Cucurbit Genetics Cooperative

Report No. 23

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Table of Contents (article titles linked to html files)

Cucurbit Genetics Cooperative Report 23: v-x (Article 0) 2000

23rd Annual Business Meeting of the Cucurbit Genetics Cooperative

Dennis T. Ray

University of Arizona, USA

The 1999 Business Meeting of the Cucurbit Genetics Cooperative (CGC) was held on 28 July 1999 in Minneapolis, Minnesota, in conjunction with the 1999 International Conference of the American Society for Horticultural Science. Tim Ng (CGC Chair) was unable to attend the meeting, and Dennis Ray (CGC Coordinating Committee member for Watermelon) presided in his place.

The meeting was called to order at 11:00 a.m., with Todd Wehner volunteering to be secretary for the minutes. After introductions around the room, Dennis passed out a handout showing the past history (size, distribution, costs) for the CGC Annual Report. He mentioned that CGC may have to increase their dues to $10 US per year soon, to cover rising costs of printing and postage**.**

Dennis reported that CGC Report No. 22 (1999) was still in preparation and due in august, and would include the watermelon gene list. The number of papers for CGC 22 was down somewhat, probably because of many papers being submitted to the recent Cucurbitaceae '98. However, while the number of reports has been dropping in recent years, the length of the reports has been increasing. As a consequence, the size of the CGC Report has remained fairly constant. However, a recurring problem is that research papers are often sent late to CGC.

A brief discussion ensued on how to increase the number of submissions to the CGC Report, and also how to decrease the number of late submissions. Some thought was given to revising the "Call for Papers" brochure to indicate the kinds of papers CGC would like to be submitted. The possibility of publishing yield trials in CGC was also discussed.

The conversation then moved to upcoming cucurbit-related meetings, in particular the 1st International Oil Pumpkin Conference (Austria, August 1999) and Cucurbitaceae 2000 (Israel, March 2000). The next CGC Business Meeting would be held in Orlando, Florida, in July 2000, again in conjunction with ASHS.

Recent books were announced, including the availability of the proceedings from Cucurbitaceae '98 (from ASHS), as well as Cucurbitaceae '96 and Cucurbitaceae '94 (both from CGC). "Cucurbits" (R.W. Robinson and D. Decker-Walters) was mentioned as in the process being reprinted after the initial print run sold out. Also, a new book on O.J. Eigsti and the seedless watermelon was described, as well as an upcoming book on vegetable breeding (author unknown, in press).

The possibility of turning the CGC Report into a web-only publication was discussed. However, there was concern that many of our members, particularly international members, may not yet have computers or Internet access. The full text of all CGC back issues is not yet available on the web, and library archives for electronic versions do not yet exist. The decision was made to stay with the printed copy of the CGC Report for a few more years.

Under new business, Dennis mentioned that Tim Ng was now ASsociate Vice President for Research at the University of Maryland, and was finding it more difficult each year to continue the CGC Chair's duties, such as preparing the CGC Annual Report, publishing and mailing it, sending renewal invoices and the "Call for papers," developing the CC website, handling correspondence, and maintaining the CGC database of members and library subscribers. The possibility of identifying an Associate Chair who could take over some of these responsibilities was discussed, with the possibility of that individual becoming Chair at some point in the future. There was also a discussion of whether ASS might take over the web and printing responsibilities for CGC, and that this should be considered.

(Note: subsequently, Tim had a conversation with Mike Neff, ASHS Executive Director, and both agreed that this would not be possible under the current CGC cost structure. Tim was unwilling to dramatically raise CGC dues, so CGC will continue with the current organization for the forseeable future.)

The meeting was adjourned at 12:00 p.m.

CGC Website Update

Timothy J. Ng

University of Maryland, USA

As many CGC members know, CGC established a website in June 1995 as a means of communicating with its members and the general public, and also to provide an electronic archive of past CGC reports and software programs (See CGC Rept. No. 19:89-90). I initially established the website on the University of Maryland server, but quickly moved it to the server for the U.S. Plant Genome Project, which was located in the USDA National Agricultural Library (NAL) in Beltsville, Maryland. For many years, we had a mutually beneficial relationship with the NAL folks.

On Sunday, 8 August 1999, I received an urgent email that the NAL site was going to be decommissioned sometime with the next month or so. Responsibility for maintaining the NAL services was being delegated to the USDA-ARS Plant Genome Database Project at Cornell University, Ithica NY. Fortunately, the folks at Cornell graciously agreed to continue hosting the CGC website, and even moved the CGC pages from NAL and updated them in the process. Because of the timing, I was even able to get the new web address into CGC Report No. 22 (1999) as it was going to press. Thus, for those of you who were wondering, this is the reason behind the change in our URl this past year.

I am grateful to the "Demeter's Genomes" staff at Cornell for the transition and web hosting, and especially to Dave Mathews, who is the USDA-ARS Plant Genome Database Curator. And of course, my fervent thanks to the many people at NAL who assisted us during the first four years of CGC's presence on the web.

Taxonomic Data for *Cucumis* and *Cucumella* on the Web

Joseph Kirkbride, USDA ARS,Beltsville, MD, has just completed a "first draft" of the taxonomic data for *cucumis* and *Cucumella* on the Internet, with the assistance of his colleagues Michael Dallwitz and David Farr. To view this, go to <*http://nt.ars-grin.gov/*> and select "Systemic Resources" from the left frame on your screen. You will then find a section on"*cucumis* and a *Cucumella* (Cucurbitaceae): Cucumbers and Melons," which links to the web page for these data.

The databases contain data on 34 species, 6 subspecies, and 2 varieties of *cucumis*, and also 11 species of *cucumella*, all the known taxa of these two genera. Joe has organized these data into two databases, one with the morphological data for identifications, and the other data with collections data so that the detailed distributions can be worked out. To use the morphological data, you must have downloaded the INTKEY software program of CSIRO, which is available on the Internet at no cost.

CGC's website may become a "mirror site" for those databases in the near future. Meanwhile, Joe is very much interested in feedback on his website so that he can continue to improve it. If you are interested in taxonomic data for these two species, please feel free to access the databases and send Joe your impressions and suggestions for improvement.

Cucurbitaceae 2000 - Short Report

Nurit Katzir & Harry S. Paris

Newe Ya'ar Research Center, Israel

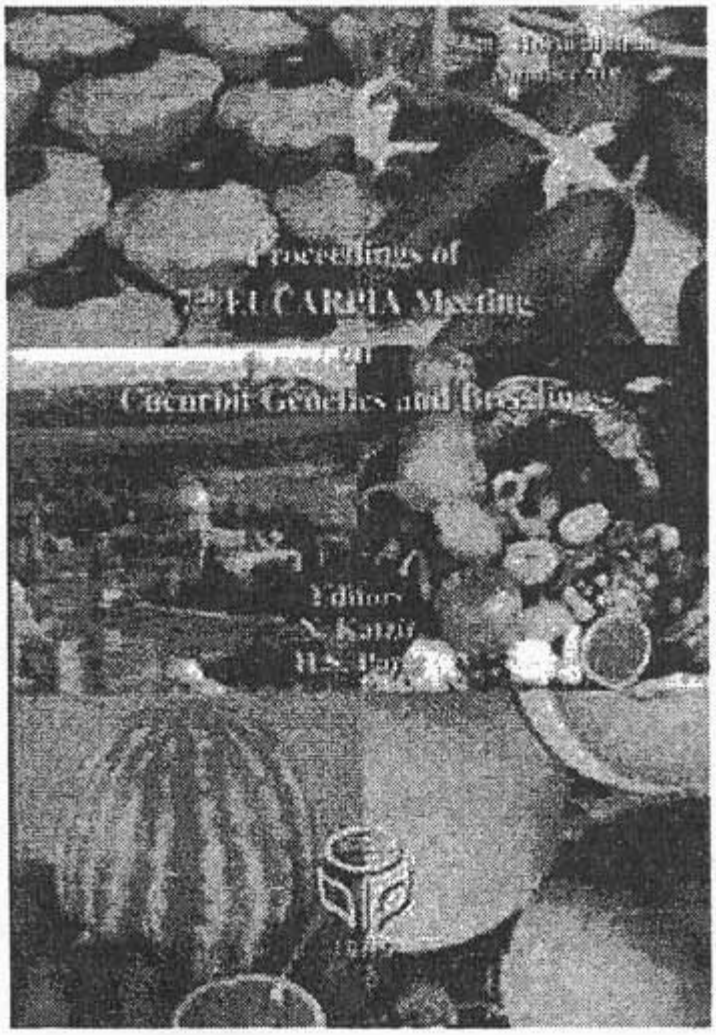
The biennial meetings on cucurbit genetics and breeding have been held over the past decade in even-numbered years and alternately on both sides of the Atlantic Ocean. This year's meeting, "Cucurbitaceae 2000," the 7th Eucarpia Meeting on cucurbit Genetics and Breeding, was held in Israel, at the Ma'ale Ha'hamisha resort near Jerusalem. Over 150 scientists from 24 countries attended, there being 77 contributed manuscripts of which 48 were presented as lectures and 29 as

posters. The meeting included tours of Israel's premiere cucurbit-growing region, an excursion to the ancient fortress of Masada, and of course to Jerusalem.

The book, "Proceedings of Cucurbitaceae 2000: the 7th Eucarpia meeting on cucurbit Genetics and Breeding: (edited by N. Katzir and H.S. Paris), was issued on the first day of the meeting. It is volume 510 (ISBN 90 6605 852 8) of the Acta Horticulture series, published by the International Society for Horticultural Science (ISHS). It contains full-length manuscripts of the 77 lectures and posters contributed at Cucurbitaceae 2000. The manuscripts are grouped within the Proceedings into subject headings: breeding and genetics, air-borne diseases, genetics and germplasm, insect pests, virology, molecular biology, and fruit quality and postharvest. This 509-page book includes some color figures and can be purchased (as supplies last) from ISHS.

The address: International Society for Horticultural Science, K. Mercierlaan 92, 3001 Leuven, Belgium. Tax: 32 16229450; Email: *orders@ishs.org;* web: *http://222.ishs.org/pub/ah510.htm/.*

At the close of Cucurbitaceae 2000 those in attendance voted the Czech Republic as the venue of Cucurbitaceae 2004, with Ales Lebeda acting as organizer.



20th Annual Meeting of the Watermelon Research and Development Working Group

20th Annual Meeting of the Watermelon Research and Development Working Group

Benny R. Bruton

USDA/ARS, Lane, Oklahoma 74555

The annual meeting of the Watermelon Research and Development Working Group (WRDWG) was held on Sunday, January 31, 2000 in Lexington, Kentucky. WRDWG met in conjunction with the southern Association of Agricultural Scientists (SAAS) and the Southern Region of the american Society for Horticultural Science (SR:ASHS). due to terrible weather conditions, particularly ice, there was a relatively low attendance with about 20 people present. Several WRDWG members got stranded in airports for the weekend and others could not even get started. In spite of the weather, there were four research reports and a highly enlightening report on watermelon germplasm from Alan Stoner.

Research Updates

Bob Maloney, Novartis Seeds, discussed problems with triploid seed germination. First, don't plant too shallow. Second, don't over-water. (If you over-water, excess water will get into the seed and you will experience germination failure.) Third, Temperature is an important factor. Some triploids tend not to germinate evenly, but after 14 days you will have all the germination you're going to get. Bob also mentioned planting a melon (OP) or wheat seed into the cell along with the watermelon seed to relieve the problem of plantlets pulling out of the planting medium during transplanting.

Don Maynard, University of Florida, mentioned that the american Society for Horticultural Science (ASHS) was sponsoring crop specific books and that one would be published on watermelon characteristics, production, and marketing.

Dan Egel, Purdue University, discussed "Sudden Wilt" of watermelon in Indiana. The disease tends to start in an area and move down the row. The disease is more severe using plastic mulch, and fumigation has not shown a beneficial response. Symptoms on roots are variable, ranging from a relatively white root system to roots having numerous lesions. To date, no fungus or bacterium has been consistently isolated. However, ground-up roots from symptomatic plants did induce some seedling disease in the greenhouse.

Benny Bruton, USDA-ARS, Lane, Oklahoma, discussed the status of "yellow Vine" of watermelon. The geographic distribution of the disease continues to increase. *Robert Wick*, University of Massachusetts, sent pumpkin samples to the Lane Research Station for PCR testing. They were positive for the yellow vine bacterium. Although the disease was not observed in watermelon, this does expand the known distribution of the disease on cucurbits to include Texas, Oklahoma, Tennessee, and Massachusetts. We suspect that the disease is more widespread than is presently known. Symptoms have often been confused with Fusarium wilt and other vine declines. the best diagnostic characteristic is a honey-brown discoloration of the phloem. You can visit the following website and go to photos and see examples of Yellow Vine on various cucurbits crops including watermelon [*http://www.lane-ag.org/scarl/scarl.htm].* If you suspect yellow vine in your state, please send us samples to run PCR.

Another topic that was discussed is a seed source for the watermelon differentials for determining race of *fusarium oxysporum* f. sp. *niveum.* Todd Wehner agreed to get the differential germplasm, test it for purity, and increase it for distribution. Germplasm can be hard to find and impossible to know the genetic purity. I hope that in the future, we can find someone to produce the differentials and offer them for sale.

Allen Stoner, USDA-ARS, Beltsville, Maryland, graciously agreed to attend our meeting and help clarify some of the questions we have had about the watermelon germplasm. he gave us an overall view of the National Germplasm system, which has about 450,000 accessions total. Dr. Stoner discussed the evolution of the Germplasm Resource Information Network (GRIN), the national Seed Storage Laboratory at Ft. Collins, and the Crop Advisory Committees. We have 40 Crop Advisory Committees at present. Dr. Stoner made a few suggestions that are worth noting here:

- We need to get duplicate samples of all the Ft. Collins, Colorado, watermelon germplasm, to Griffin, Georgia.
- We need to minimize the duplication of germplasm at Ft. Collins and at Griffin.
- Core collection may be a good idea. We need to enlist help from the Curator in making decisions and choices.

- What is the value of the PIs increased under the old system (open-pollinated)?
- Do ARS, Universities, and SEed Companies have seeds they would like to put into the system?

(Note from the Chairman: *Bob Jarret* (USDA-Griffin) has tried to get help in establishing a core collection for a long time. The Cucurbit Crop Advisory Committee and the Cucurbit Genetics Cooperative Coordinating Committee for watermelon should take the lead. Perhaps we can have input or we can take the lead and ask them for endorsement of a core collection. We need to do what we can, as a committee, to move this along if this is the direction we need to go. Please let me know what your individual thoughts are as to a Core Collection.)

News From the National Watermelon Promotion Board (NWPB)

William Watson, Executive Director of NWPB, was not able to attend due to prior commitments. However, the following NWPB information was provided:

1. A new project at the Lane Research Station, Lane, OK, will investigate the content and health properties of lycopene, a powerful antioxidant in watermelon. USDA-ARS investigator*Penelope Perkins-Veazie* will lead a team of researchers from USDA, Oklahoma State University, and Texas A&M, who will determine yield, stability, and quality of lycopene from marketable fruit and from watermelons considered culls.
2. In another new project, a team of researchers from Oklahoma State University wills et up a system to better collect and disseminate production-related research to watermelon industry members. Researchers see a need to regularly communicate and relay information to the watermelon industry about cultivar and pesticide evaluations, fertility rates, and cultural practices. The group hopes to develop a national information exchange group to establish a mechanism for distributing research results and information to all facets of the watermelon industry.
3. The NWPB Board voted to expand the work of Purdue University plant pathologist*Richard Latin*, who has developed a weather-based system designed to reduce fungicide use without increasing the risk of serious disease outbreaks. The system is called "Melon Disease Forecaster" (MELCAST). growers have relied upon MELCAST to provide temperature and moisture readings that enable them to spray at the most opportune time, thereby improving disease control while reducing fungicide costs.
4. The Board also voted to continue University of Florida research by plant pathologist *Don Hopkins*, who is investigating how to marshal a plant's natural defense system to control disease through chemicals known as plant defense activat9ors. These activators have no direct toxic effect on pathogenic fungi or bacteria and are not classified as fungicides. Early findings indicate these activators are effective in preventing the spread of bacterial fruit blotch in the greenhouse and would be effective in reducing the amount of fruit blotch in the field.

The NWPB has budgeted $50,000 annually through 2001 to support research that addresses the following five research priority areas: (1) postharvest physiology/quality, (2) gummy stem blight, including host resistance, epidemiology, and control, (3) standardization of variety evaluations and data accumulation, (4) removal and disposal of plastic mulch, and (5) disease forecast systems.

III. New Business

It was decided at the 1999 Memphis Meeting that we should invite all interested people (national and international) to become involved with out group. The new WRDWG web page is up and operational. The address is: *http://www.lane-ag.org/H2oMelon/watermelon.htm.* We have a search engine so that a person can find an expert in watermelon culture, fertility, plastic mulch, postharvest problems, foliar diseases, or soilborne diseases, etc. This information should provide a useful service to research and Extension personnel to find needed information. We do not intend to try to duplicate information that is covered on other web pages. Please pull up the forms, fill them out and send to us, since our 'expertise" section is very small and inadequate at the present time to be of much help. Hopefully, you will find our web page helpful.

Upcoming Meeting

The 21st Annual Watermelon Research and Development Working Group meting will be held from 1:00 to 5:00 p.m. on Sunday, 28 January, 2001, in Ft. Worth, Texas.

Comments.....

From the CGC Coordinating Committee: The Call for Papers for the 2001 Report (CGC Report No. 24) will be mailed in October 2000. Papers should be submitted to the respective Coordinating Committee members by 28 February 2001, although late submissions may be considered if received prior to our processing deadline. The Report will be published by July 2001. As always, wearer eager to hear from CGC members regarding our current activities and future direction of CGC.

From the CGC Gene List Committee: Lists of known genes for the *Cucurbitaceae* have been published previously in HortScience and in reports of the Cucurbit Genetics Cooperative. CGC is currently publishing complete lists of known genes for cucumber (*Cucumis sativus*), melon (*Cucumis melo*), watermelon (*Citrullus lanatus*), and *Cucurbita* spp. on a rotating basis.

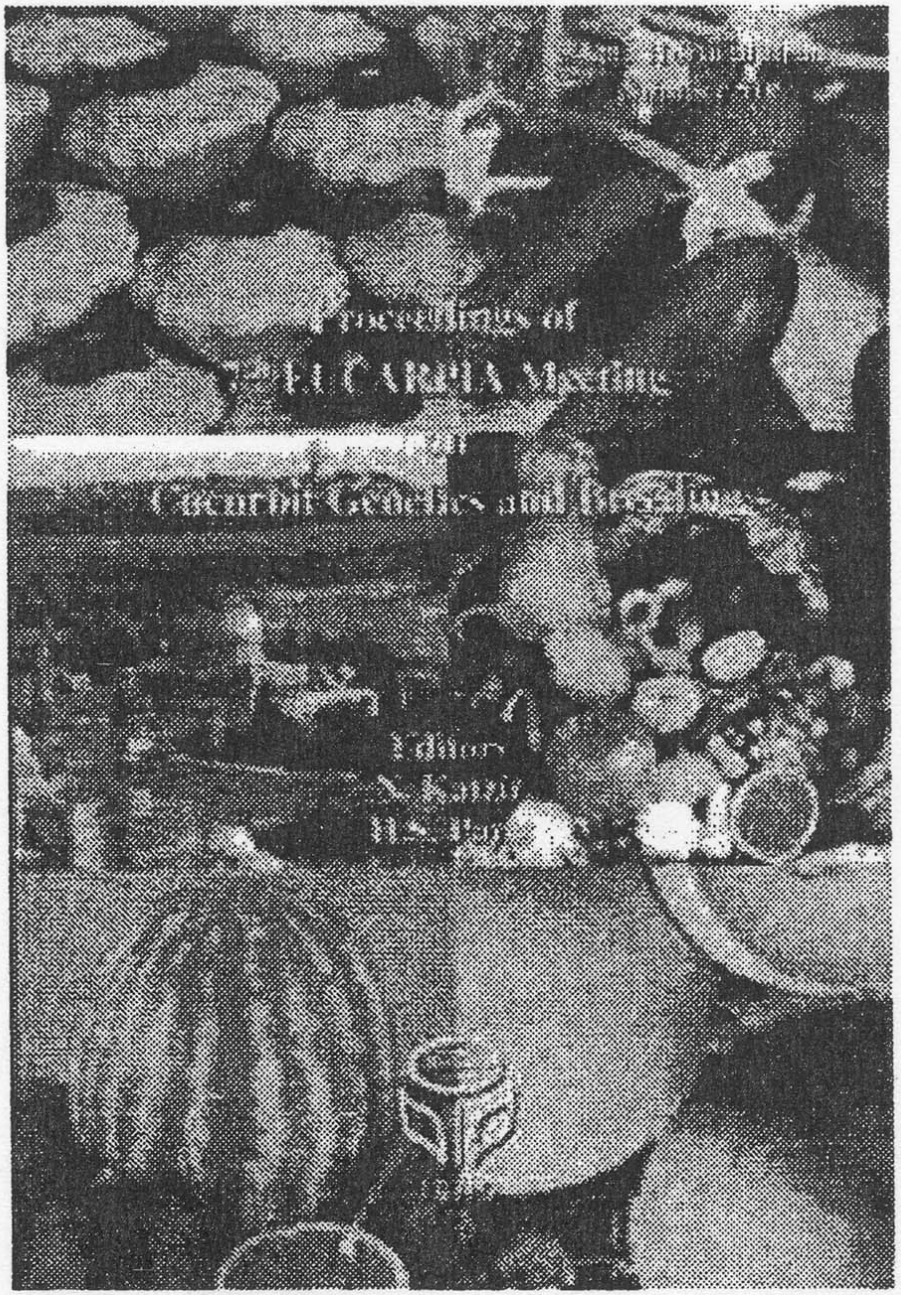
It is hoped that scientists will consult these lists as well as the rules of gene nomenclature for the Cucurbitaceae before selecting a gene name and symbol. Thus, inadvertent duplication of gene names and symbols will be prevented. The rules of gene nomenclature (published in each CGC Report) were adopted in order to provide guidelines for the naming and symbolizing of genes previously reported and those which will be reported in the future. Scientists are urged to contact members of the Gene List Committee regarding questions in interpreting the nomenclature rules and in naming and symbolizing new genes.

From the CGC Gene Curators: CGC has appointed curators for the four major cultivated crops: cucumber, melon, watermelon and *Cucurbita* spp. Curators are responsible for collecting, maintaining, and distributing upon request stocks of known marker genes. CGC members are requested to forward samples of currently held gene stocks to the respective Curator.

Upcoming Meetings of Interest to Cucurbit Researchers

Organization/Meeting	Date(s)	Location	Contact
Cucurbit Genetics Cooperative	11:00 a.m., 23 July 2000	Coronado Springs Hotel, Orlando, Florida	Tim Ng 301-405-4345 *tn5@umail.umd.edu*
	July 2001	Sacramento, California	
	August 2002	Toronto, Canada	
	September 2003	Providence, Rhode Island	
Cucurbitaceae 2002	2002	Florida, USA	Don Maynard 941-751-7636 *bra@gnv.ifas.ufl.edu*
2nd ISHS International Symposium on Cucurbitaceae	May 2001	Japan	*To be announced.* Check: *http://www.ishs.org*
Eucarpia Cucurbitaceae 2004	2004	Czech Republic	Ales Lebeda 420/68/5223325 *lebeda@rise.upol.cz*
Watermelon Research & Development Working Group	January 2001	Fort Worth, Texas	Benny Bruton 580-889-7395 *bbruton-usda@lane-ag.org*
	2002	Orlando, Florida	
	2003	Mobile, Alabama	
Cucurbit Crop Germplasm Committee	6:00 p.m., 24 July 2000	Coronado Springs Hotel, Orlando, Florida	Jim McCreight 831-755-2864 *mccreight@pwa.ars.usda.gov*
Pickling Cucumber Improvement Cooperative	24-25 October 2001	Hyatt Regency Crown Center Kansas City, Missouri	John O'Sullivan 519-426-7127 *josulliv@uoguelph.ca*

Pickle Packers International	Fall Meeting: 25-26 October 2000 Fall Meeting: 24-25 October 2001 Spring Meeting: 2001 Spring Meeting: 2002	Hyatt Regency Crown Center Kansas City, Missouri	Richard Hentschel 630-584-8950 *staff@ppi.i.org*



20th Annual Meeting of the Watermelon Research and Development Working Group

Cucurbit Genetics Cooperative Report 23 1-3 (article1) 2000

A New Source of Resistance to *Meloidogyne incognita* (Kofoid & White) Chitwood Identified in *Cucumis*

Jin-Feng Chen[1] and Stephen Lewis[2]

[1]Departmentof Horticulture, Nanjing Agricultural University, Nanjing 210095, China; [2] Department of Plant Pathology and Physiology, Clemson University, Clemson, SC 29634

Introduction: Cucumber (*Cucumis sativus* L.) is considered very susceptible to major pathogens active in temperate growing regions (Fassuliotis, 1979). In this regard, susceptibility to nematodes can be a serious limiting factor in commercial production of cucumber. Several cucumber-wide screenings have been conducted to identify sources of resistance to nematodes (*Meloidogyne ssp.*). These tests evaluated hundreds of cucumber cultigens (cultivars, breeding lines, nd plant introduction accessions), but no source of resistance to *M. incognita* was identified (Winstead and Sasser, 1956; Fassuliotis and Rau, 1963; Walters et al., 1993).

Some related *Cucumis* species, such as *C. metuliferus* E. Meyer ex Naudin, *C. anguria,* L., *C. ficifolius* A. Rich., and *C. longipes* Hook., have been found to possess high resistance to various root-knot nematode species include in*M. incognita* (Fassuliotis, 1967; Norton and Granberry, 1980). But fortunately all attempts to hybridize these wild relatives with commercial cucumber have failed. (Whitaker, 1930; Natra, 1953; Smith and Venkat Ram, 1954; Deakin et al., 1971; Fassuliotis and Nelson, 1988).

A successful cross has been made and confirmed between cucumber and *C. hystrix* Chakr. (2n = 24) (Chen et al., 1997) This interspecific hybridization is the first repeatable cross between a cultivated *cucumis* species and a wild relative. Since there is cross-compatibility between *C. sativus* and *C. hystrix*, the economically important characters if *C. hystrix* are of great interest to cucurbit scientists and breeders. In this paper, we introduce the resistance to root-knot nematode (*M. incognita*) in *C. hystrix and* the transmission of resistance from *C. hystrix to* its interspecific F_1 hybrid with cucumber.

Materials and methods:*Plant materials. C. hysteria* Xishuangbanna No. ! and No. 2 cucumber (*Cucumis sativus* L. var. *xishuangbannesis* Qi et Yuan) used in this study were from the original collection of J.F. Chen (Chen et al., 1994) the Northern Chinese cucumber cultivar 'Beijing jietou' (V05A464) was obtained from Dr. C.Z. Qi, of the Vegetable Research Institute Chinese Agricultural Academy of Sciences, Beijing. The Southern Chinese cucumber 'Erzhaozi' was obtained from Mr. Z.B. Gong, of the Chengdu Seed Company, Chengdu, Sichuan Province.

Matings. Reciprocal interspecific hybridization between *C. hystrix and* cucumber, and subsequent embryo rescue were performed as described previously (Chen et al., 1997). The diploid sterile F_1 progeny (2n = 19) went through chromosome doubling as previously described (Chen et al., 1998). The BCF_1 was made by crossing the chromosome-doubled F_1 (with genome HHCC, where H represents the genome of *C. hystrix, C* the *C. sativus*) to the original diploid cucumber stock parent.

Inoculation. Nematode and inoculation M*eloidogyne incognita* race 3 was cultured on greenhouse-grown tomato, *Lycopersicon esculentum* Mill. cv. Rutgers. Nematode inoculum was obtained by collecting eggs with 0.5% NaOCl as described by Hussey and Barker (1973).

Experimental design, plant culture and data collection. Seeds were germinated in vermiculite in a greenhouse, and seedlings at the two-leaf stage were transplanted into 4-inch pots filled with pure sand. Plantlets from in vitro culture were also transplanted into the same media at the same time. Plants were fertilized weekly with a commercial nutrient formulation (N: P: K = 20: 20: 20), and kept in a greenhouse at 28 ° C. Four days after planting, two holes 2-3 cm in depth and 00.6 cm in diam. were made with a bamboo stock around the plant roots. One ml of inoculum containing 2,500 eggs was pipetted into

each hole (5,000 eggs for each plant). There were five replications of each cultigen to determine the ability of the nematode reproduce. Plants were placed on a table in a completely randomized design. Seven weeks after inoculation, the root systems were carefully washed free of sand, and evaluated for number of galls. The number of galls for each root system was counted, and a gall index was calculated using a 0 - 5 scale, with 0 = no galls, 1 = 1 to 2, 2 = 3 to 10, 3 = 11 to 30, 4 = 31 to 100, and 5>100 galls. Normalized data were subjected to analysis of variance using SAS (SAS Inst., Cary, NC), and the means of gall indices were tested using Tukey's Studentized Range (HSD) Test.

Results and Discussion: To identify the resistance in *cucumis hystrix* Chakr. and evaluate the transmission of resistance to the progenies of its interspecific hybrid with cucumber, a screening was conducted in a greenhouse using C. hystrix, four cucumber cultigens, and three reciprocal interspecific hybrids at two ploidy levels, and one BCF_1 progeny.

After 45 days, there were, on average, only about three galls that could be seen in each *C. hystrix root* system,m. In contrast, over 100 galls could be counted in each cucumber root system tested.

C. hystrix had a high level of resistance to *M. incognita with* mean gall index of 1.8, while cucumbers were confirmed as being highly susceptible with a mean gall index of 4.8-5.0. The resistance was partially transferred to the interspecific hybrid. The mean gall index changed from 4.8 to 3.4, which is about mid-point between the resistant parent *C. hystrix* (1.8) and the susceptible parent cucumbers (4.8 - 5.0). This transmission was also observed when the chromosome=doubled F_1 and BCF_1, the statistical analysis indicated no significance between them.

There is only limited variability in mean gall index among plans of hybrids, or the parents. No significant difference in mean gall index was observed between the reciprocal F_1 plants, which indicates that the expression of resistance in the progeny is not influenced by the maternal parent. Meanwhile, the mean gall index of the F_1 plant was intermediate between both parents, indicating that neither resistance nor susceptibility is dominant.

In summary, the results revealed a high level of resistance (~ three galls in each root system) in *C. hystrix*, while cucumber was highly susceptible. The resistance was partially transmitted to the F_1 when the reciprocal interspecific hybrid was made. This resistance was further transmitted to the BCF_1 progeny when the $F_1$was backcrossed to the cucumber.

The benefit-to-cost ratio for the development of resistant crop cultivars in the United States was estimated at $300 for every $1 spent (Bottrel, 1979). Plant resistance was identified as the highest research priority in management of plant-parasitic nematodes (Bird, 1980). This success of interspecific hybridization between cucumber and its wild relatives (Chen et al., 1997) is of great importance to cucumber genetics and breeding. Introgression of root-knot nematode resistance from *C. hystrix to* cucumber must be advanced by further backcrossing. Strategies need to be developed to facilitate the gene transmission,such as more knowledge of cytogenetics and in combination with marker-assisted selection.

Literature Cited

1. Batra, S. 1953. Interspecific hybridization in the genus *Cucumis.* Sci. Culture 18:445-446.
2. Bird, G.W. 1980. Nematology status and prospects: The Role of nematology in integrated pest management. J. Nematology 12:170-176.
3. Bottrel, D.R. 1979. Integrated pest management. Washington, D.C. : United States Government Printing Office.
4. Chen, J.F., J.W. Adelberg, J.E. Staub, H..T. Knap,and B.B. Rhodes. 1998. A new synthetic amphidiploid in *Cucumis* from a *C. sativus C. hystrix* F_1 interspecific hybrid, p. 336-339. In: James McCreight (eds.), Cucurbitaceae '98 - Evaluation and enhancement of Cucurbit germplasm, ASHS Press, Alexandria, Va.
5. Chen, J.F., J.E. Staub, Y. Tashiro, S. Isshiki, and S. Miyazaki. 1997. Successful interspecific hybridization between *cucumis sativus* L. and *C. hystrix* Chakr. Euphytica 96:413-419.
6. Chen, J.F., S.L. Zhang, and X.G. Zhang. 1994. The xishuangbanna gourd (*C. sativus* var. *xishuangbannesis* Qi et Yuan), a traditionally cultivated plant of the Hanai people, Xishuangbanna, Yunnan, China. Cucurbit Genet. Coop. Rpt. 17:18-20.
7. Deakin, J.R., G.W. Bohn and T.W. Whitaker. 1971. Interspecific hybridization in *Cucumis*, Econ. Bot. 25: 195-211.
8. Fassuliotis, G. 1967. Species of *Cucumis* resistant to the root-knot nematode, *Meloidogyne incognita acrita.* Plant Dis. Rpt. 51: 720-723.
9. Fassuliotis, G. 1979. Plant breeding for root-knot nematode resistance, p. 425-453. In: J.N. Sasser and C.C. Carter (eds.). Root-knot nematodes (*Meloidogyne species*): Systematics, biology and control. Academic, New York.
10. Fassuliotis, G. and B.V. Nelson. 1988. Interspecific hybrids of *cucumis metuliferus* x *C. anguria* obtained through embryo culture and embryogenesis. Euphytica 37: 53-60.

11. Fassuliotis, G. and G.J. Rau. 1963. Evaluation of *Cucumis spp.*for resistance to the cotton root-knot nematode, *Meloidogyne incognita acrita.* Plant Dis. Rpt. 47: 809.
12. Hussey, R.S.and K.R. Barker. 1973. A comparison of methods of collecting inocula of *Meloidogyne spp.* including a new technique. Plant Dis. Rpt. 57: 1025-1028.
13. Norton, J.D. and D.M. Granberry. 1980. Characteristics of progeny from an interspecific cross of *cucumis melo* with *C. metuliferus.* J. Amer. Soc. Hort. Sci. 105: 174-180.
14. Smith, P.G. and B.R. Venkat Ram. 1954. Interspecific hybridization between muskmelon and cucumber. J. Hered. 45-24.
15. Walters, S.A., T.C. Wehner, and K.R. Barker. 1993. Root-knot nematode resistance in cucumber and horned cucumber. HortScience 28: 151-154.
16. Whitaker, T.W. 1930. Chromosome number in cultivated cucurbits. Am. J. Bot. 17:1033-1040.
17. Winstead, M.N. and J.N. Sasser. 1956. Reaction of cucumber varieties to five root-knot nematodes (*Meloidogyne spp.*). Plant Dis. Rpt. 40: 272-27.

Cucurbit Genetics Cooperative Report 23:4-7 (article 2) 2000

Method for the Development and characterization of Microsatellite Markers in Cucumber

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Introduction. Genetic markers such as isozymes, RAPDs and RFLPs have ben characterized in cucumber (*Cucumis sativus* L.) (Dijkhuizen et al., 1996; Serquen et al., 1997). However, critical documentation of these markers and their usefulness in marker-assisted selection (MAS) for applied breeding programs has been limited. This is at least partially due to the species' low polymorphism level. A higher level of polymorphism has been associated with SSR loci in preliminary studies with *C. sativus* well as in other genera (*Cucurbita and Citrullus*) of the Cucurbitaceae (Katzir et al., 1996). In this study 12 of 20 SSRs (60%) possessed two to give alleles per locus.

A moderately large molecular marker database now exists for cucumber (Meglic et al., 1996a and 1996b; Dijkhuizen et a;., 1996). The number of polymorphic loci identified in these crops is useful for variety identification and seed purity testing, but is not robust enough to be used for legal applications or some phases of germplasm management (Staub et al., 1999). The identification and characterization of SSR markers would allow for more precision in the estimation of genetic similarities among cucumber cultivars, and thus provide cucumber researchers with additional markers for genetic analysis (Staub et al., 1996). Since their initial implementation, PCR markers based on microsatellites have become the marker of choice for many genetic. Their codominant nature, and high polymorphism rate, and high through-put capacity are important methodological characteristics. The initial development of microsatellite markers requires the characterization of sequences flanking the repeat motif, followed by the design of flanking PCR primers. Many methods have been devised for the initial characterization of the flanking sequences (Litt and Luty, 1989). Other methods include: database search, construction of a genomic or cDNA library and screening with oligonucleotide probes, and construction of microsatellite-enriched libraries followed by PCR screening. We describe herein a novel method to capture sequences that contain microsatellite motifs.

Materials and Methods:DNA extraction, restriction and size fractionation. DNA from cucumber breeding line G421 was extracted from young leaves and apical meristems according to the nuclear DNA extraction protocol outlined in Maniatis (1982). Extraction products were treated with RNAse ONE (Promega, Madison, WI). DNA was quantified using a TKO 100 fluorometer (Hoefer). In separate reactions 100 mg of DNA were restricted using EcoR I restriction enzyme (Promega, Madison WI) and Acs I (Boehringer Mannheim) which recognizes the sequence (A or G)/AATT(T or C) and generates compatible ends with EcoR I. Restricted DNA was size fractionated using a low pressure 75cm long 16mm diameter chromatography column (Fisher) S-500 (Pharmacia). The column was packed according to instructions for Sephacryl S-500 and equilibrated with 200 ml elution buffer at 0.3 ml/min (0.1 M tris-HCL pH 8.0, 0.15 M NaCl, 0.001 M ETDA). Both samples of restricted DNA were fractionated separately and eluted DNA was collected at 2 ml aliquots. Eluted DNA wa precipitated, washed and resuspended in TE. Elution aliquots were sized by agarose gel electrophoresis. Fractions between 200 bp to 1200 bp were combined for ligation to vector.

Library construction, mass excision and plasmid DNA extraction. Fractionated DNA was ligated to Ziplox EcoR I arms (Life Technologies, Gaithersburg, MD) and packaged in a lambda vector with Gigapack III Gold packaging extract (Stratagene, La Jolla, CA). The resulting library was titered and efficiency of packaging was determined by blue/white colony screening. Primary Ziplox libraries were max excised in vivo into the pZL1 plasmid vector (Life Technologies, Gaithersburg, MD) in DG10B Zip (Life Technologies, Gaithersburg, MD) strain of *E. coli*. The mass excision and expansion of the plasmid library were performed at the same time in a semisolid media to minimize representational biases that can occur during expansion in liquid media. Plasmid DNA was extracted b7 a plasmid maxi-prep procedure (Quiagen, Valencia, CA).

Capture of plasmids containing microsatellites. In preparation for the GeneTrapper^{TM} (Life Technologies, Gaithersburg, MD) reaction, several oligonucleotides (20-30 bp) coded for common microsatellite sequences (CT, TG, CTT, TCC, ATT) were biotinylated (biotin-14-dCTP) with a terminal transferase enzyme (Life Technologies, Gaithersburg, MD). The "capture"

steps are described in detail in the GeneTrapper kit (Life Technologies, Gaithersburg, MD) and graphically represented in Figure 1.

1. The plasmid DNA was treated with Gene II protein and Exonuclease III to obtain single stranded circular plasmid DNA.
2. Single stranded DNA was hybridized with the biotinylated oligonucleotides. During the hybridization, the single stranded oligonucleotides hydrogen bonded with their complementary microsatellite sequences.
3. Para-magnetic beads bound to streptavidin were used to capture the microsatellite containing single stranded plasmid molecules. The product of the capture was washed several times to eliminate unbound single stranded DNA.
4. The final product of the capture was then treated with a buffer to release the captured single stranded DNA. The single stranded DNA was subsequently primed and repaired to form double-strand DNA.
5. The double-stranded plasmids were then used to transform DH10B E. coli that had been grown on ampicillin selective media. It was assumed that only E. coli harboring the intact pZL1 (putatively containing a microsatellite region) coming from the cucumber DNA library survived the selection media.
6. Single colonies were picked, named sequentially, and then amplified prior to plasmid DNA extraction. Confirmation of the presence of a microsatellite region was obtained by subjecting plasmid DNA to PCR using the standard forward or reverse M13 and a primer coding for the captured microsatellite type.
7. Plasmid DNA from positive clones was sequenced at the University of Wisconsin Biotechnology Center to identify the microsatellite type and length.

Sequence analysis and primer design. All sequences from positive clones were analyzed and compared to each other using the assembly feature in the GeneTool software package (Biotools, Edmonton, Canada) in order to discard duplicates and identify overlapping DNA regions. The resulting unique sequences were entered into the primer design software OLIGO 6.0 (Life Science Software, Long Lake, MN).

Results and discussion: The ease of application of the Gene Trapper^{TM} System (Life Technologies) made it possible to isolate and characterize several cucumber microsatellite loci in a relatively short time without the use of radiolabeled products. These results were obtained in 1998 independently of a similar technique developed by Paetkau (1999).

This methodology has been repeated five times with success and reproducibility of several types of microsatellites (di-, tri-, and tetra-nucleotides). Nevertheless, this methodology continues to be further refined and is still considered to be in its developmental stages. The first capture was performed with a (CT) 12 oligonucleotide, and the procedure yielded 457 clones. The PCR assays identified the size of the insert and its relative position. Based on these assays, 300 of the 457 clones were selected for the primer design step of the SSR construction. The insert sequences were entered into the Oligo 6.0 primer design tool (Molecular Biology Insights Inc., Long Lake, MN).

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This methodology has been repeated five time with success and reproducibility of several types of microsatellites (di-, tri-, and tetra-nucleotides). Nevertheless, this methodology continues to be further refined and is sill considered to be in its developmental stages. the first capture was performed with a (CT) 12 oligonucleotide, and the procedure yielded 457 clones. The PCR assays identified the size of the insert and its relative position. Based on these assays, 300 of the 457 clones were discarded as suspected duplicates. Sixty-five clones were selected for the primer design step of the SSR construction. the insert sequences were entered into the Oligo 6.0 primer design tool (Molecular Biology Insights Inc., Long Lake, MN).

Other captures using (TG)12, (CTT)8, (TCC)8, (ACTC)6, (ATT)9 have been successfully performed, and are currently being characterized as with the CT repeat capture. All primer pairs that have been designed are being tested on C. sativus lines G421 and H-19 using a thermal gradient PCR programmed on an Eppendorf Mastercycler Gradient thermal cycler (Hamburg, Germany) to establish the optimum annealing temperature range for each SSR primer.

This information will be used later in the development of PCR procedures for multiplexing SSR markers/ The SSR markers being developed in cucumber are being tested on C. *melo* parent lines MMR-1 and 'Topmark' to establish the extent of their cross-compatibility and potential use in melon. To date, approximately 80% of the SSR markers that have been constructed have amplified in both C. sativus and *C. melo*. the development of SSR markers will be useful for mapping, and introgressing economically important traits into cultivated varieties.

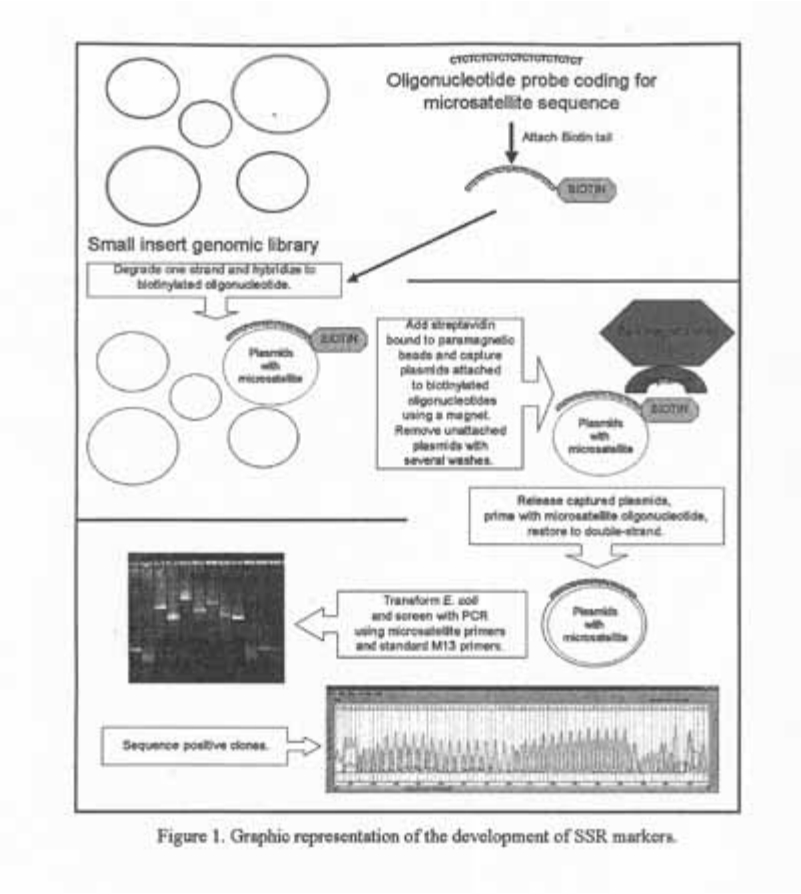

Figure 1. Graphic representation of the development of SSR markers.

Literature Cited

1. Dijkhuizen, A., W.C. Kennard, M.J. Havey, and J.E. Staub. 1996. RFLP variation and genetic relationships in cultivated cucumber. Euphytica 90: 79-87.
2. Katzir N., Danin-poleg Y., Tzuri G., Karchi Z., Lavi U., and Cregan P.B. 1996. Length polymorphism and homologies of microsatellites in several cucurbitaceae species. Theor. Appl. Genet. 93: 1282-1290.
3. Litt, M. and J.A. Lutty. 1989. A hypervariable microsatellite revealed by in vitro amplification of a dinucleotide repeat within the cardiac muscle acting gene. Am. J. Hum. Genet. 44:397-401.
4. Maniatis, T., E.F. Fritsch, and J. Sambrook. 1982. Molecular Cloning: A Laboratory Manual. cold Spring Harbor publisher, Cold Spring Harbor, NY.
5. Meglic, V., and J.E. Staub. 1996a. Genetic diversity in cucumber (*Cucumis sativus* L) II. An evaluation of selected cultivars released between 1846 and 1978. Genet. Res. Crop. Evol. 43:547-558.
6. Meglic, V., and J.E. Staub. 1996b. Inheritance and linkage relationships of isozyme and morphological loci in cucumber (*Cucumis sativus* L). Theor. Appl. Genet. 92: 865=872.
7. Paetkau, D. 1999. Microsatellites obtained using strand extension: an enrichment protocol. Biotechniques 26:690-697.
8. Serquen, F.C., J. Bacher and J.E. Staub. 1997. Mapping and QTL analysis of horticultural traits in a narrow cross in cucumber (*Cucumis sativus* L.) using random-amplified polymorphic DNA markers. Mol. Breed. 3:257-268.
9. Staub, J.E., F. Serquen, and M. Gupta. 1996. Genetic markers, map construction, and their application in plant breeding. HortScience 31:729-741.
10. Staub, J.E. 1999. Intellectual property rights, genetic markers, and hybrid seed production. J.New Seeds 1:39-65.

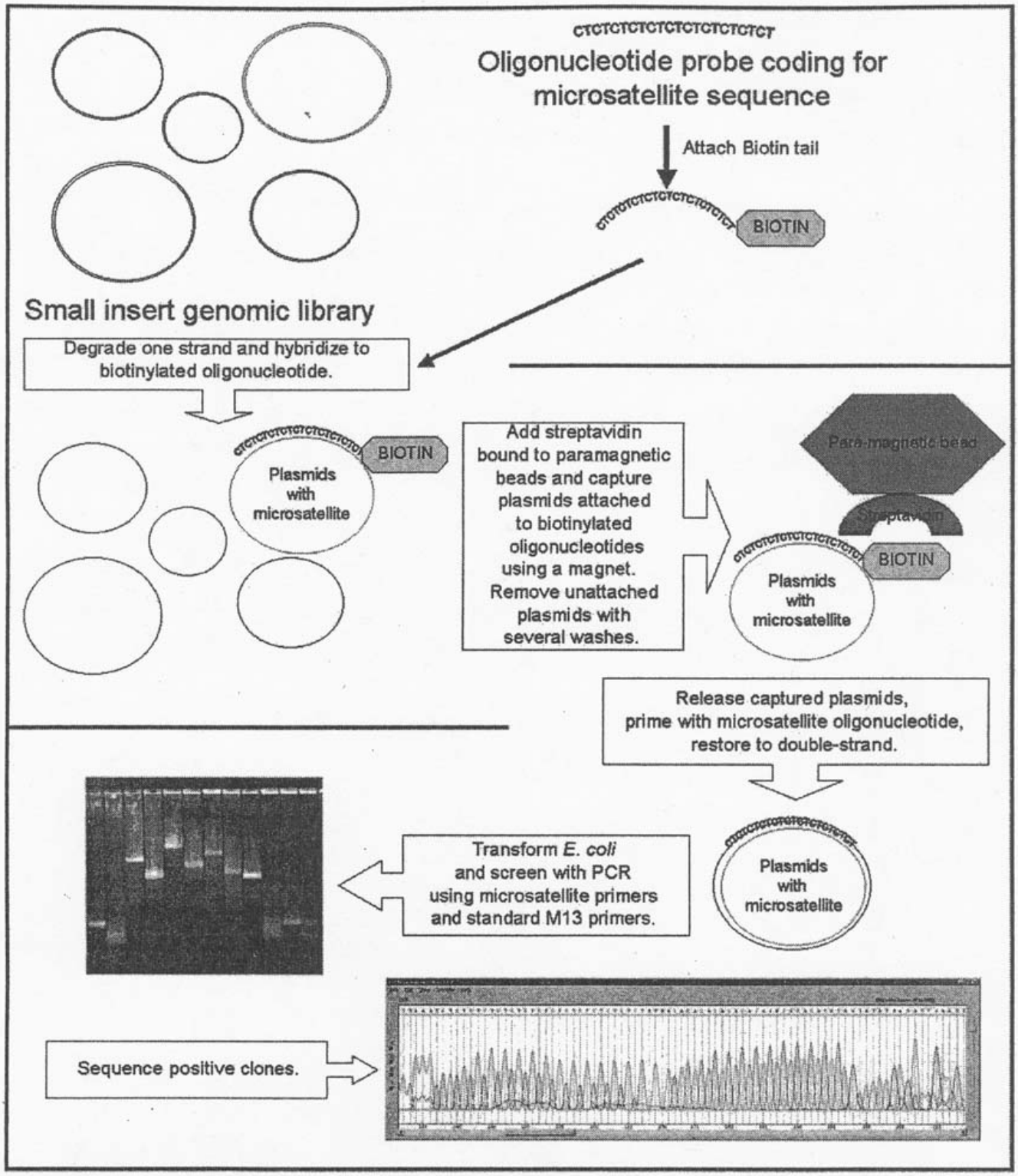

Figure 1. Graphic representation of the development of SSR markers.

Cucurbit Genetics Cooperative Report 23:8-11 (article 3) 2000

Response to Phenotypic Selection for Multiple Lateral branching in Cucumber (*Cucumis sativus* L.)

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Introduction. The acreage planted for mechanically-harvested cucumbers in the United States has increased 30 to 40% in the last 20 years. Because of rising labor costs and contract disputes this trend is projected to continue (Merchandising Guide, 1988). In the northern United States, acreage dedicated to once-over mechanical harvest is significant. For example, in Michigan, Wisconsin, Delaware, New York, Washington and Oregon mechanically harvested acreage ranges between 30 to 45%.

Curiously, the yield of pickling cucumber (gynoecious or G, and monoecious or M) has plateaued in the last 15 years. Studies by Widders and Price (1989) and Staub et al. (1989 and 1992) suggest that this recent plateau may be associated with net photosynthetic capacity. Resource limitations may explain why fruit developing from the first pollinated flower on each lateral branch inhibits the development of subsequent fruits (Denna, 1973; Fuller and Leopold, 1977).

To overcome the yield plateau and to respond to the need for cultivars suitable for mechanical harvesting, our breeding program is manipulating cucumber plant architecture to develop high yielding genotypes. Standard cucumber varieties (G x M or G x G hybrids) posses an indeterminate (*De*) plant habit and few lateral branches (`1 to 2). We are developing all female genotypes which are short in stature (determinate; *de*) and possess a multiple lateral branching habit (`5 to7 branches). This plant type can be sown at relatively high densities (compared to standard indeterminate types), and allows for an increase in early, concentrated yield (Staub et. al., 1992).

Information on quantitatively inherited traits related to yield components, however, is spare. Serquen et al. (1997) suggest that few genes (perhaps 5 to 8) control days to anthesis, sex expression, mainstem length, numbers of multiple lateral branches, and fruit number and weight. We are particularly interested in multiple lateral branching since this trait is highly correlated to yield response (). We present herein response to phenotypic selection in populations segregating for multiple lateral branches in the cross between line G421 (G, *de*, x H-19 (M, *De*). This experiment is part of a larger experiment that aims to compare the efficiency of marker-assisted selection of MLB to phenotypic selection in this cross.

Materials and Methods: The gynoecious determinate cucumber inbred line G421 possessing normal-sized leaves and low lateral branch number (approximately 1) was crossed with the monoecious indeterminate little leaf inbred line H-10 possessing high lateral branching (approximately 8) in the winter of 1997 to produce F_1 progeny. In the spring of 1998, 15 F_1 plants were used as males (after sex expression change) to pollinate 200 G421 plants to generate BC_1 seed (greenhouse in Arlington, Wisconsin).

To change sex expression, apical meristems of selected plants were treated3 times in 5 day intervals with 2-3 ml aerosol solution of 6 mM silver thiosulfate. Selected plants were then selfed and backcrossed to field-grown G421 plants (recurrent parent).

In the summer of 1999, seed of selected BC_2 (3233) and BC_1S_1 (1,367) from summer 1998 as well as parents, F_1 hybrids (36), F_2 (400), and BC_1 (397) progeny seed lots were planted at Hancock, Wisconsin (Table 1). Parents, F_1 , F_2, BC_1, $BC_1S_1$and BC_2 families were arranged in a randomized complete block design with three replications of 12 plants each (total of 36 plants per family). Seed was planted 0.45 cm apart on rows positioned 1.5 m apart. Data collection, sex conversion, and selection were performed similar to summer 1998. Selections we backcrossed to G421.

Expected gain from selection was calculated according to Falconer and Mackay (1996) as $R=1/2ih^2$ o $_P$ .The intensity of selection, *i* was adjusted to 1.2 of its tabulated value to account for the fact that no selection was applied to the females (G421). The mean h^2 (0.45) used for calculation was taken from a previously published study that estimated heritabilities for this trait in two locations (Wisconsin and Georgia) (Serquen et al., 1997).

Results and discussion: The mean MLB number of the 126 selected plants in the BC_1 population was selected, the expected gain from selection was calculated to be 0.57 units. The realized gain from selection based on the difference of the generation means was approximately 0.4 units. the replications of the base BC_1 population across years were similar. Means, standard deviations and population numbers are given in Table 1. The distributions of multiple lateral branches differ in BC_1 (MLB mean - 2.2), BC_2 (MLB mean = 2.6), and BC_2S_1(MLB ,mean 3.4) populations (Figure 1). The progressive changes (increase in MLB) in these generations are the results of selection and increased homozygosity (BC_1S_1 ; fixation) of loci affecting this trait.

A subset of approximately 200 BC_2 plants (MLB mean = 516) of the BC_2 population were selected to produce BC_3 families. As a result of this selection experiment we now possess approximately 150 BC_3 families representing independent recombination events. These families will be used to generate either nearly isogenic lines (Bernacchi et al., 1998) or congenic lines (Hill, 1998) for detailed QTL mapping and to increase our understanding the genetic control of MLB in cucumber.

Table 1. Generation means for multiple lateral branching in cucumber.

Generation	Year	Mean	SD	N
BC_1	1998	2.23	1.20	2985
G421	1999	1.75	0.84	150
H19	1999	7.90	1.60	36
F_1	1999	4.61	0.98	36
BC_1	1999	2.21	1.14	397

BC_2	1999	2.60	1.21	3233
BC_2S_1	1999	3.38	1.616	1367
F_2	1999	3.58	1.38	400

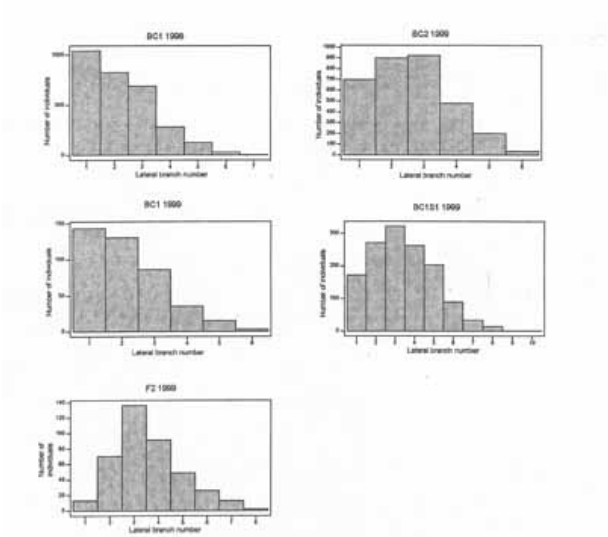

Figure 1. Distributions of multiple lateral branching among F2 and backcross cucumber populations.

Literature Cited

1. Bernacchi, D., T. Beck-Bunn, D. Emmartty, Y. Eshed, S. Inai, J. Lopez, V. Petiard, H. Sayama, J. Uhlig, D. Zamir, and S.D. Tanksley. 1988. Advanced backcross QTL analysis of tomato. II. Evaluation of near-isogenic lines carrying single-donor introgressions for desirable wild QTL-alleles derived from *Lycopersicon hirsutum* and *L. piminellifolium.* Theor. Appl. Genet. 97:170-180.
2. Denna, D.W. 1973. Effects of genetic parthenocarpy and gynoecious flowering habit on fruit production and growth in cucumber, *Cucumis sativus* L. J. Amer. Soc. Hort. Sci. 98:602-604.
3. Falconer, D.S. and F.C. Mackay. 1996. Introduction to quantitative genetics. Harlow Essex, England, Longman LTD.
4. Fuller, G.L. and C.A. Leopold. 1977. The role of nucleic acid synthesis in cucumber fruit set. J. amer. Soc. Hort. Sci. 102:384-388.
5. Hill, W.G. 1998. Selection with recurrent backcrossing to develop congenic lines for quantitative trait loci analysis. Genetics 149:1341-1352.
6. Kuper, R.S. and J.E. Staub. 1988. Combining ability between lines of *cucumis sativus* L. and *Cucumis sativus* var. *hardwickii* (R.) Alef. Euphytica 38:197-210.
7. Merchandising Guide. 1988. Pickle Packers International, Inc. St. Charles, IL.
8. Serquen, F.C., J. Bacher, and J.E. Staub. 1997. Genetic analysis of yield components in cucumber at low plant density. J. Amer. Soc. Hort. Sci. 122:522-528.
9. Staub, J.E. 1989. source-sink relationships in cucumber. Cucurbit Gen. Coop. Rpt. 12:11-14.
10. Staub, J.E., L.D. Knerr and H.J. Hopen. 1992. Effects of plant density and herbicides on cucumber productivity. J. Amer. Soc. Hort. Sci. 117:48-53.
11. Widders, I.E. and H.C. Price. 1989. Effects of plant density on growth and biomass partitioning in pickling cucumbers. J. Amer. Soc. HOrt. Sci. 11:751-755.

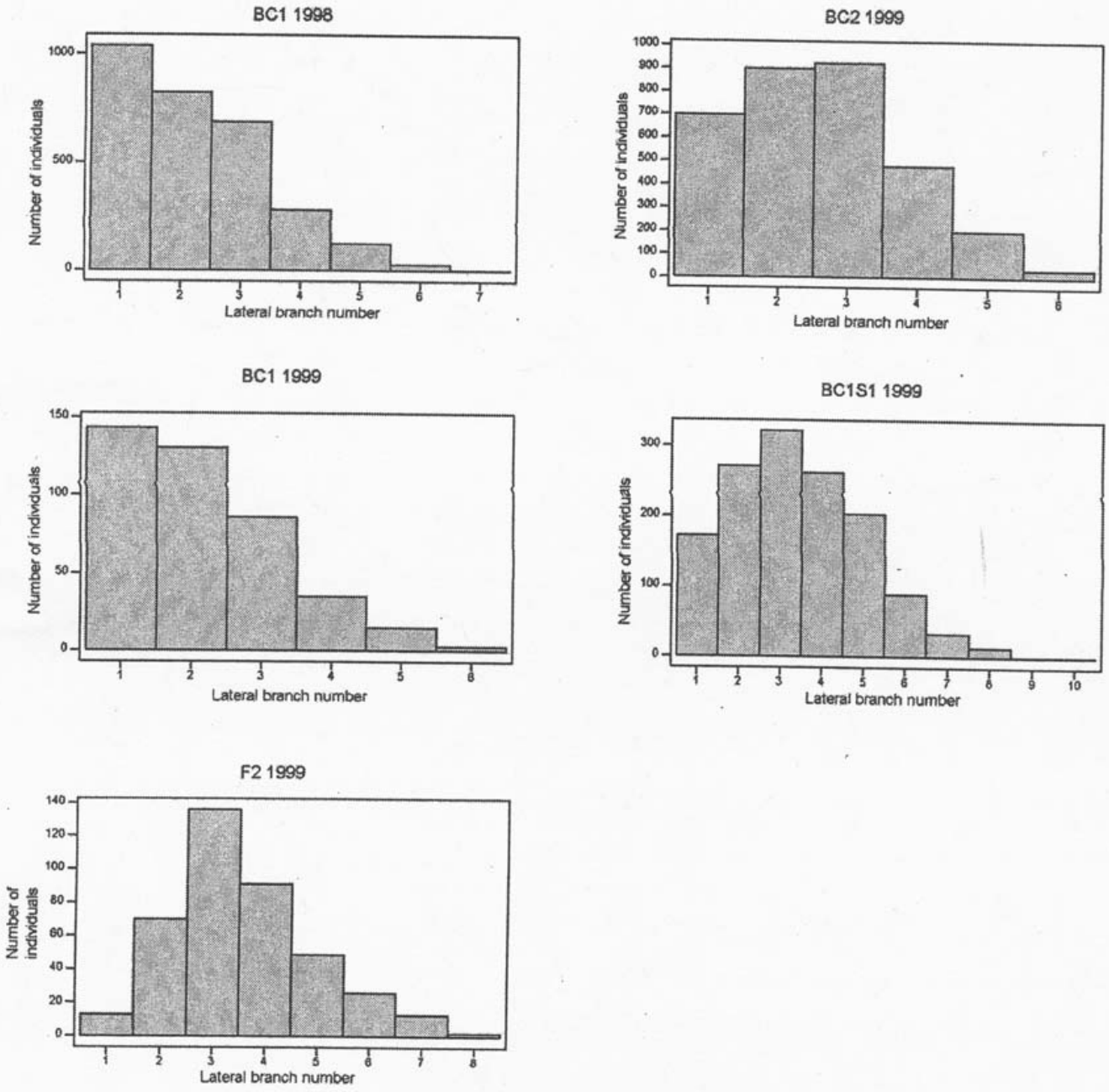

Figure 1. Distributions of multiple lateral branching among F2 and backcross cucumber populations.

Cucurbit Genetics Cooperative Report 23:12-15 (article 4) 2000

Fruit Yield and Yield Component Correlations of Four Pickling Cucumber Populations

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Introduction. Increased fruit yield has been one of the primary breeding objectives in the development of pickling cucumber cultivars (14). For purposes of selection, the most efficient trait for measurement of yield in once-over harvest in North Carolina is fruit number per plot (14). Smith et al. (12) found that fruit number had a higher heritability than fruit weight and the two were highly correlated. An alternative to direct selection for yield is to select for traits that are highly correlated with yield, but may have higher heritability. Those traits, often referred to as yield components, may include stem length, number of branches per plant, number of nodes per branch, time until first flowering, number of pistillate flower per node, and percentage of fruit set.

Yield components have been used to study fruit yields in vegetable crops such as cucumber (1, 3, 9, 11, 13, 18). In some instances, yield components have been positively correlated with yield and could be selected to improve yield. In cucumber, several strong correlations were observed between fruit yield and yield components. However, few of those studies involved genetically-diverse pickling cucumber populations. the objective of this study was to determine yield components that were strongly correlated with fruit yield in four U.S. pickling cucumber populations.

Methods. Four pickling cucumber populations, NCWPB, NCMBP, NCEP1, and NCH1, were developed at North Carolina State University (15, 16). The genetic variance for fruit yield, earliness, fruit quality, and disease resistance decreases while the mean for each trait improves from the NCWPB population to the NCMBP population, and to the NCEP1 and NCH1 populations (15, 16). After intercrossing, each population was selected using modified half-sib recurrent selection to improve fruit yield, earliness, and shape (17). For this study, eight (1995) and 4 (1996) families were taken randomly from the latest cycle of each population and self-pollinated in the greenhouse. S_1 families were evaluated in a randomized complete block design with four replications in the spring and summer seasons of 1995 and 1996 at the Horticultural Crops Research Station, Clinton, NC. forty seeds were planted on raised, shaped beds on 27 April 1995 and 29 April 1996 for the spring season, and 13 July 1995 and 8 July 1996 for the summer season.

Plots of 3.1 m length were separated by 1.5 m alleys, with guard rows and 1.5 long end plots around the field. Recommended cultural practices for North Carolina were used throughout the experiment (10). Plots were thinned to 30 plants at first true leaf stage. Plants were harvested 5 September 1995 and 19, 22, or 23 August 1996 for the summer season. Time of harvest was when the check lines had reached 10% oversized (>51 mm in diameter) fruit stage (6). each plot was evaluated for number of branches, nodes, pistillate flowers, total fruit, oversized fruit, and cull (crooked- and nubbin-shaped) fruit. Plots were corrected to 30 plants each for plots with 16 to 30 plants (plots with fewer than 16 plants were considered missing to prevent biasing from stand correction). Plant stands were corrected to reduce mean differences in yield and components resulting from differences in stand. Pearson correlation coefficients between yield components and yield, among yield components, and among yield traits were determined. Since no statistical test for comparing the magnitudes of two correlations was available, a correlation (r) between yield components and total yield of 0.7 to 1.0 or -0.7 to -1.0 ($r^2 \geq 0.49$) was considered strong, while a correlation of -0.69 to 0.69 was considered weak (2).

Results. The majority of correlations (96%) among yield components ranged from -0.69 to 0.69, defined as weak for this study (Table 1). In a similar study with slicing cucumbers, 85% of the correlations among yield components in the latest selection cycle were considered weak (3). The only strong correlation was a negative association between the percentage of

pistillate nodes and the percentage of fruit set for the NCMBP population. This negative association could be explained by the phenomenon of fruit fruit inhibition observed for cucurbits. With first fruit inhibition, the development of other fruit is limited by the development of the first pollinated fruit 95). this phenomenon is thought to be caused by a limited amount of photosynthates, which can only support the growth of one fruit at a time (7). Thus, with first fruit inhibition, a plant with a high percentage of pistillate nodes would only be able to support a few fruit. As a result, the percentage of fruit set would be higher for a plant with a lower percentage of pistillate nodes.

The correlations between yield and its components at the latest cycles exhibited a high percentage (85%) of weak correlations (Table 1). All of the strong correlations between fruit yield and yield components were positive. For both the NCMBP and NCH1 population, the number of branches per plant exhibited a strong positive correlation with the number of total and marketable fruit per plant (Table 1). The differences in correlations among populations might be attributed to germplasm used to form each population. LJ90430, a multi-branched, multi-fruiting accession of *C. sativus* var, *hardwickii,* was used in the formation of all population except the NCEP1 population (15, 16). With that germplasm in each population, the NCWBP, NCMBP, and NCH1 populations might be expected to exhibit strong correlations between the number of branches and total fruit per plant. With over 1000 breeding accessions used in the formation of the NCWBP population, the *hardwickii* germplasm originally in this population was highly diluted, and this probably accounted for the lack of strong correlations between branch and fruit number per plant. Thus, both the NCMBP and NCH1 populations would still be exhibiting the strong correlation between branch and fruit number resulting from the *C*. s. var. *hardwickii* germplasm used in their development.

The percentage of pistillate nodes of the NCH1 population was positively correlated with the number of total, marketable, and early fruit per plant (Table 1). This relationship may be associated with *C.S.* var. *hardwickii* germplasm used in the development of this population. furthermore, the NCH1 contains more of this germplasm than the other three populations. Selection for an increased percentage of pistillate nodes in this population may have promise for increasing the number of fruit produced per plant. This selection may result in more gain in yield than direct selection for yield *per se* if heritability for the percentage of pistillate nodes is higher than heritability for yield. Narrow-sense heritability of fruit number in cucumber has been reported to be between 0.00 and 0.25 for most populations 11, 12). Narrow sense heritability for gynoecy in cucumbers was also low (0.20 to 0.25) with the variance in sex expression being mainly dominance variance (11). Sex expression in cucumbers is primarily governed by three major loci, *a, F, and G* (8). The minimum number of effective factors involved in sex expression has been reported to be five (11). Thus, even with a strong correlation between the percentage of pistillate nodes and the number of fruit per plant, indirect selection for yield based upon the percentage of pistillate nodes may or may not be advantageous for improvement of yield for once-over harvest.

Regarding correlations among yield traits, the total number of fruit per plant was positively correlated with the number of marketable fruit for each population (Table 1). Therefore, selection for an increased number of fruit per plant should also increase the number of marketable fruit per plant. A strong, positive correlation also existed between total and marketable fruit number when these populations were tested at a low plant density (4). Total fruit number was positively correlated with the number of early fruit per plant for the NCWBP and NCH1 population (Table 1). Selection for increased yield in these populations would increase the number of early-maturing fruit produced per plant. The number of early fruit per plant was positively correlated with the number of marketable fruit per plant for the NCWBP and NCH1 populations (Table 1). In both of these populations, selection for an increased number of marketable fruit per plant also would increase the number of early-maturing fruit per plant. the NCH1 population also exhibited a strong, positive correlation between marketable and early fruit number when the population was grown at a reduced planting density (4).

Table 1. Correlation coefficients[z] among yield components (branches per plant, nodes per branch, percentage of pistillate nodes, percentage of fruit set), between yield components and yield traits (total, marketable and early yield per plant), and among yield traits for the latest cycle in each population.

Trait	Nodes/branch	Pistillate nodes (%)	Fruit set (%)	Fruit number per plant		
				Total	Marketable	Early
NCWBP Cycle 5						
Branches/plant	**0.00**	**00.25**	**0.61***	**-0.01**	**0.06**	**-0.04**
Nodes/branch		**0.05**	**-0.29**	**0.38**	**0.21**	**0.06**
Pistillate nodes (%)			**0.16**	**0.51**	**0.32**	**0.36**
Fruit set (%)				**0.15**	**0.20**	**0.24**

Total fruit number					**0.92*** **	**0.80*** **
Marketable fruit number						**0.87*** **
NCMBP Cycle 10						
Branches/plant	**0.04**	**-0.50**	**0.16**	**0.72****	**0.76*** **	**0.07**
Nodes/branch		**0.31**	**-0.61***	**0.44**	**0.26**	**-0.40**
Pistillate nodes (%)			**-0.75****	**-0.20**	**-0.43**	**-0.17**
Fruit set				**0.02**	**0.21**	**0.55***
Total fruit number					**0.91*** **	**0.24**
Marketable fruit number						**0.35**
NCEP1 Cycle 9						
Branches/plant					**0.58***	**0.05**
Nodes/branch					**0.34**	**0.30**
Pistillate nodes (%)	**0.34**	**-0.61***	**-0.08**	**0.57***	**-0.19**	**-0.18**
Fruit set (%)		**-0.58***	**-0.38**	**0.21**	**0.37**	**0.31**
Total fruit number			**0.02**	**-0.14**	**0.97*** **	**0.53***
Marketable fruit number				**0.43**		**0.61***
NCH1 Cycle 10						
Branches/plant	**-0.54***	**0.41**	**-0.28**	**0.74****	**0.72****	**0.59***
Nodes/branch		**-0.26**	**-0.05**	**-0.26**	**-0.21**	**-0.13**
Pistillate nodes (%)			**-0.59***	**0.79*** **	**0.77*** **	**0.77*** **
Fruit set (%)				**-0.29**	**-0.29**	**-0.40**
Total fruit number					**0.99*** **	**0.83*** **
Marketable fruit number						**0.82*** **

*, **, *** Significant at P≤ 0.05, 0.01, 0.01, respectively.

[Z] Strong correlations (bold) were considered to be 0.7 to 1.0 and -0.7 to -1.0.

Literature Cited

1. AbuSaleha and O.P. Dutta. 1988. Interrelationship of yield components in cucumber. Veg. Sci. 15:79-85.
2. Cramer, C.S. 1997. Specific combining ability for fruit yield and shape, yield and yield components of cucumber (*Cucumis sativus* L.) populations improved using recurrent selection. Ph.D. Diss. North Carolina State Univ., Raleigh, North Carolina.
3. Cramer, C.S. and T.C. Wehner. 1998a. Fruit yield and yield component means and correlations of four slicing cucumber populations improved through six to ten cycles of recurrent selection. J. Amer. Soc. Hort. Sci. 123:388-395.
4. Cramer, C.S. and T.C. Wehner. 1998b. Fruit yield and yield components of cucumber populations grown at low plant density. *In*: J.D. McCreight (ed.) Cucurbitaceae '98, evaluation and enhancement of cucurbit germplasm, p. 277-285. ASHS Press, Alexandria, VA.
5. McCollum, J.P. 1934. Vegetative and reproductive responses associated with fruit development in the cucumber,. Memoir 163, Cornell Univ. Agric. Exp. Sta., Ithica, NY.
6. Miller, C.H. and G.R. Hughes. 1969. Harvest indices for pickling cucumbers for once-over harvested systems. J. Amer. Soc. Hort. Sci. 94: 485-487.
7. Pharr, D.M., S.C. Huber and H.N. Sox. 1985. Leaf carbohydrate status and enzymes of translocate synthesis in fruiting and vegetative plants of *Cucumis sativus* L. Plant Physiol. 77: 104-108.
8. Pierce, L.K. and T.C. Wehner. 1990. Review of genes and linkage groups in cucumber. HortScience 25:605-615.
9. Prasad, V.S.R.K. and D.P. Singh.1994. Genetic association and interrelationship between yield components in cucumber. J. Maharshtra Agric. Univ. 19:147-148.

10. Schultheis, J.R. 1990. Pickling cucumbers. N.C. State Ag. Extension. Horticulture Information Leaflet No. 14-A.
11. Serquen, F.C., J. Bacher, and J.E. Staub. 1997/ Genetic analysis of yield components in cucumber at low plant density. J. Amer. Soc. Hort. Sci. 122: 522-528.
12. Smith, O.S., R.L. Lower and R.H. Moll. 1978. Estimates of heritiblities and variance components in pickling cucumbers. J. Amer. Soc. Hort. Sci. 103L 222-225.
13. Solanki, S.S. and A. Shah. 1989. Path analysis of fruit yield components in cucumber. Prog. Hort. 21:322-324.
14. Wehner, T.C. 1989. Breeding for improved yield in cucumber. In. J. Janick (ed.), Plant Breeding Reviews 6:323-359., AVI Press, Stamford, CT.
15. Wehner, T.C. 1997. Three pickling cucumber populations: NCWBP, NCMBP, and NCEP1. HortScience 32: 941-944.
16. Wehner, T.C. 1988. Two special cucumber populations: NCH1 and NCBA1. HortScience 33:766-768.
17. Wehner, T.C. and C.S. Cramer. 1996. Gain for pickling cucumber yield and fruit shape using recurrent selection. Crop Sci.36: 1538-1544.
18. Yin, M. and H. Cui. 1994. Analysis of component traits for early yield in cucumber. Cucurbit Genet. Coop. Rpt. 17: 27-29.

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Testing Method and the Correlation Between Fruit Yield and Yield Components in Cucumber

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Introduction. An alternative to improving fruit yield *per se* in cucumber (*Cucumis sativus* L.) would be to select for characters which were highly correlated with yield and that had a higher heritability than yield. Recently, Cramer and Wehner (2, 5) identified certain yield components that were correlated with fruit yield of pickling and slicing cucumber populations. The potential exists for the selection of those components to improve yield in those populations.

The test method used by the plant breeder influences yield and yield components. Cramer and Wehner (2, 3, 5) observed that cucumber populations grown in hills (spaced plants) at low density (6,450 plants/ha) produced more branches, and total, marketable and early fruit per plant, pistillate nodes, and nodes per branch than populations grown in plots at a normal density (6,500 plants/ha). Wehner (8) also observed an increase in the number of fruit per plant as the plant density decreased from 123,500 to 10,300 plants/ha. In addition, gynoecious hybrids produced fewer pistillate nodes as the plant density increased from 84,000 to 256,000 plants/ha (7). Plot measurement of yield and yield components can be labor-intensive, time-consuming, difficult, and inefficient. Testing in single-plant hills would be less labor-intensive, faster, easier, and more efficient.

The objective of this study was to determine the effects of two testing methods on the 1) correlation among yield components, 2) correlation among fruit yield traits, and 3) the correlation between yield components and total fruit yield of cucumbers. We were interested in selecting yield components using the easier method of hills (low density) but only if the strong correlations were maintained.

Methods. The four pickling cucumber populations used were the North Carolina wide base pickle (NCWBP), medium base pickle (NCMBP), elite pickle 1 (NCEP1), and hardwickii 1 (NCH1). The four slicing cucumber populations used were the wide base slicer (NCWBS), medium base slicer (NCMBS), elite slicer 1 (NCES1), and Beit Alpha 1 (NCBA1). The populations differed in their genetic diversity and mean yield performance (9, 10, 11). Populations were developed using modified half-sib recurrent selection to improve fruit yield, earliness, and shape of the population (12, 13). Three cycles of selection were chosen from each population (0, 3, 5) for NCWBP; 0. 5. 10 for NCH1; 0, 5, 9 for NCEP1; 1, 5, 10 for NCES1; 0, 4, 8 for NCBA1) to represent early, intermediate and late cycles of selection (2, 5). Eight families were chosen at random from each population-cycle combination (four in 1995 and four in 1996) and self-pollinated in the greenhouse.

The experiment was a split-split plot treatment arrangement in a randomized complete block design with four replications in each of two seasons (spring, summer) in each of two years (1995, 1996) with two testing methods [plot (64,500) or hill (6,450 plants/ha)] (2, 3, 5). Whole plots were the right cucumber populations, subplot were three cycles of recurrent selection (early, intermediate, late) and sub-subplots were testing method [hill (3) or plot (30 plants per 3.1 m)]. The experimental factors, planting and harvesting dates, plot size, border plots, soil type, and cultural practices were identical to those reported by Cramer and Wehner (2, 4, 5).

Each test plot was evaluated for number of branches, leaves, pistillate flowers, and total, early (oversized), and culled (crooked and nubbined) fruit. Plants which had fewer than five leaves, no flowers, and a stem length less than 40 cm were considered weak and were eliminated from the plot. Plots were corrected to three plants per plot for the hill method, or to 30 plants per plot for the plot method if they had two plants for hills or 16 to 34 plants for plots (2, 3, 4, 5). Plots with fewer than

two plants for hills, or 16 plants for plot were considered missing. PathSAS (6) was used to determine correlations among yield components, among yield traits, and between yield components and total fruit yield per plant (2, 3, 5). Correlations of 0.70 and higher (positive or negative) were considered to be strong correlations while correlations between -0.69 and 0.69 were considered weak (2, 3, 5).

Results. The total correlations between yield components and fruit yield, among yield components, and among yield traits discussed for each population, season, and cycle combination for plot (1, 2, 5) and hill (1, 3) testing method have been published and will not be presented in this paper. Of interest here is the proportion of instances in which changes in correlation strength occurred when the testing method was changed. Correlations for certain population-cycle-season combinations will be presented when changes in correlation strength occurred with a change from plot to hill testing method.

For the majority of yield components and populations, correlation strength among yield components remained unchanged when testing method was changed (Table 1). The proportion of instances in which correlation strength did not change with testing method was greater than 0.75 for most yield component-population combinations. When the number of instances was averaged over populations for each crop type, the correlation strength among yield components was more stable over testing methods for the pickle populations than for the slicer populations (Table 1). When averaged over all eight populations, the proportion of instances in which the correlation strength did not change with a change in testing method was high for each yield component. However, several instances existed in which the change in testing method directly influenced correlations among yield components (Table 1). Both the NCMBS and NCES1 populations exhibited a proportion of correlation strength between the number of branches per plant and other yield components when the testing method was changed from plot to hill. In addition, the correlation strength of nodes per branch and other yield components varied with testing method in several instances for the NCWBS and NCMBS populations. For the NCEP1 and NCMBS populations, the correlation strength between percentage fruit set and other yield components varied with testing method in several instances.

Of the populations examined, the NCMBS population exhibited the lowest proportion of instances in which the correlation strength among yield components did not change with a change in testing method. The differences in correlation strength among yield components with a change in testing method may have resulted from similar differences in yield component means between plot and hill testing methods (1, 2, 3, 5). When averaged over all populations, the number of branches per plant, the number of nodes per branch, and the percentage of pistillate nodes were greater when plants were grown in hills than when plants were grown in pots. Those changes in component means would alter the correlations among yield components and would result in changes in correlation strength with a change in testing method.

Correlation strength between yield components and total fruit yield per plant remained unchanged when testing method changed for a majority of the population-yield component combinations (Table 2). The proportion of instances in which the correlation strength did not change with testing method was 0.67 or greater for 75% of the population-yield component combinations. Most of the correlations between yield components and total yield per plant at either testing method were considered weak because they ranged from -0.69 to 0.69 (1, 2, 3, 5). With this wide range of correlation values, correlations could change with testing method without a change in correlation strength. This wide range of correlation values for the weak correlation classification could explain the high number of population-yield component combinations where the correlation strength did not change. When the number of instances was averaged over populations for each crop type, the correlation strength between yield components and total fruit yield per plant was more stable over testing methods for the pickle populations than for the slicer populations (Table 1). When averaged over all eight populations, the proportion of instances in which the correlation strength did not change with a change in testing method was high for each yield component.

However, several population-yield component combinations were observed where the proportion of instances in which the correlation strength did not change with testing method was 0.50 or less (Table 2). More of these population-yield component combinations were observed for slicer populations than for pickle populations. For both the NCES1 and NCBA1 population, the correlation between the number of branches per plant and total yield per plant changed strength with a change in testing method in a number of instances. The NCBA1 population also exhibited two instances in which the correlations of total fruit yield per plant with the number of nodes per branch and percentage of pistillate nodes changed strength from plots to hills (Table 2). For both the NCWBS and NCES1 population, the correlation between percentage of pistillate nodes and total fruit number per plant changed strength with a change in testing method in 50% of the instances. the correlation between the percentage of fruit set and total fruit yield per plant changed strength from plots to hills for the NCMBP and NCWBS populations. The changes in correlation strength between total fruit yield per plant and yield components with a change in testing method may have resulted from similar differences in yield and yield component mean values between plot and hill testing methods (1, 2, 3, 5). When averaged over all populations, the total number of fruit per plant, the number of branches per plant, the number of nodes per branch, and the percentage of pistillate nodes were

greater when plants were grown using the hill method than when plants were grown using the plot method. These changes in yield and yield component means would alter the correlations between total yield and yield components and would result in changes in correlation strength with a change in testing method.

With regard to the correlations of total fruit yield per plant with marketable and early fruit yield, the proportion of instances in which correlation strength did not change with a change in testing method was 0.67 or greater for a majority of population-yield trait combinations (Table 3). Several population-yield trait combinations existed in which changes in correlation strength occurred for 50% of the cycle-season combinations (Table 3). For the NCBA1 population, a change in testing method changed the correlation strength between total and marketable fruit number per plant when the population was tested in both seasons (Table 2). For both the NCMBP and NCH1 populations, the correlation between total and early fruit yield per plant changed strength from the plot to the hill testing method for 50% of the instances observed (Table 3).

Table 1. The percentage of instances[z] in which the correlation between the yield component of interest and the other three yield components did not change when testing method was changed from plot to hill for eight cucumber populations.

Population	Branches/plant	Nodes/branch	Pistillate nodes (%)	Fruit set (%)	Average
NCWBP	0.89	0.72	0.72	0.89	0.81
NCMBP	0.83	0.83	0.89	0.89	0.86
NCEP1	0.94	0.78	0.78	0.61	0.78
NCH1	0.72	0.89	0.83	0.78	0.81
All Pickles	0.82	0.81	0.81	0.79	0.81
NCWBS	0.72	0.61	0.78	0.89	0.75
NCMBS	0.56	0.44	0.72	0.50	0.56
NCES1	0.50	0.67	0.83	0.67	0.67
NCBA1	0.78	0.78	0.78	0.78	0.78
All Slicers	0.64	0.63	0.78	0.71	0.69
Average	0.74	0.72	0.79	0.75	0.75

[z] The number of observations for correlation among yield components is 18 (population-yield component), 72 (population, crop-yield component), 144 (yield component), 288 (crop) and 576 (overall).

Table 2. The percentage of instances[z] in which the correlation between yield component and total fruit yield per plant did not change when testing method was changed from plot to hill for eight cucumber populations.

Population	Branches/plant	Nodes/branch	Pistillate nodes (%)	Fruit set (%)	Average
NCWBP	0.67	0.67	1.00	1.00	0.83
NCMBP	0.83	0.67	0.83	0.50	0.71
NCEP1	0.83	1.00	0.67	0.67	0.79
NCH1	1.00	0.67	0.67	0.83	0.79
All Pickles	0.83	0.75	0.79	0.75	0.78
NCWBS	0.83	0.83	0.50	0.50	0.67
NCWBS	0.67	0.83	0.83	0.67	0.75
NCES1	0.50	0.83	0.50	0.83	0.67
NCBA1	0.17	0.50	0.50	0.83	0.50
All Slicers	0.54	0.75	0.58	0.71	0.65

Average	0.69	0.75	0.69	0.73	0.71

[z] The number of observations for correlation between yield components and yield is 6 (population-yield component), 24 (population, crop-yield component), 48 (yield component), 96 (crop) and 192 (overall).

Table 3. The percentage of instances[z] in wyhich the correlation between yield traits and total fruit yield per plant did not change when testing method was changed from plot to hill for eight cucumber populations.

		Fruit yield per plant	
Population	**Marketable**	**Early**	**Average**
NCWBP	0.83	0.67	0.75
NCMBP	1.00	0.50	0.75
NCEP1	1.00	0.83	0.92
NCH1	1.00	0.50	0.75
All Pickles	0.96	0.63	0.79
NCWBS	0.83	0.83	0.83
NCMBS	0.67	0.67	0.67
NCES1	0.67	0.67	0.67
NCBA1	0.50	0.67	0.58
All Slicers	0.67	0.71	0.69
Average	0.81	0.67	0.74

[z] The number of observations for correlation between yield components and yield is 6 (population-yield trait), 12 (population), 24 (crop-yield trait), 48 (yield trait, crop), 96 (overall).

Literature Cited

1. Cramer, C.S. 1997. Specific combining ability for fruit yield and shape, yield, and yield components of cucumber (*Cucumis sativus* L.) populations improved using recurrent selection. Ph.D., Dissertation, North Carolina State Univ., Raleigh, North Carolina.
2. Cramer, C,S, and T.C.Wehner. 1998a. Fruit yield and yield component means and correlations of four slicing cucumber populations improved through six to ten cycles of recurrent selection. J. Amer. Soc. Hort. Sci. 123:388-395.
3. Cramer, C.S. and T.C. Wehner. 1998b. Fruit yield and yield components of cucumber populations grown at low planting density. In: J. D. McCreight (Ed.), Cucurbitaceae '98, evaluation and enhancement of cucurbit germplasm, pp. 277-285. ASHS Press, Alexandria, VA.
4. Cramer, C.S. and T.C. Wehner. 1998c. Stand correction methods for cucumber fruit yield. cucurbit Genet. Coop. 21: 18-20.
5. Cramer, C.S. and T.C. Wehner. 1999. Fruit yield and yield component correlations of four pickling cucumber populations improved through recurrent selection. Crop Sci. (in review).
6. Cramer, C.S., T.C. Wehner, and S.B. Donaghy. 1999. PATHSAS: A SAS computer program for path coefficient analysis of quantitative data. J. Hered. 90: 2560-262.
7. Lower, R.L., O.S. Smith, and A. Ghaderi. 1983. Effects of plant density, arrangement, and genotype on stability of sex expression in cucumbers. HortScience 18: 7370738,
8. Wehner, T.C. 1986. Efficiency of 3 single-harvest tests for evaluation of yield in pickling cucumber. Euphytica 35: 493-501.
9. Wehner, T.C. 1997. three pickling cucumber populations: NCWBP, NCMBP and NCEP1. HortScience 32: 941-944.
10. Wehner, T.C. 1998a. Three slicing cucumber populations: NCWBS, NCMBS, and NCES1. HortScience 33: 168-170.
11. Wehner, T.C. 1998b. Two special cucumber populations: NCH1 and NCBA1. HortScience 33: 766-768.
12. Wehner, T.C. and C.S. Cramer. 1996a. Gain for pickling cucumber yield and fruit shape using recurrent selection. Crop Sci. 36: 1538-1544.
13. Wehner, C.S. and C.S. Cramer. 1996b. Ten cycles of recurrent selection for fruit yield, earliness, and quality in three

slicing cucumber populations. J. Amer. Soc. Hort. Sci. 134: 322-326.

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Artificial Inoculation Methods for Screening Melons Against Melon Vine Decline

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Development of disease resistant melon cultivars is the most promising strategy to reduce the economical damage caused by melon vine decline. To date, field assays have been conducted to screen melon collections and segregating populations (2,7). However, in field conditions the lack of environmental stresses could lead to the absence of aerial symptoms (6). Then, root analysis for fungal damage is necessary to select resistant genotypes. However, root inspection in field is a very tedious task. Moreover, the variability found in aggressiveness of pathogenic fungal isolates from different geographic areas (1) requires quantification of inoculum pressure in each assay. Artificial inoculation methods to overcome all these difficulties are needed.

An assay was conducted using the resistant accession*C. melo var.agrestis* 'Pat 81' (3), nd the susceptible control *C. melo*'VC-187' (Tendral type). Both pathogens, *Acremonium cucurbitacearum* Alfaro Garcia, W. Gams Garcia-Jimenez (A) and *Monosporascus cannonballus* Pollack & Uecker (M), reported as the two main causal agents of melon vine decline in Spain (4), were included. The assay was designed as a 2 x 2 x 2 factorial, the three factors being: accessions ('Pat 81' and 'VC-187'), soil (NS: natural soil from a commercial melon field affected by melon and vine decline, mixed with peat 2:1 and fertilized, and SS: sterilized soil, NC autoclaved twice) and treatments (T1: BS: basic substrate plus 10^5 colony forming units of *A. cucurbitacearum* (isolates A-419 and A-499)/g of soil, T3: BS + M, basic substrate plus 40 colony forming units of *M. cannonballus* (isolates C-29 and C-31)/g of soil, and T4: BS+A+M. The pathogenic composition of the natural soil was previously studied, confirming the presence of aggressive isolates of *A. cucurbitacearum* and *M. cannonballus.*

A significant effect of the soil employed was observed (Tables 1 and 2) with higher root severity indexes in NS inoculated roots. This may be due to the existence of other secondary agents (opportunistic and saprophytes parasites) in natural soil. A significant soil x treatment interaction was obtained, as differences among treatments were only found when SS was used as basic substrate, probably due to a threshold effect of pathogen concentration in NS (5). In SS inoculated soil the more severe symptoms were found after M or A + M addition, suggesting that *M. cannonballus* is a more aggressive pathogen than *A. cucurbitacearum.*

The high level of resistance of 'Pat 81', previously reported in field assays (3), was confirmed, and mild symptoms (RSI<1.8) were found in the roots of this accession. Highly significant genotype x soil and genotype x treatment interactions were found, as NS increased the severity of symptoms in 'VS-187', much more than did SS. Also, the artificial inoculation treatments resulted in similar effects on the susceptible cultivars. However, the severity of vine decline symptoms in 'Pat 81' did not increase significantly due to the partial resistance of this accession.

The results obtained indicated that the NS is the most rapid and simple inoculation method. However, in order to obtain comparable results we recommend using it in preliminary screening assays, confirming resistance in soil artificially inoculated with a known pathogenic composition.

Table 1. ANOVA results of root severity index in two melon accessions after inoculation with two soil types and 4 inoculation treatments (See Table 2 for soil and treatment descriptions).

Root severity index	df	MS	F ratio
Genotype	1	209.40	322.80**
Soil	1	140.00	229.70**
Treatment	3	7.35	11.30**
GxS	1	18.55	28.60**
GxT	3	4.08	6.29**
TxS	3	3.49	5.37**
GxTxS	3	3.25	5.01**

**Significant at 1% level

Table 2. root severity index in two melon accessions after inoculation with two soils and four inoculation treatments.

Soil/Inoculum treatment[z]	'Pat 81'	'VC-187'
SS	0.02aA[y]	0.00aA
SS + A	0.20aA	2.07bB
SS + M	00.37aA	0.86bB
SS + A + M	0.26aA	3.00bB
NS	1.50bA	4.60cB
NS + A	1.33bA	4.95cB
NS + M	1.75bA	4.93cB
NS + A + M	1.80bA	5.00cB

[z] SS = sterilized soil (2X), NS - natural infested soil, A = *A. cucurbitacearum* inoculation (10^5 CFU/g), M = *M cannonballus* inoculation (40 CFU/g).

[y] Root severity index evaluated as 0 (healthy) to 5 (severely affected). Lower-case letters for comparison among treatments and capital letters for comparisons between accessions by a Duncan's multiple ranges test.

Literature Cited

1. Bruton, B.D., M.E. Miller and J. Garzia-Jimenez, 1996. comparison of *Acremonium* sp. from the lower Rio Grande Valley of Texas with *Acremonium* sp. from Spain. Phytopathology 86:3.
2. Cohen, R., Elkind, Y., Burger, R. Offenbach and H. Nerson, 1996. Variation in the response of melon genotypes to sudden wilt. Euphytica 87 91-95.
3. Iglesias, A., B. Pico and F. Nuez. 1999. *C. melo* spp. *agrestis.* Pat 81, an interesting genetic resource highly resistant to melon dieback. Phytopathology 89:S35.
4. Martyn, R.D. and M.E. Miller, 1996. *Monosporascus* root rot/ vine decline: an emerging disease of melon worldwide. Plant Disease 80:716-725.
5. Mertely, J.C., R.D. Martyn, M.E. Miller and B.D, Bruton, 1993. quantification of *Monosporacsus cannonballus* ascospores in three commercial muskmelon fields in South texas. Plant Disease 77:766-771.
6. Pivonia, S., R. Cohen, U. Kafkafi, I.S. Ben Ze'ev, J. Katan, 1997. Sudden wilt of melons in Southern Israel: Fungal agents and relationship with plant development. Plant Disease 81:1264-1268.
7. Wolff, D. and M. Miller, 1998. tolerance to *Monosporascus* root rot and vine decline in melon (*Cucumis melo* L.) germplasm. HortScience 33:287-290.

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A Strategy for Selection *Cucumis melo* L. Resistance Sources to Melon Vine Decline in Field Assays

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Vine decline is a major root-rot disease of melon crops (*cucumis melo* L.) around the world (3). Plants affected by this disease suffer root damage that leads to a gradual vine following and decay as the plant approaches fruit maturity Several soil-borne fungi, virus and even bacteria have been related with this complex disease. In Eastern Spain *Acremonium cucurbitacearum* Alfaro Garcia, W. Gams, Garcia-Jiminez and *Monosporascus cannonballus* Pollack & Uecker seem to be the main causal agents (3). The development of melon varieties resistant to vine decline is difficult. Most screening assays have been performed in the field, due to the lack of artificial inoculation procedures (2,7). In these conditions, it is necessary to characterize fungal isolates from each screening field to obtain comparable results. In field assays, the rate of collapsed plants is highly dependent on certain environmental conditions such as high temperatures, warm winds, and horticultural characteristics of each accession (growth cycles, fruit load, etc) (5,6). Evaluation of the conditions of the roots could allow for a better evaluation of the resistance level of each variety.

The pathogenicity of 2 isolates of *A. cucurbitacearum* and 2 isolates of *M. cannonballus* from our screening field were compared to other Spanish and American isolates (Table 1). Fungi were grown in artificial medium as previously reported (1), and 10^5 colony forming units of *A. cucurbitacearum* per g of soil, and 40 colony forming units of *M. cannonballus* per g of soil were used to inoculate 1.5 L pots, containing sterilized substrate, in which plants (2nd true leaf stage) of the susceptible *C. melo* cultivar VC-185 were transplanted. Forty-eight days after transplanting the roots were inspected for symptoms, and a root severity index (RSI) similar to that used by Mertely *et al.* (4) was recorded.

Significant differences in aggressiveness were found among isolates (Table 1), AC-1 and MI-2 caused the most severe root damage among *A. cucurbitacearum* and *M. cannonballus* isolates, respectively. The former was more aggressive than A-419. the reference isolate for this fungus. The latter was significantly more severe than isolates from Texas.

After characterizing the fungal isolates, the field resistance of different melon accessions was tested. Incidence of vine decay was variable with assay conditions. The first year all accessions completed their reproductive cycle without collapse. However, on observation of the roots, lesions caused by fungal pathogens were found (Table 2) All muskmelon cultivars displayed high RSI (3-4). However, RSI was significantly lower in *C. melo* var. *agrestis* 'Pat-81' (2.1), indicating a partial resistance to melon vine decline. In the absence of vine collapse, the scoring of root damage allowed for the selection of this resistance source.

The resistance of 'Pat 81' was assayed three more consecutive years. These years vine decline was more severe and most plants of susceptible cultivars died before completing their growth cycle, whereas the percentage of symptomless plants in 'Pat 81' ranged from 45 to 85%. Despite the high severity of aerial symptoms, all plants were inspected for root damage the last year (Table 2). Results were consistent with those observed the first year. Indeed, when stresses appeared the root severity index was highly correlated with the above ground disease symptoms, so RSI provided a measure of the potential risk of each genotype of suffering vine decline. due to the high severity of field attack and o the aggressiveness of *A. cucurbitacearum* and *M. cannonballus* isolates from our screening field, 'Pat 81' may be a useful resistance source against melon vine decline not only in Spain but in other affected areas.

Table 1. Aggressiveness of different isolates of *A. cucurbitacearum* and *M. cannonballus* on melon cv. VC-185.

Isolate (Origin)	N	RSI[z]
Control	17	0.00 ± 0.00^{y}

A. cucurbitacearum		
A-499 (Central Spain)[X]	20	1.20 ± 0.22
A-419 (Eastern Spain)[X]	41	2.30 ± 0.19
AC-1 (Screening field)	31	3.00 ± 0.15
AC-2 (Screening field)	6	2.08 ± 0.40
M. cannonballus		
MI-2 (Screening field)	10	3.50 ± 0.52
C-31 (Eastern Spain)[X]	10	1.05 ± 012
TX-970059 (Texas)[V]	10	1.38 ± 0.14
MI-1 (Screening field)	10	2.05 ± 0.39
TX-970064 (Texas)[V]	10	2.60 ± 0.30

[Z] Root severity index evaluated as) (healthy) to 5 (severely affected).

[Y] Mean values + standard errors.

[X] Provided by Dr. Garcia-Jiminex (Department of Vegetable Pathology of the UPV, Valencia, Spain).

[V] Provided by Dr. Bruton (USDA-ARS, Oklahoma, USA).

Table 2. Root severity index of different *C. melo* accessions tested against vine decline under field conditions in two years.

Accessions	**RSI[z]**
First year	
C. melo var. *agrestis* Pat 81	2.18 ± 0.11[y]
UPV-5079 (Amarillo type)	2.92 ±.25
Cantaloupe	3.13 ± 0.26
VC-21 (Piel de sapo type)	3.21 ± 0.31
VS-120 (Amarillo type)	3.32 ± 0.20
Acc-6 (Cantaloupe	3.53 ± 0.15
Mu-C-32 (Amarillo type)	3.57 ± 0.20
Kaffer (Egyptian cultivar)	4.20 ± 0.11
Fourth year	
C. melo var. *agrestis* Pat 81	2.66 ± 0.01

VC-185 (Amarillo type)	3.87 ± 0.09
VC-187 (Tendral type)	4.16 ± 0.14

[z] Root severity index evaluated as 0 (healthy) to 5 (severely affected)

[y] The mean values ± standard errors

Literature Cited

1. Armengol, J., R. Sales and J. Garcia-Jimenez, 1999. Evolucion de los danos causados por *Acremonium cucurbitacearum* en raiz de melon en sus primeros estados de desarrollo. Boil, San. Veg. Plagas, 25:265-277.
2. Cohen, R., Y. Elkind, Y. Burger, R. Offenbach and H. Nerson, 1996. Variation in the response of melon genotypes to sudden wilt. Euyphytica 87:91-95.
3. Martyn, R.D. and M.E. Miller, 1996. *Monosporus* root rot/vine decline: An emerging disease of melon worldwide. Plant Disease 80:716-725.
4. Mertely, J.C., R.D. Miller and B.D. Bruton, 1993. An expanded host range for the muskmelon pathogen *Monosporascus cannonballus* Plant Disease 77:667-673.
5. Pivonia, S., R. Cohen, U. Kafkafi, I.S. Ben Ze'ev, J. Katan, 1997. Sudden wilt of melon in Southern Israel: Fungal agents and relationship with plant development. Plant Disease 81:1264-1268.
6. Wolff, D.W. 1996. Genotype, fruit load, and temperature affect *Monosporascus* root rot/vine decline symptom expression in melon. In: Cucurbits Towards 2000. Proceeding of the VIth Eucarpia Meeting on Cucurbits Genetics and Breeding, Malaga, Spain, 280-284.
7. Wolff, D. and M. Miller, 1998. Tolerance to *Monosporascus* root Rot and Vine Decline in Melon (*cucumis melo* L.) Germplasm. HortScience 33:287-290.

Cucurbit Genetics Cooperative Report 23:27-29 (article 8) 2000

Selection of Snake Melon Lines (*Cucumis melo* var. *flexuosus*) Resistant to Different Races of Powdery Mildew (*Sphaerotheca fuliginea* (Schlecht ex Fr.) Poll. in Sudan

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Snake melon or snake cucumber is widely cultivated in the Sudan as well as in other countries from North Africa to India. It is used in fresh salad and in pickles. Only landraces that resulted from farmer selections are cultivated, and they are all susceptible to the prevailing pests and diseases. Among the most important diseases, powdery mildew is a limiting factor of melon production in all producing countries and conditions (8). In the Sudan the disease is widely spread, and chemical control, it possible, is economically and technically difficult to practice and may present some health and environmental risks. *Sphaerotheca fuliginea* (Schlecht ex Fr.) Poll. and *Erysiphe cichoracearum* DC were reported as powdery mildew causal agents in Sudan with *S. fuliginea* being more prevalent (4). Fusarium wilt is another serious disease of melon in sudan. viral diseases are prevalent in different cucurbits growing agro-ecosystems and now are considered as serious factors threatening cucurbit production. Among these, Zucchini Yellow Mosaic Potyvirus (ZYMV), Cucurbit Aphid-Borne Yellows Luteovirus (CABYV), Watermelon chlorotic Stunt Gemini virus (WCSV), Squash Mosaic Comovirus (SqMV) and Melon Rugose Mosaic Thymovirus (MRMV) were reported (1,2).

PI 414723 is resistant to powdery mildew, ZYMV, CABYV, *Aphis gossypii* Glover, fusarium wilt and Papaya Ringspot Potyvirus (PRSV) (6). Another melon accession from India, PI 124112, also was reported to have resistance to downy mildew (7, 9), powdery mildew, fusarium wilt and CABYV (5, 6), A program was initiated in 1994-95 to introduce the powdery mildew resistance of the PI 414723 line obtained from cornell University into the susceptible cultivar Shendi. the experiment was conducted at the University Research farm. Seeds were sown on raised beds 1m wide with 50 cm spacing between plants. Recommended cultural practices were used throughout the experiment. In the winter season (Nov. - Feb.) 1998-00 the F_1 ('Shendi' x PI 414723), F_2 , F_1BC_1 (F_1 x 'Shendi'), F_2BC_1 and (BC_1 X PI 124112) were planted. In the second season (Nov. - Feb.) 1999 - 2000 the same material was included with the addition of F_1BC_2[F_2 BC_1 (PMR=9) X 'Shendi']. In both seasons parents and differential genotypes for powdery mildew resistance were included. A rating scale of 1 to 9 (1=highly resistant) was used to evaluate powdery mildew resistance under natural inoculum conditions. One rating was done when 'Shendi' was completely infected (PMR=1).

In both seasons, *Erysiphe cichoracearum* and race 0 of *Sphaerotheca fuliginea* were not considered to be present since 'Nantais oblong' was infected (Table 2). 'Nantais oblong' is resistant to *E. cichoracearum* and race 0 of *S. fuliginea* (Table 1). A laboratory test of samples collected from the field confirmed *S. fuliginea* as the causal agent. In the first season (1998-99), *S. fuliginea* race 2 was predominant according to the reaction of the differential genotypes (Table 2). The differential genotype "PMR 45', which is resistant to race 0 and 1 (Table 1), was infected together with the susceptible 'Shendi' at the beginning of the season (Table 2). Since race 2 arrived first it is difficult to know the presence or absence of race 1 because there is no differential genotype resistant to race 2 and susceptible to race 1. Both PI 414723 and PI 124112 were resistant (PMR=9) while 'Shendi' was completely infected (PMR=1). The F_1 ('Shendi' x PI 414723) and F_1BC_1 (F_1 x 'Shendi') were intermediate in resistance to powdery mildew (mean PMR-4.9 and mean PMR=4, respectively. The F_2 ('Shendi' x PI 414723) showed a segregation with about 23% of the plants having the level of resistance of the PI 414723 whereas the F_2BC_1 showed a segregation with about 9% of the plants having the level of resistance of the PI 414723. The progeny of the cross (BC_1 x PI 124112) were completely resistant (PMR=9).

In the second season (1999-2000), *S. fuliginea* race 1 was the causal agent since the differential genotype 'PMR 45' remained resistant (Table 2) until evaluation time. At the same time the susceptible check 'Shendi' was completely infected (PMR=1) and PIO 414723 and PI 124112 were completely resistant (PMR=9). Towards the end of the season (3^{rd} week of Feb.) 'PMR 45' was infected suggesting the arrival of race 2. Therefore, during this season the rating was done mainly for race 1. The F_1 ('Shendi' x PI 414723) and the F_1BC_2 had a resistance rating of 5.0 and 4.0, respectively. the F_2 ('Shendi' x PI 414723) was segregating with 38% of the plants having the level of resistance of the PI 414723. The progeny of the cross (BC_1 X PI 124112) were segregating this time with 37% of the plants resistant and 63% of the plants susceptible.

It is obvious from the results that resistance to race 1 and 2 conferred by the PI 414723 is incompletely dominant since the F_1 is intermediate in resistance level. It is also evident from this study that race prevalence of *S. fuliginea* is changing from one season to another. In a previous study it was reported that race 1 prevails during the summer (June-Sept.) and race 2 in the winter (Nov.-Feb.) (4). For *S. fuliginea* race 2 resistance in melons, two recessive genes against the U.S.A. strain and one dominant gene against the French strain from PI 414723 were reported (3). Segregation observed in this study does not fit a single dominant gene or a two recessive gene model for resistance. This could be because *S. fuliginea* race 2 in Sudan is different from that of USA and the French strain. Further study of the race 2 strains in sudan is needed for the advancement of breeding for powdery mildew resistance. Artificial inoculation under controlled conditions is important to select for resistance to race 1 and 2 of *S. fuliginea.* Since the donor genotypes have multiple resistances, backcrossing is done on the plants that are free of other disease symptoms in an attempt to advance multiple disease resistant selections. The two loci that confer resistance to powdery mildew in PI 414723 and PI 124112 are not

allelic (Pitrat and Dogimont, unpublished data); therefore, breeding powdery mildew resistant genotypes using the two donor parents might permit the recombination of the two loci in F_1 hybrids.

Table 1: The reaction of differential genotypes to powdery mildew pathogens and races.

	Sphaerotheca fuliginea			*Erysiphe cichoracearum*	
Genotype	**Race 0**	**Race 1**	**Race 2**	**Race 0**	**Race 1**
Iran H	S[z]	S	S	S	S
Nantais oblong	R	S	S	R	R
PMR 45	R	R	S	R	S
WMR 29	R	R	R	R	R
MR 1	R	R	R	R	R
PMR 5	R	R	R	R	R
PI 124112	R	R	R	R	R
PI 414723	R	R	R	R	R

[z] R=resistant, S=Susceptible

Table 2 Observed reaction of differential hosts to infection of powdery mildew in two field seasons.

Season	**Iran H**	**Nantais oblong**	**PMR 45**	**WMR 29**	**PMR 1**	**PMR 5**	**PI 414723**	**PI 124112**
1998-99	S[z]	S	S	R	R	R	R	R
1999-00	S	S	R	R	R	R	R	R

[z]R=resistant, S=susceptible

Literature Cited

1. Hanan, A.M., C. Wipf-Scheibel, B. delecolle, M. Pitrat, G. Dafalla, and H. Lecoq. 1997. Melon rugose mosaic virus: characterization of an isolate from Sudan and seed transmission in melon. Plant Disease 81:656-660.
2. Lecoq, H., G. Dafalla, Y.F. Mohamed, H.M. Ali, C. Wipf-Scheibel, C. Desbiez, A.E. El Jack, S.K. Omara, and M. Pitrat. 1994. survey of virus diseases infecting cucurbit crops in eastern central and western Sudan. Univ. Khartoum J. Agric. Sci. 2:67-82.
3. McCreight, J.D., M. Pitrat, C.E. Thomas, A.N. Kishaba, and G.W. Bohn. 1987. Powdery mildew resistance genes in muskmelon. J. Amer. Soc. Hort. Sci. 112:156-160.
4. Mohamed, Y.F., M. Bardin, P.C. Nicot, and M. Pitrat, 1995. Causal agents of powdery mildew of cucurbits in Sudan. Plant Disease 79:634-636.
5. Pitrat, M., C. Dogimont, and M. Bardin. 1998. Resistance to fungal diseases of foliage in melon. p. 167-173, *In*: J.D. McCreight, (ed.), cucurbitaceae '98: Evaluation and enhancement of cucurbit germplasm, 30 Nov - 4 Dec, Pacific Grove, CA.
6. Pitrat, M., G. Risser, F. Bertrand, D. Blancard, and H,Lecoq. 1996. Evaluation of a melon collection for disease resistance. p. 49-57, *In*: M.L. Gomez-Guillamon, C. Soria, J. Cuartero, J.A. Tores, R. Fernandez-Moniz (eds.), Cucurbits Towards 2000: Proceedings of the Vth Eucarpia Meeting on Cucurbit Genetics and Breeding, 28-30 May, 1996, Malaga, Spain.
7. Sitterly, W.R. 1972. Breeding for disease resistance in cucurbits. Annu. Rev. Phytopathol. 10:471-490.
8. Sitterly, W.R. 1978. Powdery mildews of Cucurbits. p 360-379. *In*: D.M. Spencer, (ed.), The powdery mildews. Academic Press, London.
9. Thomas, C.E. and E.L. Jourdain. 1992. Evaluation of melon germplasm for resistance to downy mildew. HortScience 27:434-436.

Cucurbit Genetics Cooperative Report 23:30-31 (article 9) 2000

Variability Among Israeli Isolates of *Sphaerotheca fuliginea*: Virulence Races, DNA Polymorphism, and Fatty Acid Profiles

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Introduction. Powdery mildew is a limiting factor for the production of cucurbits worldwide (Sitterly, 1978). In Israel, *Sphaerotheca fuliginea is* the causal agent of the disease, whereas *Erysiphe cichoracearum*, though it occurs on cichory, has yet to be found in cucurbits (Cohen and Eyal, 1995). Different races of *S. fuliginea have* been identified on the basis of differential host specificity (Thomas, 1978, Bardin et al., Pitrat et al. 1998). However, *S. fuliginea race* determination can be difficult, as plant response to inoculation may differ for a number of reasons including environmental conditions, genetic variability within a fungal population and shifts in pathogen populations.

The application of molecular markers has been demonstrated as a powerful tool for the study of populations of pathogenic fungi. Therefore, the aim of our study was to assess three techniques, RAPD, ISSR and FA analyses, as additional tools for the identification of powdery mildew races.

Materials and Methods. Five melon cultivars were used to identify *S. fuliginea* races based on their response to inoculation (Cohen et al., 1996). *Sphaerotheca fuliginea isolates* were collected from various cucurbit species during different seasons and at selected locations in Israel.

To obtain sufficient quantities (30-50 mg) of conidia for DNA or fatty acid analyses, cucumber plants at the age of 2-3 true leaves were inoculated with the different isolates (prepared from cultures originating from single spores). Two weeks postinoculation, conidia were harvested by washing infected leaves with sterile water. The conidial suspensions were filtered through a glass fiber filter (GFA, Whatman) over a Buchner funnel. After filtration, conidia were vacuum dried, collected to 1.5 ml microfuge tubes and maintained at -80°C until analyzed.

Fungal DNA was isolated as described by Danin-Poleg et al. (1998). This protocol was found to be efficient for DNA extraction from the 30-50 mg of spores collected from each isolate. RAPD analysis was performed according to Williams et al. (1990) and ISSR analysis was performed according to Danin-Poleg et al. (1998).

Profiles of fatty acids: conidia (30-60 mg) from each isolate were processed as descriobed by Ben-Ze'ev et al., (1997) for fatty acid extraction. fatty acid profiles were identified by gas chromatography.

Results. RAPD and ISSR analyses were applied to detect polymorphism among 26 isolates of *S. fuliginea from* Israel. Only 2% of the 440 RAPD primers that were tested on isolates of *S. fuliginea yielded* reproducible polymorphic patterns. The nine primers that detected polymorphism were UBC primers: 807, 834, 840, 841, 861, 873. Cluster analysis of RAPD and ISSRT products did not result in grouping related to biological races.

The fatty acid composition of the 25 *S. fuliginea* isolates was analyzed and profiles were obtained using the method and library 'Fungi' (MIDI, 1992; 1993). They clustered in two groups of 12 and 14 isolates, with the larger cluster consisting of 2 subgroups. The 2 subgroups linked at <9 Euclidean distance units (EDU) while the 2 groups linked at ~27 EDU. Such distances would indicate two subspecific entities within one of two congeneric species (Greenberg, 1997). More detailed

information will be published elsewhere.

Discussion. Traditionally, races of *S. fuliginea were* identified on the basis of differential host specificity. Three races of *S. fuliginea have* been identified in the US (Thomas, 1978) and recently six races have been described in France (Pitrat et al., 1998). This implies that the number of possible or pathotypes is larger than could be identified by differential plants. Additional approaches for race identification and for assessment of genetic variability have therefore been tested.

In general, using RAPD and ISSR analyses, a low level of polymorphism was detected among isolates of *S. fuliginea* that were collected from a narrow geographic range, in which sexual mating occurs rarely, if at all. ISSR was slightly more efficient than RAPD in detecting polymorphism among the isolates (10% versus 2% of the primers detected polymorphism). However, the RAPD and ISSR profiles were not useful in distinguishing among *S. fuliginea* races, confirming the previous observations by Bardin et al. (1997) who employed RAPD and RFLP analyses. Fatty acid profiles obtained for the same isolates were found to be more promising for the distinction among races. The fatty acid profiles as belonging to two races definitely belonged to two different FA subgroups. Further studies are required to confirm this observation.

Literature Cited

1. Bardin, M., P.C. Nicot, P. Normand and J.M. Lemaire. 197. Virulence variation and DNA polymorphism in *Sphaerotheca fuliginea*, causal agent of powdery mildew of cucurbits. Eur J Plant Pathol 103:545-554.
2. Ben-Ze'ev, I.S., E. Levy, P. Goldshlag, T. Eliam and Y. Anikster. 1997. Characterization of *Puccinia* species by teliospore fatty acids. Phytioparasitica 25:265-266.
3. Cohen, R., Y. Burger, S. Shraiber, Y. Elkind and E. Levin. 1996. Influence of the genetic background and environmental conditions on powdery mildew of melons. Phytoparasitica: 162.
4. Cohen, Y., and H. Eyal. 1995. Differential expression of resistance to powdery mildew incited by race 1 or 2 of *Sphaerotheca fuliginea* in *Cucumis melo* genotypes at various stages of plant development. Phytoparasitica *23:223-230.*
5. Danin-Poleg, Y., G. Tzuri, N. Reis and N. Katzir 1998. Application of inter-SSR markers in melon Cucumis melo L.). Cucurbit Genetics Cooperative 25-28.
6. Greenberg, R. 1997. Variation in virulence, DNA polymorphism, and fatty acid profiles among isolates of *Sphaerotheca fuliginea.* M.Sc. thesis, The Hebrew University of Jerusalem.
7. MIDI (1992) Microbial Identification System -- Library Generation System -- User's Manual. Microbial ID, Inc. (MIDI), Newark, Delaware, USA.
8. MIDI (1993) MIS Software Update, February 1993. An introduction to the fungal database. Microbial ID, Inc. (MIDI), Newark, Delaware, USA.
9. Pitrat, M., C. Dogimont and M. Bardin. 1998. Resistance to fungal diseases of foliage in melon. In: J. McCreight (ed.), Proceedings of cucurbitaceae 1998, (Asilomar, CA USA, 30 Nov..-4 Dec., 1998).
10. Sitterly, W.R. 1978. Powdery mildews of cucurbits. In: The powdery mildews (ed D.M. Spencer) Academic Press, NY (pp 359-379).
11. Thomas, C.E. 1978. A new biological race of powdery mildew in cantaloupes. Plant Dis Rep 62:223.
12. Thomas, C.E., J.D. Kishaba, J.D. McCreight and P.E. Nugent. 1984. The importance of monitoring races of powdery mildew on muskmelon. cucurbit Genetics Cooperative 7:58-59.
13. Williams, J.G.K., A.R. Kibelic, K.H. Livak, J.A. Rafalsky, S.V. Tingey. 1990. DNA polymorphisms amplified by arbitrary primers are useful as genetic markers. Nucleic Acids Res. 18:6531-6535.

Cucurbit Genetics Cooperative Report 23:32-36 (article 10) 2000

Genetics of Resistance to Powdery Mildew and Aphids, and Screening of DNA Markers Linked to the Resistance Genes in Melon (*Cucumis melo* L.)

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Introduction. Many sources of resistance to *Sphaerotheca fuliginea in* melon (*Cucumis melo* L.) are currently available and a number of resistant genes have been identified (3) and are listed by Pitrat (4). In this report, the inheritance of powdery mildew and aphid resistance in the breeding line 'PMAR No.5' (introduced to Japan from the University of California USA in 1981) (7) has been studied. furthermore we have screened random amplified polymorphic DNA (RAPD) markers linked to these resistance genes.

Materials and Methods. The F_1, F_2 and backcross generations (BC-S = F_1x Susceptible parent and BC-R - F_1 x Resistant parent) from the crosses between 'PMR No.5' and 'Harukei No. 3' were obtained. 'PMR No.5' is a cantaloupe type and resistant to powdery mildew and aphids. 'Harukei No.3' is an Earl;s Favorite type and susceptible to them.

A leaf-disk assay for powdery mildew r4esistance was carried out as described (2). Race 1 of *S. fuliginea was* isolated from field grown

Har8ukei No. 3'. After seven days sporulation was recorded on a scale of 0 (= no sporulation) to 7 (= entire disk covered with heavy sporulation). The plants were grouped in three categories for X^2 analysis, based on disease ratings: resistant (score 0), intermediate (score 1) and susceptible (scores 3, 5 and 7).

At the 1-2 leaf stage, ten to fifteen aphids were placed on each plant for a mass infection test (6). After seven days we checked leaf-curling. The plants were grouped in two categories for X^2 analysis: resistant (no leaf-curling) and susceptible (leaf-curling).

Genomic DNA was extracted from young true leaves by using the plant DNA extraction kits, Nucleon Phytopure (Scotlab, Scotland), according to the manufacture's protocol. The PCR protocol was adapted from that of Yui et al. (8). Four DNA bulks from the F_2 population were used for a bulked segregant analysis (PMR - 10 powdery mildew resistant plants, PMS - 6 susceptible plants, AR - 10 aphid resistant plants, and AS - 10 aphid susceptible plants). A total of 1412 primers were screened for detection of RAPD between the PMR and PMS, and between the AR and AS. Unique fragments successfully amplified in the resistant bulk were named after the primer name with their size in base parts.

Results and Discussion. *Powdery mildew resistance.* All the F_1 plants and BC-R were resistant and the observed segregations fitted well with 13 resistant: 2 intermediate; 1 susceptible in the F_2, and 2:1:1 in the BC-S (Table 1). Segregation ratios suggested a digenic control of a completely and an incompletely dominant genes, in which the former is epistatic over the later.

Melon resistance to powdery mildew has been studied for a long time. The genetics, however, remain confusing. One of the reasons for the confusion is the different categorization of resistance among authors.

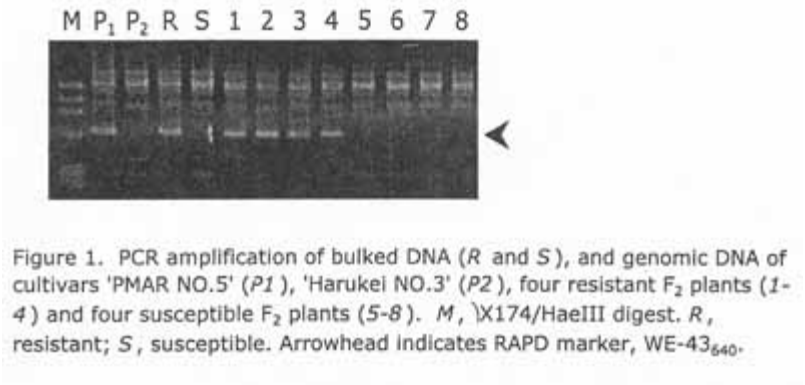

Figure 1. PCR amplification of bulked DNA (*R* and *S*), and genomic DNA of cultivars 'PMAR NO.5' (*P1*), 'Harukei NO.3' (*P2*), four resistant F_2 plants (*1-4*) and four susceptible F_2 plants (*5-8*). *M*, \X174/HaeIII digest. *R*, resistant; *S*, susceptible. Arrowhead indicates RAPD marker, $WE\text{-}43_{640}$.

By grouping in three categories (resistant, intermediate and susceptible), 'PMR No. 5' is found to have two resistance genes. To clarify whether these two genes are the same or different to those reported by other authors, allelism tests are needed.

A total of 60 primers successfully amplified unique fragments in the PMR bulk. For example, the marker, $WE\text{-}43_{640}$ amplified with the WE-43 primer (Table 2), was frequently observed in the PMR individuals and was scarcely found in the PMS ones in the F_2 population (Fig. 1). By further analysis, a total of 4 fragments were selected as candidates of linked markers. The segregation of these fragments in the BC-S population (120 plants) was further examined. The markers, $WE\text{-}43_{640}$, $OPX\text{-}15_{1100}$ and $UBC411_{770}$, were found to be linked to a completely dominant resistance gene at a distance of 43.4, 43.3 and 49.8 cM, respectively (Figure 2). The maker $UBC475_{970}$ was found to be linked to an incompletely dominant resistance gene at a distance of 47.5 cM (Figure 3).

*Aphid resistance.*The data produced evidence for a dominant monogenic control in 'PMR No.5' as all the plants in the F_1 and BC-R were resistant and the observed segregation fit a 3:1 resistant to susceptible ratio in the F_2 and a 1:1 ratio in the BC-S (Table 3). Yoshida and Iwanaga (6) also reported that the flat and curled phenotypes were controlled by a single gene (*Ag*) with flat leaves being dominant.

A total of 16 primers successfully amplified unique fragments in the aphid resistant bulk. By further analysis, the marker, $UBC401_{800}$, was found to e linked to the aphid resistance gene at a distance of 30.2 cM (Figure 4).

In this report, four RAPD markers linked to the PMR genes and one marker linked to the AR gene were found by bulked segregant analysis. However, the distance between the resistance gene and the marker is 30 to 50 cM. There could be several reasons why more tightly linked RAPD markers were not found. One possibility is the presence of other resistance genes with minor effects that make it difficult to correctly identify plants for the appropriate bulks. Baudracco-Arnas & Pitrat (1) and Wang et al. (5) also concluded that RAPD analysis was not the best solution or melon map construction because of skewed segregation.

Presently we are trying to identify DNA markers more tightly linked to powdery mildew resistance genes and aphid resistance gene.

Table 1. Observed segregation for powdery mildew resistance and the goodness of fit test.

Pedigree	**Number of plants**			**Expected [y] ratio**	***x*²**	***P***
	R[z]	**I**	***S***			
PMR No.5	37	0	0	37:0:0		
Harukei No. 3	0	0	53	0:0:53		

F_1	50	0	0	50:0:0		
F_2	164	24	12	13:2:1	0.074	0.964
BC-S	56	27	37	2:1:1	2.200	0.333
BC-R	116	0	0	116:0:0		

[z] R = resistant, 1 + intermediate, S = susceptible.

[y] The genetic model tested is one completely dominant gene and another incompletely dominant gene, with the former being epistatic to the later.

Table 2. RAPD markers for powdery mildew resistance (PMR) and aphid resistance (AR) in *Cucumis melo* L.

RAPD marker	**Trait**	**Size (bp)**	**Primer**	**Sequence of primer**	**Origin of primer**
$WE\text{-}43_{640}$	PMR (completely dominant)	640	WE-43	5'ACTCACAAATTG-3'	Wako Pure Chemical Ind. Ltd.
$OPX\text{-}15_{1100}$	PMR (completely dominant)	1100	OPX-15	5'CAGACAAGCC-3'	Operon Technologies, Inc.
$UBC411_{770}$	PMR (completely dominant)	770	UBC-411	5'-GAGGCCCGTT-3'	University of British Columbia
$UBC475_{970}$	PMR (incompletely dominant)	970	UBC-475	5'-CCAGCGTATT-3'	University of British Columbia
$UBC401_{800}$	AR	800	UBC-401	5'-TAGGACAGTC-3'	University of British Columbia

Table 3. Segregation and goodness of fit test of aphid resistance in *Cucumis melo* L.

Pedigree	**Number of plants[z]**		**Expected[y] ratio**	***x*²**	***P***
	R	**S**			
PMR No.5	10	0	10:1		
Harukei No. 3	0	20	0:20		
F_1	40	0	40:0		
F_2	145	51	3:1	0.1096	0.947
BC-S	35	25	1:1	1.667	0.435
BC-R	60	0	60:0		

[z] R = resistant, S = susceptible.

[y] A single, dominant gene model.

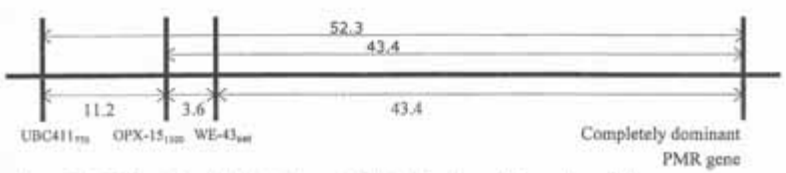

Figure 2. RAPD markers linked to the completely dominant gene for powdery mildew resistance (PMR) in *Cucumis melo* L. Distances between markers are shown in centiMorgans. For the markers see Table 2.

Figure 3. A RAPD marker linked to the incompletely dominant gene for powdery mildew resistance (PMR) in *Cucumis melo* L. Distance is shown in centiMorgans. For the marker see Table 2.

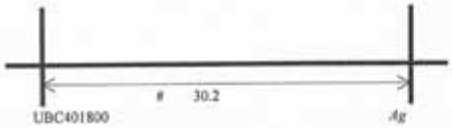

Figure 4. A RAPD marker linked to the aphid resistance gene (*Ag*) in *Cucumis melo* L. Distance is shown in centiMorgan. For the marker see Table 2.

Literature Cited

1. Baudracco-Arnas, S. and M. Pitrat. 1996. A genetic map of melon (*cucumis melo* L.) with RFLP, RAPD, isozyme, disease resistance and morphological markers. Theor. Appl.Genet. 93:57-64.
2. Epinat, C., M. Pitrat and F. Bertrand. 1993. Genetic analysis of resistance of five melon lines to powdery mildew. Euphytica 65: 135-144.
3. Floris, E. and J.M. Alvarez. 1995. Genetic analysis of resistance of three melon lines to *Sphaerotheca fuliginea.* Euphytica 81-186.
4. Pitrat, M. 1998. 1998 Gene list for melon. CGC Report 21: 69-81.
5. Wang, Y.H., C.E. Thomas and R.A. Dean. 1997. A genetic map of melon (*Cucumis melo* L.) based on amplified fragment length polymorphism (AFLP) markers. Theor. Appl. Genet. 95: 791-798.
6. Yoshida, T. and Y. Iwanaga, 1991. Resistance to cotton aphid (*Aphis gossypii* G.) in melon: its mechanism and selection methods. JARQ. 24:280-286.
7. Yoshida, T. and T. Kohyama. 1986. Mechanisms, genetics and selection methods of aphid resistance in melons, *cucumis melo*. Bull., Veg. & Ornam. Crops Res. Sta. Jpn. Ser. C9; 1-12 (In Japanese with English summary).
8. Yui, M., S. Monma, M. Hirai, S. Nishimura, Y. Ukai and S. Enomoto. 1999. Random amplified polymorphic DNA (RAPD) markers for the selection of tomatoes resistant to bacterial wilt. Bull. Natl. Res. Veg., Ornam. & Tea, Japan. 14: 189-98.

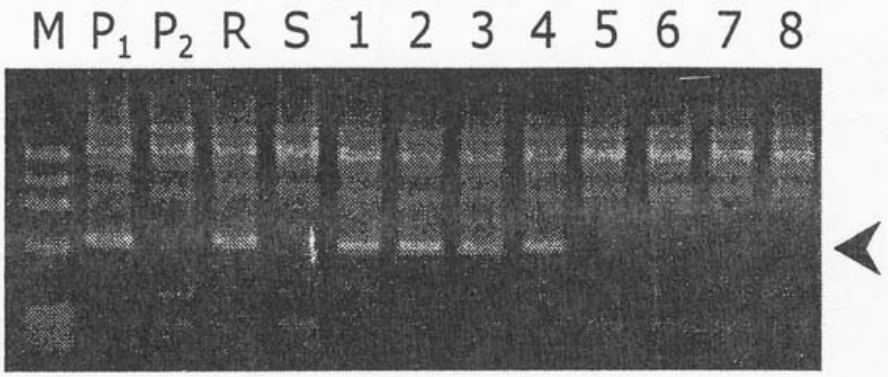

Figure 1. PCR amplification of bulked DNA (*R* and *S*), and genomic DNA of cultivars 'PMAR NO.5' (*P1*), 'Harukei NO.3' (*P2*), four resistant F_2 plants (*1-4*) and four susceptible F_2 plants (*5-8*). *M*, \X174/HaeIII digest. *R*, resistant; *S*, susceptible. Arrowhead indicates RAPD marker, WE-43_{640}.

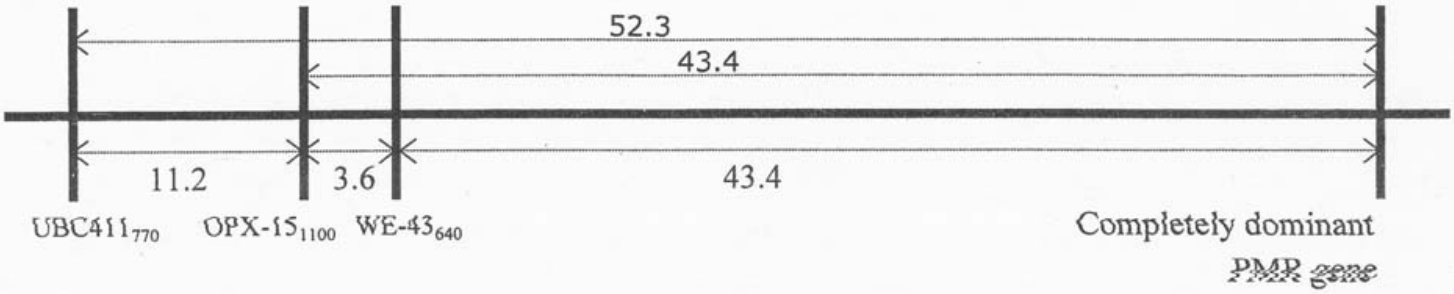

Figure 2. RAPD markers linked to the completely dominant gene for powdery mildew resistance (PMR) in *Cucumis melo* L. Distances between markers are shown in centiMorgans. For the markers see Table 2.

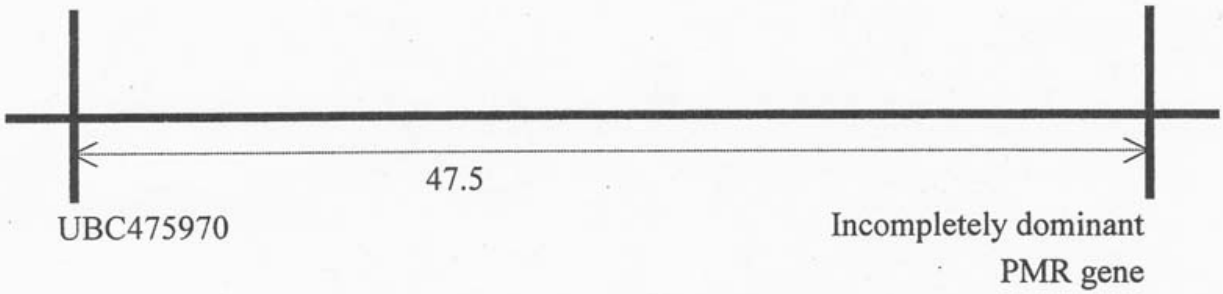

Figure 3. A RAPD marker linked to the incompletely dominant gene for powdery mildew resistance (PMR) in *Cucumis melo* L. Distance is shown in centiMorgans. For the marker see Table 2.

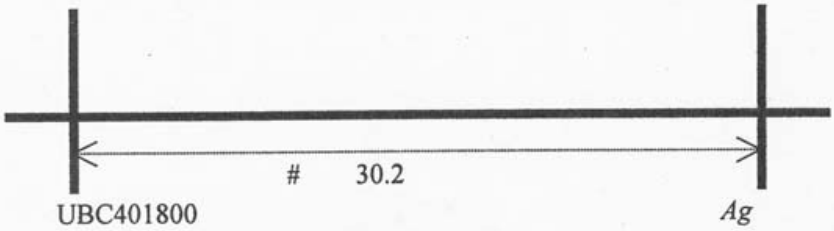

Figure 4. A RAPD marker linked to the aphid resistance gene (*Ag*) in *Cucumis melo* L. Distance is shown in centiMorgan. For the marker see Table 2.

Cucurbit Genetics Cooperative Report 23:37-40 (article110) 2000

Characterization of Identified Disease Resistant Lines in Melon, *Cucumis melo* L.

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Muskmelon, *Cucumis melo* L. is an important cucurbitaceous crop, being grown extensively in the garden land and riverbeds. Most important limiting factors in the production of muskmelon are devastating diseases like Fusarium wilt, GCMMV and mildews. Therefore, emphasis has been given to utilize available genotypes/accessions to identify and incorporate genes for disease resistance (Fusarium wilt, CGMMV and powdery mildew etc.), fruit quality attributes (fruit weight, fruit shape, flesh color, flesh thickness, bitterness/sweetness etc.) and vegetative characters (e.g. seedling marker, plant habit).

Several multiple-disease resistant genotypes and accessions have been identified and screened as a step towards variety improvement. Efforts are underway to combine multiple-disease resistance and superior quality attributes.

Materials and Methods. Seventeen genotypes were selected based on available descriptions in the *Cucumis melo L.*gene list (4) and information obtained through correspondence. These lines were characterized for vegetative and fruit traits. Seed viability was tested under controlled greenhouse conditions. Major disease screening (Fusarium wilt, powdery mildew, virus [e.g., CGMMV].) was conducted under field conditions at various stages of development up to fruit harvest. Several fruit characters were evaluated (See Table 2) and nutritional traits (total soluble solids [TSS], ascorbic acid) were also collected (Table 3). Standardized cropping practice was in the field during three years.

Total soluble solids (TSS) was measured using a hand refractioner and ascorbic acid content (mg/l 100 gm sample) was calculated based on a 2,6-dichlorophenol-indophenol visual titration method (6).

Results. Eight accessions (Table 1) obtained from INRA, Montfavet, France, were identified based on their field resistance (PDI < 25%) to *Fusarium* wilt, powdery mildew, and native viruses. Flesh color and plant habit were also characterized. 'MR-1' is a multiple disease resistant genotype (Table 1), known to possess genes for Fusarium wilt resistance (*Fom*-1, *Fom*-2) and powdery mildew resistance (*Pm*-*H*), PI 414723 (*Pm*-x), 'PMR 5' (*Pm*-2, *Pm*-E) and 'WMR 29' (*Pm*-w). Flesh color was found to be white, green and salmon in eight secessions with compact plant habit known to be contributed by *si*-1 gene in 'Top Mark Bush'.

Seed viability based on percent germination was found to be greater than 33% under controlled greenhouse conditions. In the field, maximum fruit settling occurred in WMR-29 (38.08 $\pm$ 7.12 percent).

Fruit characters of eight identified resistant genotypes given in Table 2 reveal small fruit size (Ogon-9), medium (Honey-dew, MR-1, Top Mark Bush and WMR-29), medium to large sized (PI 414723) and large (Nantais-oblong, PMR-5) with fruit weight in direct proportion to fruit size. Fruits differ widely in shape (round, oval, flattish round, oblong, pear-shaped and elliptically long), flesh color (white, green and orange), rind (netted, smooth and ribbed), and flesh-to-cavity ratio (1:1.54 [Nantais-oblong] to 1:2.83 [Top Mark Bush]).

TSS varied between 3.82 to 8.16, and ascorbic acid content was equal or greater than 20l0 mg/100 gm sample in five of the eight genotypes (Table 3).

Discussion. Eight genotypes with resistance to three major diseases and diversity in fruit quality traits can be utilized as parents in melon resistance breeding through conventional and non-conventional techniques. Artificial screening studies will further highlight the feasibility of incorporation of these lines in the improvement of existing cultivars.

A number of resistant accessions ('WMR-29' (2), PI 414723 (5), 'Casaba' (3)) have been reported and are being utilized further for multiple disease resistance and under other economically important attributes.

TSS and ascorbic acid differ with accession. Low TSS and high ascorbic acid content may be associated with field resistance to major virus and fungal diseases.

Molecular mapping of these identified genetic markers would facilitate the rapid marker assisted selection and cloning of the resistant genes. For example, markers have been identified for the *Fom*-2 and can be utilized in marker assisted selection (7,9).

Table 1. Description of eight resistant accessions obtained from INRA, Montfavet, France.

Accession	Description
Honeydew	Green flesh color, recessive to salmon.
	+ +

MR-I	Monoecious, (*a _g <_) , fusarium oxysporum melonis resistance (Fom-1, Fom-2), powdery mildew resistance (Pm-3, Pm-6), white seeds, white flesh*
Nantais oblong	Powdery mildew resistance (*Pm-H*)
Ogon 9	Yellow epicarp of mature fruit (*y*), white flesh, (*wf* epistatic to *gf*)
PI 414723	Powdery mildew resistance (*Pm-x*)
PMR 5	Powdery mildew resistance (*Pm-2, Pm-E*)
Topmark Bush	Compact plant habit (*si-1*)
WMR 29	Powdery mildew resistance (*Pm-w*), Papaya ringspot resistance (Prv^1)

Table 2. Fruit traits of eight disease resistant accessions.

Accession	Fruit weight (g)	Size and shape of fruit	Rind Color	No. of ribs	Fruit dia. (cm)	Flesh color	Flesh thickness	Cavity dia. (cm)	Fresh seed weight (g)	Nature and thickness of skin (cm)
Honeydew	725 ± 177	medium, round to oval	pale yellow	10	9.95 ± 2.18	diffused green-orange	2.32 ± 0.44	5.88 ± 1.24	125 ± 35	smooth, 0.25 ± 0.07
MR-1	610 ± 65	medium, flat-round to oval (splits on maturity)	pale orange	10	10/85 ± 0.31	orange-cream	2.68 ± 0.21	6.61 ± 1.05	121 ± 65	smooth, 0.21 ± 0.0
Nantais oblong	2063 ± 439	large, elliptically oblong (25.25 ± 2.10 cm long)	orange	absent	13.35 ± 2.25	orange	3.88 ± 0.31	6.0 ± 1.04	200 ± 58	smooth, 0.1 ± 0.0
Ogon 9	75.0	small, pear-shaped	yellow	10	3.97	green-white	0.9	2.23	-	0.1
PMR 5	2250 ± 707	large elliptically long (29.00 ± 7/07 cm long)	wild greenish orange	absent	13.18 ± 8.45	orange	3.40 ± 0.18	7.3 ± 0.23	225 ±106	rough, 0.2 ± 0.0
WMR 29	522 ± 40	medium, round	yellow	green, 10	10.20 ± 0.49	dark orange	2.61 ± 0.21	5.98 ± 0.52	88 ± 18	netted, 0.38 ± 0.2
PI 414723	650 ± 283	cylindrically long (15.5 ± 3.59 cm long)	green	absent	9.54 ± 0.05	cream-green (juicy fresh)	2.14 ± 0.67	5.75 ± 0.56	750 ± 35	smooth, 0.1 ± 0.00
Topmark Bush	783 ± 532	flat-round (3 sided) fruit	yellow	10	12.12 ± 1.09	cream-green (dry, crisp fresh)	2.88 ± 0.58	8.17 ± 0.83	154 ± 33	smooth, 0.15 ± 0.00

Literature Cited

1. Jain, J. and T.A. More, 1996. Identification and selection of genetic marker donor lines for incorporation of disease resistance in cultivars of *Cucumis melo* L. Cucurbit Genetics Coop Report 19: 53-56.
2. Nandpuri, K.S., Lal Tarsem and J.S. Dhiman, 1995. Muskmelon 'Punjab Rasila'. Ind. Hort 40 (3): 4,9.
3. Norton, J.D. 1981. Multiple disease resistant 'Casaba'. Cucurbit Genetics Coop. Report 4:24.
4. Pitrat, Michel, 1994. Gene list for *Cucumis melo* L. Cucurbit Genetics Coop. Report 17:135-142.

5. Guitton, L., L. Hagen, and M. Pitrat. 1999. genetic Control and linkages of some fruit characters in melon. Cucurbit Genetics Coop. Report 22:16-18.
6. Ranganna, S. Vitamins, *In*: Handbook of analysis and quality control for fruit and vegetable products. 2nd edition, pp. 105-106.
7. Wolff, D.W. and Z. Jianling. 1996. Potential utility of RAPD markers linked to Fom-2 gene in melon (*Cucumis melo* L.) - Cucurbit Genetics Coop. Report 19:61-62.
8. Zheng, X.Y., D.W. Wolff, S. Baudracco-Arnas and M. Pitrat. 1999. Development and utility of cleaved amplified polymorphic sequences (CAPS) and restriction fragment length polymorphisms (RFLPs) linked to *Fom*-2 Fusarium wilt resistance gene in melon (*Cucumis melo* L.) Theor. Appl. Genet. 99:453-463.

Cucurbit Genetics Cooperative Report 23:41-45 (article 12) 2000

Characterization of Local Varieties of *Cucumis melo*

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Introduction. Life cycles and the reproductive biology of plants are fundamental in determining cultivation and/or breeding strategies. Cucurbit species have an annual and a perennial cycle, and may have intraspecific variation in the phenological cycle. Thus, the determination of the phenological cycle may lead to the determination of genotypes which produce earlier fruits.

Various factors related to cucurbit reproductive biology have been investigated: proportion of male and female flowers, anthesis time, flowering time and duration, time of flower opening, and stigma receptivity and flower bud development (4). Studies on sex expression in some cucurbit species have shown that there are diverse phenotypes (monoecious, gynoecious, gynomonoecious and androecious). The most common type is monoecious, with the sexes in separate flowers (5).

Morphological characterization enables cultivars to be differentiated, as was shown by Wendel and Weeden (11) and Brandao et al., (1) who identified three *Cucurbita pepo* L. cultivars and five *cucurbita maxima* (Duschesne) cultivars, respectively, using seedling traits. Thirty-nine *Citrullus lanatus accesses* from Northeast Brazil were characterized and the genetic variability quantified using a descriptor list (7). *Cucumis melo fruits* are extremely polymorphic for shape and color (globe shaped, oval, elongated oblong, pubescent or glabrous, pale yellow, canary yellow or green in color).

'Melao coalhada' (Fig. 1) and 'Maxixe italiano' (Fig. 2) are commercialized on a small scale for consumption in juices and salads, respectively. Because of their potential expanded cultivation and breeding work, this study was carried out to obtain data on some of their phenological, plant, morphological and genetic (cytogenic and isoenzymatic) aspects.

Material and Methods. The plants described in this study were collected in the semi-arid area of the state of Bahia, in Baixa Grande county, where they are locally known as 'Melao coalhada' and 'Maxixe italiano'. The experiment was conducted in the field at Universidade Estadual de Feira de Santant (UEFS). Twenty seed per variety were sown at 2 seeds per hole, 1.5 m plant spacing, and 2.0 m row spacing in December, 1997.

The germination period was observed in days, along with the germination percentage and cotyledon and true leaf colors. The plant development was assessed by scoring the percentage of established plants, cotyledon and first true leaf color, length of the main branch, number of side branches, flower traits and flowering time, days to harvest of the first fruit, and the harvest peak. The presence of visiting insects was recorded. The fruit were characterized for size, shape, flesh color and mean number of seeds produced. They were later stored at room temperature and at 5°C, and the postharvest shelf-life was assessed after 15 days.

Cytogenetic analyses were carried out using the conventions Feulgen method (8) and electrophoresis in starch gels (12%) for the following enzymatic systems: esterase (EST), phosphoglucoisomerase (PGI), shikimate dehydrogenase (SKDH), malic enzyme (ME), peroxidase (PER), catalase (CAT), glutamate dehydrogenase (GDH), and glutamate oxaloacetate trasaminase (GOT). *Cucumis anguria*and *C. sativus varieties* were used to compare.

Isoenzymatic patterns from 'Melao coalhada' and "Maxixe italiano' were compared with one cantaloupe type. Cotyledon leaf tissue was used in the analyses and the migration methods were lithium-borate pH 8.3 for PGI, PER, GOT, SKDH (10), and tris-citrate pH.7.5 for CAT, EM, EST, and GDH (9).

Results and Discussion. Both the varieties germinated completely in 8 to 13 days and had a high percentage of plant establishment (Table 1).

'Melao coalhada' was approximately 130 cm long from the main branch while 'Maxixe italiano' was only 88 cm long (Table 2). Similarly, 'Maxixe italiano' flowers are smaller in size (Figure 3) than the commercial melon varieties. flowering occurred between 40 and 75 days after planting and the flowers of both varieties are single sex. The only visiting insects observed were *Apis melifera*, and the flowering periods coincided allowing cross fertilization and exchange of genotypes favorable to the crop.

'melao coalhada' was the earliest variety, with ripe fruit at 68 days after planting, and it had a lower number of seeds compared to 'Maxixe italiano' (Table 3). Further study should be done at different planting times to get a clearer picture of these traits.

'Maxixe italiano' has potential for large scale cultivation in the Brazilian northeast because of its organoleptic characteristics, which are similar to the cucumber (*Cucumis sativus*), its adaptation to semi-arid climatic conditions, and its small plant size, which makes it suitable for cultivation in smaller areas than those required by other cucurbits. Also, it maintains its fruit characteristics during a storage period (15 days) at 5°C.

The chromosome analyses revealed a diploid number 2n = 24 for both varieties (Figure 4), as observed for the other varieties of *C. melo*(2, 6). *Cucumis has* two basic numbers: species originating in India such as the cucumber are 2n = 14, whereas species such as *C. melo andC. agrarian* in Africa are 2n = 24. These were introduced to Brazil by the Negro slaves, and are today widely distributed throughout the Brazilian Northeast, where they are found with great morphological diversity which is reflected in innumerable varieties.

No differences were observed at the genomic group level in any enzymatic system x = 12 (*C. melo andC. anguria*) and x = 7 (*C. sativus*). The electrophoretic patterns were monomorphic for PER, EST, GOT and PGI systems. In other systems, differences were observed between *Cucumis melo, Cucumis anguria* and *Cucumis sativus*, showing characteristic patterns for each species (Figure 5). GDH, SKDH and ME had a polymorphic locus within *C. melo.* CAT showed two loci, with Cat-2 being polymorphic and having three different alleles, each one observed in a different genotype (Figure 6).

Table 1. Assessment of germination and seedling stages of two local varieties of *Cucumis melo.*

Variety	Germination (days)	Germination (%)	Cotyledon color	First true leave color
Melao coalhada	10	90	green	green

Maxixe italiano	13	100	green-yellow	green

Table 2. Establishment, growth, and reproductive cycle evaluation of two local varieties of *Cucumis melo*

Variety	Established plants (%)	Main branch length (cm)	Side branch No.	Days to first flower	Days to harvest of the first fruit	Days to harvest peak
Melao coalhada	90	129.11	5.7	41	68	89
Maxixe italiano	70	88.28	4.4	41	75	99

Table 3. Fruit characteristics of two local varieties of *Cucumis melo*

Variety	Fruits per plant	Fruit length (cm)	Fruit diameter (cm)	Fruit shape	Fruit weight (g)	Flesh color	Seed number
Melao coalhada	1.89	24.11	22.90	oblong	411.61	pale yellow	547
Maxixe italiano	2.14	9.38	17.66	globular	126.60	white	689

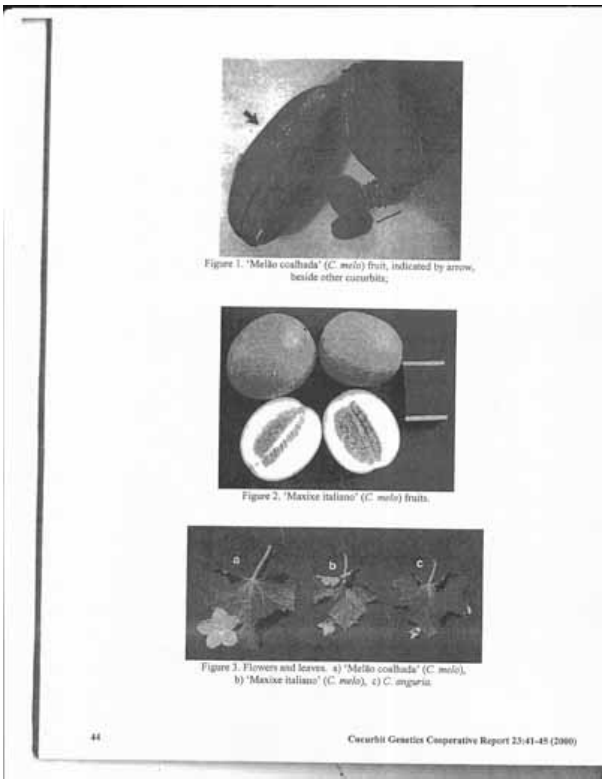
Figure 1. 'Melão coalhada' (*C. melo*) fruit, indicated by arrow, beside other cucurbits;

Figure 2. 'Maxixe italiano' (*C. melo*) fruits.

Figure 3. Flowers and leaves. a) 'Melão coalhada' (*C. melo*), b) 'Maxixe italiano' (*C. melo*), c) *C. anguria*.

Figure 4. Mitotic metaphase of 'Maxixe italiano' (*C. melo*.) (2n=24)

Figure 5. EST isozyme patterns of *Cucumis*. Right cucumber varieties (*C. sativus*); center right, 'Maxixe italiano' (*C. melo*); center left, null allele from *C. anguria;* left, varieties of *C. melo* ('Melão coalhada', cantaloupe and yellow melon).

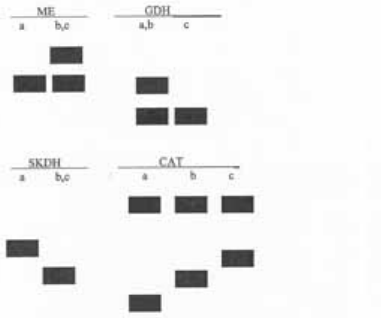

Figure 6. Polymorphic isozyme patterns between three genotypes of *Cucumis melo*. a) 'Melão cantalupe', b) 'Melão coalhada', c) 'Maxixe italiano'. See Materials and Methods for the enzyme system names.

Literature Cited

1. Brandao, J.C., V.M. de M. Andrae, and M.E. S.P. Dematte. 1981. Caracterizacao de Cucurbitaceae em fase inicial de desenvolvimento 2. Comparacao de plantulas de diferentes cultivares de *Cucurbita maxima*Duchenese. Cientifica 9(1): 113-119.
2. Dane, F. and T. Tsuchya,. 1976. Chromosome studies in the genus *Cucumis.* Euphytica 25:367-374.
3. Esquinas - Alcazar, J.T., and P.J. Gulick. Genetic Resources of Cucurbitaceae. Rome, OBPGR, 1983. 101p. (IBPGR - 82184).
4. Katrodia, J.S., P. Nath, and O.P. Dutta. 1974. Studies on floral biology in parents, F_1 . F_2 and backcross generations in *Citrullus lanatus* Thunb Mausf. Indian Journal of Horticulture 31(1):56-65.
5. Lopes, J.F. 1982. Melhoramento genetico (chuchu melanicia, melao e pepino). Inf. Agropec. 8(85):61-64.
6. Ramachandran, C., V.S. Seshardri, and R.A. Pai. 1985. Cytogenetical studies on dessert and non-dessert forms of muskmelon (*cucumis melo* L.). Cytologia 50:631-641.
7. Romao, R.L. 1995. Dinamica evolutiva e variabilidade de populacoes de melancia *Citrulus lanatus*(Thunb.) Matsum. & Nakai em tres regioes do Nordeste brasileiro. Piracicaba. Dissertacao (Mestrado) 75p.
8. Sharma, A.K, and A. Sharma. 1989. Advances in cell and chromosome research. Oxford and IBH Pub. Co., New Delhi.
9. Soltis, D.E., C.H. Haufler and D.C. Darrow. 1983. Starch gel electrophoresis of ferns: a compilation of grinding buffers, gel and electrode buffers, and staining schedules. American Fern Journal 73:9-27.
10. Wendel, J.P. and N.F. Weeden. 1990. Visualization and interpretation of plant isozymes. *In*: Soltis, D.E. and P.S. Soltis (eds.), Isozymes in plant biology. London: Chapmam and Hall p. 5-45.
11. Zaccaro, R.PO., M.E.S.P. Dematte, and V.M. de M. Andrade. 1981. Caracterizaco de Cucurbitaceae em fase inicial de desenvolvimento. 3. Comparacao de plantulas de diferentes cultivares de *Cucurbit pepo* L. (aboboreira). Cientifica 9(2):281-287.

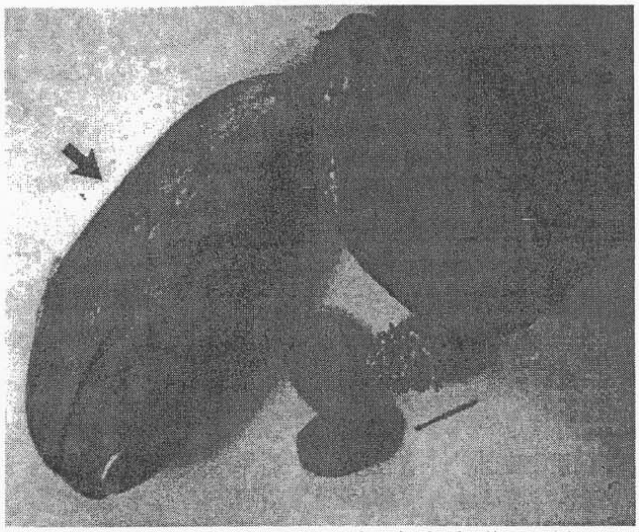

Figure 1. 'Melão coalhada' (*C. melo*) fruit, indicated by arrow, beside other cucurbits;

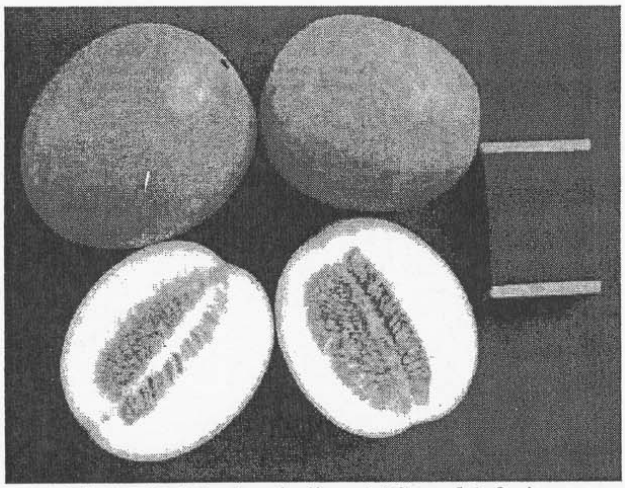

Figure 2. 'Maxixe italiano' (*C. melo*) fruits.

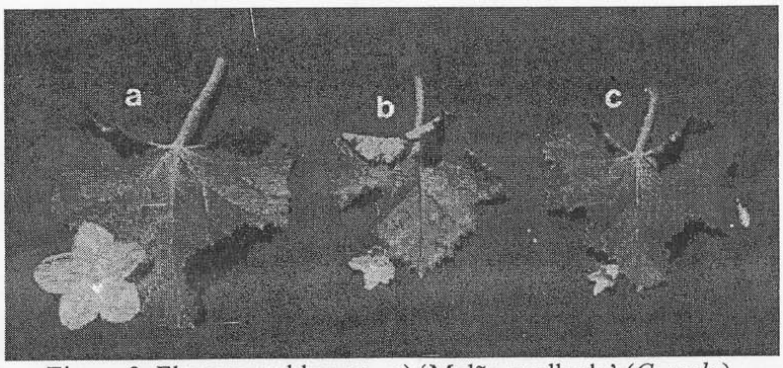

Figure 3. Flowers and leaves. a) 'Melão coalhada' (*C. melo*), b) 'Maxixe italiano' (*C. melo*), c) *C. anguria.*

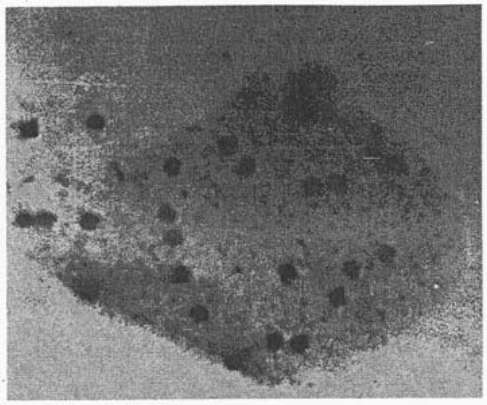

Figure 4. Mitotic metaphase of 'Maxixe italiano' (*C. melo*.) (2n=24)

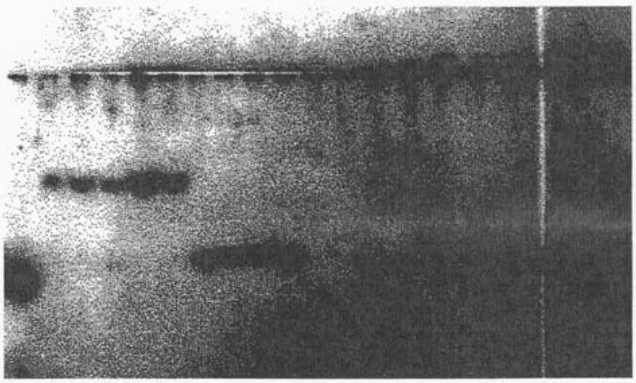

Figure 5. EST isozyme patterns of *Cucumis.* Right cucumber varieties (*C. sativus*); center right, 'Maxixe italiano' (*C. melo*); center left, null allele from *C. anguria;* left, varieties of *C. melo* ('Melão coalhada', cantaloupe and yellow melon).

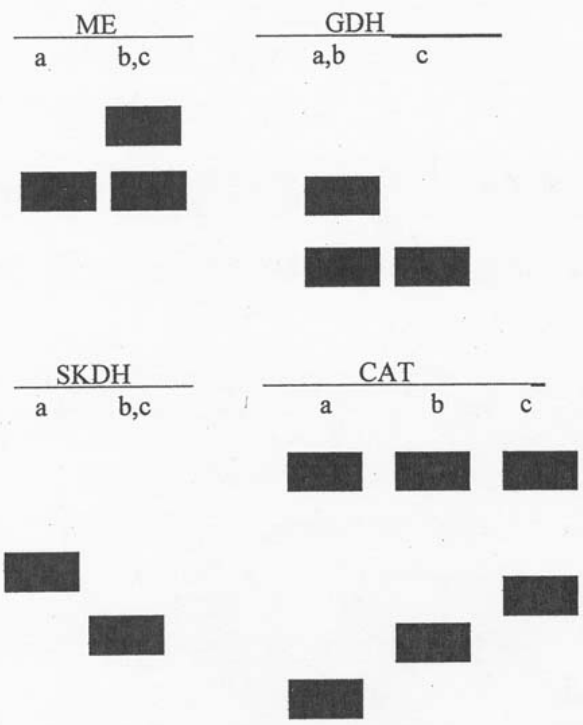

Figure 6. Polymorphic isozyme patterns between three genotypes of *Cucumis melo*. a) 'Melão cantalupe', b) 'Melão coalhada', c) 'Maxixe italiano'. See Materials and Methods for the enzyme system names.

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Citrullus lanatus - a Potential Host of Powdery Mildew in the Czech Republic

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Introduction .Within the cucurbits, watermelon (*Citrullus lanatus /* Matsum./ Thunb/ et Nakai) is generally considered the most resistant to powdery mildew (*Erysiphe cichoracearum (Ec), Sphaerotheca fuliginea (Sf) (8).*Its reaction to the Czech isolates of powdery mildew was evaluated as a part of our pathotype determination (3).

Material and Methods. A total of 72 isolates of powdery mildew (*Ersiphe cichoracearum / Ec/, Sphaerotheca fuliginea / Sf/*and mixed isolates of both species) were collected in 1997-1998 from field cultures of cucurbits (*Cucurbita pepo* and *Cucurbita maxima*) at 56 locations in the Czech Republic. they were maintained *in-vitro* on the cotyledons of *Cucumis sativus* cv. Marketer according to methods described by Bertrand (1).

Plant genotypes proposed by Bertrand (1) for pathotype determination were used. The differential set included *C. melo* genotypes Vedrantais and PMR 45, *C. sativus* cv. Marketer, *C. pepo* cv. Diamant F_1 , *C. lanatus* cv. Sugar Baby and a Czech cv. Golias of *C. maxima.* Seeds of *C. pepo andC. lanatus were* kindly provided by Dr. F. Bertrand (France).

Response of differential genotypes to the powdery mildew isolates was evaluated *in-vitro* on leaf discs. Intensity of sporulation was assessed at 4, 7, 10 and 14 days after inoculation on a scale of 0 (no sporulation) to 4 (more than 75% of the disc surface covered by mycelium). Isolates with an intensity of sporulation 0-1 were classified as virulent and those with scores 2-4 were considered virulent. The average value of infection degree (ID) on each genotype was expressed as a % of disc surface covered by mycelium at time of the last evaluation.

Results and discussion. Sporadic mycelium development on disc margins and surface during the first two evaluations, followed by a reduction of mycelium growth, were observed on 2 *Sf, 7 Ec* and 8 *Ec+Sf* isolates. None of 8 *Sf* isolates were virulent to *C. lanatus.* Of 33 isolates of *Ec* and 31 mixed isolates of *Ec + Sf*, only eight *Ec* and one *Ec + Sf* isolates were significantly virulent to *C. lanatus* (Table 1). All of these virulent isolate were collected in 1998. Similar high virulence to the watermelon within isolates acquired in 1997 was not observed. Isolate 3.98 (*Ec*) was virulent also to the *cucumis melo*MR-1 and isolate 19/98 (*Ec + Sf*) was virulent to the *C. melo* PI 124112 (4).

Isolates originating from Praha (Prague) (altitude of 352 m a.s.l. and an average temperature during the vegetative growth period of 14.2 °C) were collected in the early September and isolates from the Bohemian-Moravian Highlands (surroundings of Trebic; altitude of 406 m a.s.l. and an average temperature during the vegetative period of 12.6 °C) were collected August 24-25. These locations are not suitable for commercial cultivation of watermelon. Isolates originating from the warmest parts of the country did not express such a high virulence, in fact they were not virulent to the watermelon.

C. lanatus and *C. colocynthis* are mentioned as hosts of *S. fusca* and *E. orontii* in several countries of the West, Central and Eastern Europe (2). Both powdery mildew species were identified on watermelon in Slovakia in the 1980s (6) and symptoms of this disease were observed by A. L Kebeda on *C. lanatus grown* in a glasshouse in Moravia (Prostejov district) in the beginning of the 1990s/ At that time powdery mildew was not considered an economically important pathogen of watermelon.

A severe powdery mildew infection of *S. fulginea* on *Citrullus lanatus* was observed in Spain in the late 1990s (Dr. F. Bertrand, Avignon, France, 1999, pers. com.) and *Sf* caused economic losses in seedless watermelon production in California (USA) (7). Based on our investigation similar susceptibility of watermelon to the *e. cichoracearum can* be expected in the Czech Republic.

The geographic origin of the virulent isolates excludes their possible issue from host- pathogen interaction under natural and/or artificial ecopathosystems. On the contrary, the origin of these isolates could predict the probability of their virulence to watermelon. The host species *C. pepo* and *C. maxima are* grown throughout the Czech Republic and could serve as potential bridge species in spreading these isolates. Monitoring the virulence of pathogens is important not only under field conditions but also *in-vitro*. This strategy allows predicting epidemics and can be exploited by plant breeding programmes.

Table 1. Compatible response of *Citrullus lanatus* Sugar Baby to the Czech isolates of powdery mildew.

Isolate number	Pathotype[z]	ID (%) on *C. lanatus*`	Host plant	Location
E. cichoracearum				
12/98	AB1CD	33	*C. pepo* ZU[y]	Trebic
4/98	AB1B2CCmD	33	*C. maxima*	Trebic
21/98	AB1B2CCmD	33	*C. pepo* VM	Praha
9/98	ACCmD	50	*C. pepo* ZU	Trebic
7/98	B1B2CCmD	50	*C. pepo* PU	Trebic
1/98	AB1B2CCmD	50	*C. maxima*	Trebic
3/98,11/98	AB1B2CCmD	52	*C. pepo* ZU	Trebic
E. cichoracearum* + *S. fulginiea				
19/98	AB1B2CCmD	50	*C. maxima*	Praha

[z] *C. pepo fruit* shape according to (5): PU pumpkin, VM vegetable marrow, ZU zucchini.

[y] Comparable reaction to:

A	*C. sativus* cv. Marketer		C	*C. pepo* cv. Diamant F_1
B1	*C. melo* Vedrantais		Cm	*C. maxima* cv. Golias
B2	*C. melo* PMR 45		D	*C. lanatus* cv. Sugar Baby

Literature Cited

1. Bertrand, F. 1991. Les oidiums des Cucurbitacees. These Universite Paris-Sud-Orsay. Specialite "Phytopathologie", 225 pp.
2. Braun, U. 19915. Thepowdery mildes (Ersiphales) of Europe. Gustav Fischer Verlag, Jena, Germany 337 pp.
3. Kristkkova, E. and A. Lebeda. 1999. Powdery mildew of cucurbits in the Czech Republic - species, pathotype and race spectra. In. The First International Powdery Mildew Conference, Programme and Abstracts, August 29 - September 2, Avignon, France, pp 14-15.
4. Kristkova, E. and A. Lebeda. Reaction of *Cucumis melo* MR-1 and PI 124112 to the powdery mildew in the Czech Republic (prepared for submission to press).
5. Paris, H,S, 1986. A proposed subspecific classification for *cucurbita pepo.* Phytologia 61 (3):133-138.
6. Paulech, C. 1995. Huby mucnatkotvare (Erysiphales). Flor Solvenska X/1, Veda, Bratislava, Slovak Republic, 291 pp.
7. Paulus, A.O., G.J. Holmes, M. Vilchez and J.L. Aguiar. 1999. Fungicides for the cotrol of cucurbit powdery miildew *Sphaerotheca fuligine*a in California and North Carolina, USA. In: The First International Powdery Mildew Conference, Programme and Abstracts, August 29 - September 2, Avignon, France, pp. 44-45.
8. Sitterly, W.R. 1978. Powdery mildews of cucurbits. In. Spences, D.M. (ed.) The powdery mildews. Academic Press New York, San Francisco, pp. 359-379.

Cucurbit Genetics Cooperative Report 23: 49-50 (article14) 2000

In vitro Watermelon Genotype Screening by Adventitious Shoot Induction from Juvenile and Immature Cotyledons

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Introduction: Diploid watermelon (*Citrullus lanatus* (Thunb.) Matsum. & Nakai) genotypes were screened in order to determine the best explant material, as well as genotypes, for *in vitro regeneration.* This information will help other researchers who are conducting genetic transformation studies in watermelon, or any other *in vitro manipulation*, such as *in vitro* selection.

Two *in vitro* explant regeneration systems have been reported: one from juvenile cotyledons (1), and the other from immature cotyledons (2). Both systems use similar culture media, and regeneration is by adventitious shooting. For juvenile cotyledons, explants are taken five days after germination, however for immature cotyledons, explants are taken from fruits 28 days after pollination.

Materials and Methods: Sixteen genotypes of watermelon were used in this study, including commercial varieties, breeding lines and landraces. Their names and origin of each genotype are listed in Table 2.

At least 30 explants of each genotype and assay (juvenile or immature cotyledons) were used to evaluate *in vitro regeneration.* Regeneration was considered to have occurred if at least one shoot from an explant had developed by six weeks post-induction.

In vitro culture were observed after four and six weeks, and the following data were collected: explant number per genotype; number of callusing explants; number of explants with structures potentially regenerative; and number of shooting explants.

Analysis of Variance (ANOVA) was performed with each explant considered a replication (Table 1). The results are reported as the percentage of regenerated explants per genotype at six weeks post-organogenesis (Table 2).

Results: The ANOVA (Table 1) shows that there are differences among genotypes, as well as between the source of the explants. Comparisons among genotypes for the percentage of regenerable explants are found in Table 2.

The results found in Table 2, suggest that the best genotype for regeneration LPKKA when considering both sources of explants. LPKKA had 71.4% regenerated explants from juvenile cotyledons and 100% from immature cotyledons. PLKKA is followed by the landraces LG7 and LG8 when considering only the juvenile explants. LG8 had lower regeneration percentage with immature cotyledon. LPKKA is a breeding line, and therefore, could be used directly to introduce a trait into elite germplasm. The results from the landraces indicate that there is a great amount of available variability for *in vitro* selection that could be used in a crossing program.

In general, the best explant for regeneration was the immature cotyledon (Table 2). However, from a practical point of view, it is important to note that the immature cotyledon explant system, requires growing the plant past fruit setting, whereas juvenile cotyledon explants can be obtained after only two weeks growth.

Table 1. Analysis of Variance - Type III sums of Squares

Source	Sums of squares	Df	Mean squares	F-ratio	P-value
Main effects					
Genotype	2817.99	15	187.866	2.25	0.0135
Explant	72915.2	1	72915.2	872.30	0.0000
Interaction (G X E)	1524.06	15	101.604	1.22	0.2849
Residual	5182.53	62	83.5892		
Total (corrected)	95630.2	93			

Table 2. Percentage of regenerated explants

Genotype	Origin	Juvenile cotyledon	Immature cotyledon
DULCE	Novartis, S.A.	15.3 B[x]	94.4 bed
MARAVILLA	Ramiro Arnedo		
PANONIA	Seminis	33.3 abc	100 d
PATA NEGRA	Seminis	16.1 ab	100 d
LPKKA	Fito	71.4 c	100 d
LP3B	Las Gabias	18.45 ab	97.2 cd
LG4[z]	Las Gabias	8.33 ab	84.72 ab
LG6	Las Gabias	8 a	91.66 bcd
LG7	Las Gabias	45.11 bc	100 d
LG8	BGH[y]	33.3 bc	83.3 abcd
L12899	BGH	24.64 bc	100 d

L12900	BGH	18.2 ab	88.8 abcd
L12903	BGH	20.31 abc	100 cd
L12904	BGH	13.69 ab	100 cd
L12907	BGH	23.47 bc	100 d
L12911	BGH	30.35 bc	77.7 a
L12917	BGH	15.4 ab	100 d

[z] LG = Watermelon landraces from Las Gabias (Granada).

[y] BGH = Watermelon landraces from Horticulture germplasm, Spanish Bank (F. Nuez; Univ. Politecnica de Valencia. - Valencia Spain).

[x] Different letters indicate statistically significant differences between genotypes, P = 0.05% after LSD test.

Acknowledgements: *We appreciate the help of all the seed companies cited in Table 2 for their generous contributions of the genotypes used in this study. We thank Emila Romero for her help in the laboratory and field experiments. This project was partially funded by Fundacion para la Investigciou de la Provincia de Almeria (FIAPA).*

Literature Cited

1. Compton, M.E. and D.J. Gray, D.J. 1993. shoot organogenesis and plant regeneration from cotyledons of diploid, triploid and tetraploid watermelon. J. Amer. Soc. Hort. Sci. 118:151-157.
2. Zhang, X.P., B.B. Rhodes and J.W. Adelberg, 1994. Shoot regeneration from immature cotyledon of watermelon. CGC Rep. 17: 36-39.

Cucurbit Genetics Cooperative Report 23:51-53 (article 15) 2000

Watermelon Cultivars in the United States in 2000

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Plant breeders, seed industry personnel, crop advisors, and growers are interested in cultivars that are being grown in various parts of the country. Wehner (1) developed a list of cucumber cultigens to represent the diversity of the American cucumber market based on his genetic studies. The list of watermelon cultivars presented here was obtained by survey of knowledgeable individuals in ten of the most important watermelon producing states.

Diploid (seeded) cultivars are shown in Table 1. 'Allsweet', 'Black Diamond', 'Calsweet', 'Crimson Sweet', 'Jubilee II' and 'Legacy' are open-pollinated cultivars (opc) whereas the other 18 cultivars are listed as hybrids. The opc are grown mostly in one state suggesting regional adaptation or local demand. On the other hand, hybrids generally are grown in several states suggesting wider adaptation. the Allsweet type which usually is associated with high quality is represented by more than half of the listed cultivars m three opc and eleven hybrids. 'Sangria' and 'Royal Sweet' are popular in seven states. 'Fiesta' in six states and 'Mardi Gras' and 'Regency' in five states each.

Triploid (seedless) cultivars are shown in table 2. Almost half of the triploid cultivars are 'Tri-X-313' look-alikes. 'Tri-X-313 is popular in all ten states, 'Summer Sweet 5244' in nine states, and 'Millionaire' in eight states, 'Genesis' in five states and 'Tri-X-Shadow' in four states.

What conclusions can be drawn from these data?

1) The transition from diploid opc to diploid hybrid cultivars is nearly complete. Some unique opc will continue to be grown for local sales, but hybrids will be favored for large-scale production because of greater uniformity, reliability and generally enhanced yields and quality.

2) Only a relatively few watermelon cultivars are widely grown commercially even though there are hundreds of available cultivars.

Table 1. Diploid watermelon cultivars currently being grown in some of the principal producing states.

Cultivar	Arizona	California	Delaware	Florida	Georgia	Indiana	North Carolina	Oklahoma	South Carolina	Texas
Allsweet								X		
Athens	X									
Big Stripe										X
Black Diamond								X		
Calsweet		X								
Celebration				X			X		X	
Crimson Sweet			X				X		X	
Emperor						X				
Fiesta	X	X		X	X		X		X	
Jade Star			X							
Jubilee II								X		

Legacy								X		
Mardi Gras				X	X		X		X	
Pinata	X									
Regency				X	X	X	X		X	X
Royal Flush										
Royal Majesty			X			X				
Royal Star				X			X	X		
Royal Sweet		X		X	X	X	X	X	X	X
Sangria	X	X	X	X						X
Starbrite					X		X			
Stars-N-Stripes			X							X
Summer Flavor 500	X					X				
Summer Flavor 800					X	X				X

Table 2. Triploid watermelon cultivars currently being grown in some of the principal producing states.

Cultivar	Arizona	California	Delaware	Florida	Georgia	Indiana	North Carolina	Oklahoma	South Carolina	Texas
Gem Dandy								X		X
Genesis			X	X	X	X			X	
Laurel		X								
Millennium			X							
Millionaire	X	X	X	X	X	X	X		X	
Nova		X				X				
Revolution									X	X
Scarlet Trio								X		
Sugar Time								X		X
Summer Sweet 5244	X	X	X	X	X	X	X	X	X	
Summer Sweet 5544						X				X
Triple Sweet										

Tri-X-Carousel					X					
Tri-X Palomar				X	X					
Tri-X Shadow				X			X		X	
Tri-X 313	X	X	X	X	X	X	X		X	X
Ultra Cool	X									

Acknowledgments: *The assistance of Frank Dainello, Tim Hartz, Richard Hassell, Terry Kelley, Liz Maynard, Jim Motes, Jonathon Schultheis, Kai Umeda, and Tracy Wooten in providing information on watermelon cultivars grown in their state is gratefully acknowledged.*

Literature Cited

1. Wehner, T.C. 1994. A set of cucumbers to represent the American market. Cucurbit Genet. Coop. Rpt. 17: 12-13.

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Evidence for the Center of Diversity of *Cucurbita moschata* in Colombia

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Until recently, most researchers accepted southern Mexico-northern guatemala as the likely center of origin of *C. moschata as* proposed by Whitaker and Davis (5). However, others (1,2,6) have noted that pumpkins in northern Colombia are morphologically diverse. Nee (3) has speculated that this area might be the center of diversity of *C. moschata*. In contrast to the case of central America, little is known (or at least documented) about the extent of genetic variability in South america *C. moschata.* Collections exist in Colombia, Brazil and Bolivia, but are largely uncharacterized. Older literature from South America is difficult to access and has been largely ignored. The literature that is available often misidentifies species making the usefulness of this information somewhat suspect. My purpose here is to document what I have learned about genetic variability of South American *C. moschata*, either by personal observation in markets, communications with colleagues, or field evaluations of germplasm, collections, and thus lend support to the hypothesis that the center of diversity and domestication of this species is northern South American, and possibly the north coast of Colombia.

The large size of *C. moschata* severely limits its characterization compared to other crops. I have either grown out or observed thousands of accessions of *Cucurbita*, including well over 500 accessions of *C. moschata.* Several characteristics in materials from Colombia and Bolivia not observed in germplasm originating in Central America, has lead me to believe that further investigation into variability of South American *C. moschata* would be valuable in the understanding of this crop. Of particular interest was the frequent presence of brown seeded morphs in land races from Panama, Columbia and Bolivia, and occasional Colombian land races from Panama, Colombia and Bolivia, and occasional Colombian land races with extremely small (5 mm) dark seeds. Bukasov (1) reported this trait in Colombian land races. Interestingly,this trait has not been noted in Brazilian germplasm, (M.A. de Queiroz, personal communication). Nothing in the literature, nor in my personal observations, indicates that this trait occurs in *C. moschata* germplasm originating north of Panama. Previous to my trip to Colombia I also noticed that fruits and plants of some Colombia accessions were particularly primitive in appearance. The fruits were small (<0.5 kg) with highly lignified and warty rinds. The plants were very viney, small leafed and indeterminate.

I visited markets in colombia from 15 - 22 May 1999 in two areas of the country: (1.) Cali and nearby towns (including Jamundi, Santander de Quyilichao, Puerto Tejada and Palmira, and (2.) Cartagena and small towns along the coastal highway between Cartagena and Baranquilla. Cartagena is located on the Caribbean coast of Colombia at the mouth of the Cauca river, while Baranquilla lies at the mouth of the Magdalena river. This is the general area where Nee (1990) suggests that the wild ancestor of *C. moschata* might be found. Cali is located in the upper Cauca Valley, an area now dedicated almost exclusively to sugarcane. However, small plots of *C. moschata* are grown all over the valley. The presence of feral *C. moschata* plants along almost every roadside is testament to how widely this crop was grown in the past. There are many important archeological sites in Colombia. Agriculture in the Americas may have its origin in areas similar to parts of the Cauca and Magdalena valleys that were once covered with tropical deciduous forests (4). Very likely *C. moschata* was part of those earliest agricultural systems.

The fruits I observed in markets around and in Cali were very variable in shape and color, but generally smooth skinned with little or no lignification. In fact, the fruits I observed at these markets were not markedly different from fruits I have seen in other places in central America or the Caribbean except that they may have been more commonly furrowed. However, one trait did stand out: approximately half of the fruits I observed here were dark-seeded. Interestingly, the market at Jamundi had only a few fruits (less than 10%) with brown seeds, while all fruit at the Puerto Tejada market were brown seeded.

In and near Cartagena fruits were much more primitive in appearance compared to Cali. As a group, these fruits were more primitive looking than fruits I have seen from Mexico or Guatemala. Nearly every fruit I observed was heavily furrowed and warty and often highly lignified. Seeds were brown with few exceptions. As in Cali, vendors and buyers alike seemed indifferent to variations in seed color. However, I was consistently told that people preferred the smooth, non-warty skin

types and that farmers tried to select for that trait. My visit coincided with the beginning of the main growing season when *C. moschata* fruits are relatively rare in the markets of the coast. Many vendors commented that what I was seeing were the *criollo*, or unimproved types. Nevertheless, a great deal of farmer selection must have been carried out over the millennia since *C. moschata* was first domesticated since these *criollo* fruits possessed thick (often >5 cm), fine textured and intensely orange flesh. Very attractive flesh color and thickness were observed in fruits in the Cali area as well. Brown seeded types at both locations were variable for seed size, intensity of brown color (ranging from almost black to golden brown), smoothness (from deeply etched to very smooth) and size, color and smoothness of margins.

I did not observe any wild species of *Cucurbita* while in Colombia, although I only had limited access to areas outside of markets because of the precarious security situation in that country. The area between Cartagena and Baranquilla deserves further study as a site for the possible location of the wild ancestor of *C. moschata*. Recent studies carried out in collaboration with D. Piperno and O. Sanjur (Smithsonian Institute for Tropical Research, Panama City, Panama) and T. Andres suggest that this may be *C. argyrosperma* subsp. *sororia,*or something like it. *C. sororia* was recently found in Panama (T. Andres and D. Piperno, personal communication; 4). It's previous range was thought to be the southern U.S. to Nicaragua. It is adapted to ecological conditions (hot and dry) that are similar to those found seasonally in some areas of the Caribbean coast of Colombia.

The primitive and variable appearance of *C. moschata in* Colombia, particularly on the Caribbean coast, the presence of ecological conditions favorable for the growth of a putative wild ancestor, and the side-by side occurrence of traits otherwise found only in either South or Central America, suggest that this species was domesticated in Colombia and later carried north and south.

Acknowledgements: wish to thank Proyecto Atlantea, University of Puerto Rico for financial support for travel to Colombia.

Literature Cited

1. Bukasov, 1930. The cultivated plants of Mexico, Guatemala and Colombia. Bull. Appl. Bot. Genet. and Plant Breeding, Suppl. 47:1-553.
2. Lira-Saade, R., T.C. Andres and M. Nee. 19195. Chapter 1: *Cucurbita* L. *In:*Lira-Saade, R. (ed.), Taxonomic and ecogeographic studies of Latinamerican Cucurbitacea of economic importance. International Plant Genetic Resources Institute, Rome, Italy. [in Spanish]
3. Nee, M. 1990. The domestication of *cucurbita* (Cucurbitaceae). Economic Botany, 44(3) Supplement):56-68.
4. Piperno, D.R. and D.M. Pearsall, 1998. The origin of agriculture in the lowland neotropics. Academic Press.
5. Whitaker, T.W. and G.N. Davis. 1962. Cucurbita: Botany, cultivation and utilization. Interscience Publishers, New York.
6. Zhiteneva, N.E. 1929-30. The world's assortment of pumpkins. Trudy Prikl. Bot. 23:157-207.

Cucurbit Genetics Cooperative Report 23:56-57 (article 17) 2000

Duchesne is the Botanical authority for *Cucurbita moschata* and *Cucurbita maxima*

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The botanical authority cited for *cucurbita moschata* in scientific papers has varied among various writers. Whitaker and Davis (10), in their book "Cucurbits," gave the authority as Poiret, in reference to volume 11 of J.L.M. Poiret's "Dictionnaire des Sciences Naturelles." In "Hortus Third" (7), the authority for this species is given as (Duchesne) Poir. Other authors have given the authority as (Lam.) Poir., referring to the work of the renowned J.B.P.A. de M. de Lamareck, "Encyclopedie Methodique, Botanique." Both, Lamareck's Encyclopedie and Poiret's Dictionnaire were written in French and contain detailed descriptions of many plants, in alphabetical order.

In his Dictionnaire, Poiret (5) clearly gave credit to Duchesne for naming the species *Cucurbita moschata.* Although Lamarck himself had written almost all of the articles contained in the first two volumes of his Encyclopedie, two paragraphs before the end of the article "Courge, *Cucurbita*" (2), there appears the insignia "*Duch.*", indicating that this article had been prepared almost entirely buy Duchesne. However, in this article, the epithet *moschata* had been used as a subspecific entity of *C. pepo* and not as a specific entity. therefore, on the basis of these two well-known articles on *Cucurbita* in the works of Lamareck and Poiret, the botanical authority for *C. moschata* should be given as (Duchesne in Lam.) Duchesne ex Poir., or (Duchesne) Poir., or simply Poir. As *C. maxima* had been named and described as a species in the article "Courge, *Cucurbita*" in Lamarck's Encyclopedie, the authority for *C. maxima* should be given as Duchesne in Lam., or simply Duchesne.

However, *cucurbita moschata* was named and described by Duchesne in two publications that appeared decades before Poiret's Dictionnaire. The latter of these two was the article "Courge, *Cucurbita*" in volume 3 of "Encyclopedie Methodique, Agriculture", edited by Tessier and Thouin (4).

The first publication of *Cucurbita moschata* was the "Essai sur l'histoire naturelle des courges" (3). This is a 46-page duodecimo book. It does not bear a date or place of publication, nor the name of the publisher. from the methodology of printing during the late 18th century (8),from careful study of the print, and from the second title page of the book which indicates that it is an excerpt for Lamarck's Encyclopedie, it can be established without question that the Essai, like the part of the Encyclopedie containing the article "Courge, *Cucurbita*", was published in 1786 by C.J. Panckoucke of Paris.

The Essai is not merely a reprint of the article "Courge, *Cucurbita*" from the "Encyclopedie Methodique, Botanique." as it differs in several major points. Most relevant to this discussion is that it contains the binomial *Cucurbita moschata.* From the "Essai" it can also be learned that it was Lamarck who had considered this entity to be merely a subspecies of *C. pepo* , and that it had been published as such in the Encyclopedie because Lamarck was responsible for editing. Duchesne had insisted that *C. moschata* was a species separate from his *C. polymorpha* (-*C. pepo*). This point is also outstanding in the hand written, rough draft of what was to become the Essai and the article "Courge, *Cucurbita*" in the Encyclopedie (3).

Cucurbita maxima was presented as a separate species in both,the "Essai" and the article "Courge, *cucurbita" .* A minor point would be to establish priority of publication for this species. Part 1 of volume 2 of the "Encyclopedie" (the part that contains the article "Courge, *cucurbita*") was issued on 16 october 1786 (6). I have not been able to establish the exact date of issuance of the "Essai", but it contains a note added in proof dated 18 August 1786. The "Essai" contains some text which is slightly improved over that of the "Encyclopedie." indicating that it was printed subsequently. However, from the method of printing during that time we can establish with certainty that the "Essai" most have been printed immediately after the corresponding pages of the article. Thus, the printing of the "Essai" was completed long before that of the entire part 1 of volume 2 of the "Encyclopedie." It is more likely than not that the "Essai" was issued before part 1 of volume 2 of the "Encyclopedie." Thus, referring *C. maxima* to Duchesne in Lam. may well be incorrect.

In publications where the species name followed by the authority is given, these two species of *Cucurbita* are best presented as follows:

- *Cucurbita moschata* Duchesne
- *Cucurbita maxima* Duchesne

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Literature Cited

1. Duchesne, A.N. 1786. Courge, *Cucurbita*, in: J.B.P.A. de M. de Lamarck. Encyclopedie Metodique, Botanique 2: 148-159.
2. Duchesne, A.N. 1786. Essai sur l'histoire naturelle des courges. Paris, 46 pp.
3. Duchesne, A.N. 1786. Plan d'un article pepon pour l'encyclopedie de ....., 55 pp., in: Melange agronomique. Manuscript no. 12333, Bibliotheque Nationale, Paris.
4. Duchesne, A.N. 1793. Courge, *cucurbita*, in A.H. Tessier and A. Thouin. eneyclopedie Methodique, Agriculture 3: 605-614.
5. Poiret, J.L.M. 1818/ Courge, in J.L.M. Poiret. Dictionnaire des Sciences Naturelles 11:231-243.
6. Rickett, H.W. and F.A. Stafleu. 1961. Nomina genera conservanda et rejicienda sermatophytorum 8 bibliography. Taxon 10:111-121.
7. Staff of the L.H. Bailey Hotorium. 1976. Hortus Third, p. 343. New York.
8. Stafleu, F.A. 1964. Introduction o Jussieu's genera plantarum, in Jussieu, A.L. de. Genera Plantarum. Weinheim.
9. Stafleu, F.A. 1971. Lamarck: the birth of biology. Taxon 20:397-442.
10. Whitaker, T.W. and G.N.Davis. 1962. Cucurbits, p. 47. New York.

Cucurbit Genetics Cooperative Report 23:58-59 (article 18) 2000

Significance of Paintings (1769-1774) of *Cucurbita pepo* Fruits by A.N. Duchesne

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Cucurbita entered recorded history in the botanical herbals of the Renaissance period. From 1542 through 1700, over 50 original illustrations were published of various forms of this genus, almost all of them *C. pepo.* The period 1701-1850 has only a few published illustrations of Cucurbita. By far the greatest collection of *Cucurbita* illustrations of the period 1701-1850 is the paintings by A.N. Duchesne (1747-1827). These paintings, life-like and drawn true to color and size, were not published.

Duchesne began his study of *Cucurbita* in 1768, two years after the publication of his classic work on strawberries (3). His objective had been to determine taxonomic relationships among the various forms of *Cucurbita*, a genus that had been defined 15 years previously by Linne (8). Duchesne obtained seeds from nearly 100 *Cucurbita* cultigens and made cross-pollinations among them. the plants obtained through cross-pollinations were planted out and cross-pollinated, and so on for several generations. Duchesne documented his results by drawing, paying attention to the finest detail, the fruits of the cultigens and of their cross-pollinated progeny over successive generations He observed which stocks cross-pollinated and gave fertile offspring, and which did not. In this fashion, he was able to establish that the accessions of *Cucurbita* in his possession belonged to three species. One, which was highly polymorphic and for which Linne (8) had established four species, Duchesne called *Cucurbita polymorphia* (=*C. pepo*). Another, which usually had large, round fruits, that is, pumpkins, or in common French, Potirons, he appropriately named *C. maxima.*The third, whose fruit flesh had a musky flavor and aroma, known in French under several names including citrouille musquee, he named *C. moschata.*. Duchesne presented the result of his study before the French Royal Academy of Sciences in 1779. He read from a manuscript, using the paintings to illustrate and document his discussion. This manuscript has been lost, but a summary of his work was published in 1786 as a 46-page duodecimo book. Entitled "Essai sur l'histoire naturelle des courges" (5), its existence has been known only to several historians of botany. Modifications of this work appeared in two installments in encyclopedias published by C.J. Panckoucke of Paris (4,6).

The paintings reside today in the Central Library of the Museum National d'Histoire Naturelle in Paris. They are catalogued as manuscript no. 5007 (even though unaccompanied by any manuscript) and contain 258 64 x 48 cm plates containing 364 drawings of 615 fruits. Black and white photographs of approximately one-half of the drawings are in the herbarium of the Bailey Hortorium in Ithica, New York. L.H. Bailey had earlier learned of the existence of the Duchesne drawings from his article (4) in volume 2 of Panckoucke's "Encyclopedie Methodique" Botanique," of which Lamarck had been editor, and in 1946 Bailey requested and obtained the photographs now in the possession of the Hortorium bearing his name (1). The only other publications known to me that refer to these paintings, besides the works authored by Duchesne, were written by Buc'hoz (2), Sageret (9), and more recently, Duprat (7).

Bailey had studied the paintings at length and admitted that he did not know what taxonomic significance they might have, as he could not decipher Duchesne's numbering system. From the text (4,5,6), supported by the dates which many of the paintings bear, I have been able to decipher the numbering system. Significantly, the numbers not bearing a letter suffix represent fruits obtained form plants grown from the original seed stocks. Those bearing letter suffixes were borne by offspring resulting from cross-pollinations. Although the summaries of Duchesne's work (4,5,6) do not inform us as to how the original seed stocks were obtained, the paintings do allow us to determine the kinds of *Cucurbita* that had existed at the time. Some of the more interesting ones of *C. pepo* are:

- No. 1: Orange gourd
- No. 7: Bicolor, striped (=quadricolor) flat gourd
- No. 14: Striped pear gourd

- No. 14[1] : Bicolor, striped (=quadricolor) pear gourd
- Nos. 8, 15, and 17: Other bicolor gourds.
- Nos. 36 and 37: Orange warted gourds
- Nos. 62 and 63: Striped pumpkins
- Nos. 73 and 76: Cocozelle squash
- No. 83: Straightneck squash
- No. 85: Acorn squash
- No. 91: Scallop squash
- No. 92: Striped crown of thorns gourd

Nos. 73 and 76 are especially significant, being perhaps the first illustrations of cocozells squash. No. 83, likewise is significant, being perhaps the first illustration of straightneck squash.

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Literature Cited

1. Bailey, L.H. 1948. Jottings in the Cucurbitas. Gent. Herb. 7: 447-477.
2. Buc'hoz, P.J. 1777. Histoire universelle du regne vegetal 7 (text): 88-91.
3. Duchesne, A.N. 1766. Histoire naturelle des fraisiers. Didot,. Paris.
4. Duchesne, A.N. 1786. Courge, cucurbita. In: J/B/P/A. de M. de Lamarck, ed. encyclopedie Methodique, Botanique 2: 148-159. Panckoucke, Paris.
5. Duchesne, A.N. 1786. Essai sue l'histoire naturelle descourges. Panckoucke, Paris.
6. Duchesne, A.N. 1793. courge, Cucurbita. In: A.H. Tessier and A. Thouin, eds. Encyclopedie Methodique, Agriculture 3: 605-614. Panckoucke, Paris.
7. Duprat, G. 1964. Les dessinateurs d'histoire naturelle, en France au 18e siecle. In: M. Adanson. the bicentennial of Michel Adanson's "Families des plantes," part 2, pp. 451-470. Hunt Botanical Library, Pittsburgh.
8. Linne, C. von. 1753. Species plantarum 2: 1010-1011. Salvii, Stockholm.
9. Sageret, A. 1826. Considerations sur la production des hybrides des variantes et des varietes en general et sur celles de la famille des Cucurbitacees en particulier. Ann. Sci. Nat. Bot., I, 8.: 294-314.

Cucurbit Genetics Cooperative Report 23:60-61 (article 19) 2000

Cucurbita spp. and *Lagenaria siceraria* Collection at the Center for Conservation and Breeding of Agricultural Biodiversity (CCMAV), Polytechnical University of Valencia

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The Center for Conservation and Breeding of Agricultural Biodiversity, (CCMAV) located at the Polytechnical University of Valencia (UPV), is the reference center for the *Cucurbitaceae* family in the European Cooperative Programme for Crop Genetic Resources Network (ECP/GR), which is included in the International Plant Genetic Resources Institute (IPGRI). Considering the importance of genetic diversity preservation as a tool for crop improvement, it becomes essential to collect material, as well as conserve and characterize collections. The *Cucurbita* spp., and *Lagenaria siceraria* collection conserved at the Genebank of the CCMAV include 900 accessions belonging to 5 cultivated species, *Cucurbita pepo* L., *C. maxima* Duchesne, *C. moschata* Duchesne, *C. ficifolia* Bouche and *Lagenaria siceraria* (Mol.) Standl.

Most of these accessions were collected in Spain (2), and the remainder came from Central America and North Africa. A majority are traditional landraces, adapted to very variable ecological conditions, from mountainous dry lands to irrigated lands of the plains. Part of this collection was characterized during 1998 and 1999, following in part the *Cucurbita* descriptor of the IPGRI (1). A great diversity of types was found (3).

***C. pepo*:** Three hundred and eight accessions of *C. pepo* were collected in all the Spanish regions, Greece and North Africa, in variable ecological conditions (from sea level up to 1300 m). These pumpkins are used for human consumption, fried, roasted or tinned, and in vegetable stew. They also have less common uses as an ingredient of sausages, sweets (like 'Meloja"), pumpkin "bunuelos" or jam. The seeds are also consumed roasted.

Fifty-nine accessions were morphologically characterized, the weight ranging from 100g to 8.75 kg. In this species there is a great diversity in size, shape and colors. The elongated or elliptical zucchini, with yellow, orange or green colors predominate, even though some flattened, spherical and curved pumpkins can be observed, showing sometimes more or less pronounced ribs. the great variability found in this collection is consistent with previous information on this species, one of the most variable species of the vegetable kingdom with regard to fruit characters (4).

***C. maxima*:** The Genebank maintains 174 accessions of *C. maxima* collected in all the Spanish regions, Ecuador and Morocco, that were grown in low and intermediate altitudes (from nearly sea level up to 1300 m.). These accessions are basically destined for human consumption and are used boiled, roasted or fried, or in sausages and jam. they are also used for animal feeding and for decoration. One hundred accessions have been morphologically characterized, and a great variability in size, shape and colors has been found. The fruit weight ranged from 1.5 up to 20 kg. The turban-shaped pumpkins, with smooth skin and red and white colors; the flattened , wrinkled and dark ones; and the smooth grayish or orange-colored, which can reach considerable sizes, are most remarkable types.

***C. ficifolia*:** *The 81 existing accessions of C. ficifolia* have been collected in spain and Ecuador, in regions of higher altitude (from 12 to 2500 m. above sea level) than the aforementioned species. This pumpkin is basically used in confectionery, such as in "cabello de angel" elaboration. Some types are used boiled for human consumption and for animal feeding. the accessions characterized are highly monomorphic, weighing around 20 kg, with elliptical shape and with yellow and green veins when ripe.

C. *moschata*: The 187 accessions of *C. moschata,* collected in Spain and Ecuador are raised, like *C. ficifolia*, in intermediate and high altitudes, from 30p up to 1890 m above sea level. It is used for human consumption, boiled, fried, roasted or in vegetable stews, in sweets or desserts. It is also used, like the rest of species in the genus *Cucurbita*, for animal feed. The 40 accessions characterized weighed from 1 to 9 kg. The predominant shape of the fruit is pear-shaped, but some accessions with flattened, oblong, elliptic, heart-shaped and curved fruits are also found. The fruit usually have orange, yellow or green colors, with veins in the major part of the pear-shaped fruits. Most of them show ribs.

***Lagenaria siceraria*:** Fifty-five accessions of *L. siceraria* have been collected in Spain and Morocco in low altitudes, from 13 to 800 m above sea level.This species is not suitable for human consumption. It is basically used as a recipient for liquids, as a float and as decoration. This species shows a greater morphological uniformity than the rest of the species. All the fruits of the 10 characterized accessions are pear-shaped without ribs, green in color and occasionally with little white spots.

Literature Cited

1. Esquinas-Alcazar, J.T. and P.J. Gulick. 1983. Genetic resources of *Cucurbitaceae*: A global report. IBPGR Secretariat, Rome, 101 pp.
2. Nuez, F. M.J. Diez, J. Costa and J. Cuartero (1988). Germplasm resources of *Cucurbita* from Spain. Cucurbit Genetics Cooperative Rpt. 11:86.
3. Nuez, F., J.J. Ruiz, J.V. Valcarcel y P. Fernandez de Cordova. 2000. Coleccion de semillas de calabaza del Banco de Germoplasma de la Universidad Politecnica de Calencia. Ministerio de Agricultura, Pesca y Alimentacion. Instituto Nacional de Investigacion y Tecnologia Agaria y Alimentaria.
4. Paris, H.S. 2000., Segregation distortion in *Cucurbita Pepo.* Proceedings of 7th Eucarpia meeting on Cucurbit Genetics and Breeding, Acta Horticulturae 510: 199-202.

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Cucurbita argyrosperma Sets Fruit in Fields where *C. moschata* is the Only Pollen Source

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Cucurbita argyrosperma Huber and *C. moschata* Duchense can often be found growing in close proximity, or even in the same fields in Mexico and Guatemala. While Whitaker and Knight (9) state that these species seldom overlap in Mexico, Merrick (4) found that, at lower elevations in Mexico, these two species are commonly paired. The same is true in Guatemala where both species grow from 0 to 1500 m above sea level (Cesar Azurdia, University of San Carlos, Guatemala City, Guatemala, Personal communication). *C. argyrosperma* consists of two subspecies: *argyrosperma* and *sororia* (5). Subspecies *sororia* is either the progenitor or a weedy escape of subsp. *argyrosperma.* No wild or weedy populations are known for *C. moschata.* However, among species of *Cucurbita, C. argyrosperma* is clearly the most closely related to *C. moschata.* This is evident from crossing studies carried out by several workers (3, 4, 7, 8, L. Wessel0Beaver and T. Andres, unpublished). These studies indicate that fertile F1 plants can be rather easily obtained when using *C. argyrosperma* as the female parent in a manual interspecific cross. The reciprocal cross was never successful, indicating there are reproductive barriers between the species. I have found no reference in the literature directly confirming spontaneous hybridization under field conditions between *C. argyrosperma* and *C. moschata*. Spontaneous hybridization seems to occur between the domesticated and wild subspecies of *C. argyrosperma* (2, 6), although even in that case the evidence is indirect. However,m allozyme studies support the hypothesis that introgression occurs between *C. moschata and C. argyrosperma*(1). The objective of this study was to test whether open-pollinated fruit set with seed formation occurs in *C. argyrosperma under* field conditions where pollen is available only from *C. moschata.*

Materials and Methods: In Experiment #1, three plants of each of three populations of *C. argyrosperma* subsp. *sororia* (sor 80-1 and sor 177-1 from Mexico and sor 1 (P) from Panama) and three populations of subsp. *argyrosperma* (arg 46-3, arg 551-5, all from Mexico) were planted within a field of various genotypes of *C. moschata* on 17 September 1999 at the Isabela substation of the University of Puerto Rico (northwestern Puerto Rico, at an elevation of 138 m). Staminate flowers of the *C. argyrosperma* plants were removed every few days, before the flowers were able to open. Pistillate flowers were allowed to set fruit by open pollination. In experiment #2, five plants of each of one population of subsp .argyrosperma(arg 182-2 from Mexico) and one population of subsp *sororia* (sor 177-1 from Mexico) were planted within a field of *C. moschata on* 31 January 2000 at the Lajas Substation of the University of Puerto Rico (southwestern Puerto Rico, at an elevation of 80 m). As in Experiment #1, staminate flowers were removed and pistillate flowers were allowed to set fruit by open pollination during a two week period with *C. moschata* being the only source of pollen. Plants were pruned leaving 1 vine for subspecies *argyrosperma* and 2 to 3 vines for subspecies *sororia.* This was done to reduce the number of staminate flowers having to be removed, which could be hundreds in the case of *sororia.* Seed was removed from harvested fruits and embryo development was noted.

Results and discussion: all plants of all six populations of both subspecies of *C. argyrosperma* set at least one fruit during Experiment #1. However, due to heavy rains, I was unable to harvest fruits and evaluate seed development. In Experiment #2 each plant of both subspecies set several fruit during the two week period when staminate flowers were removed (Table 1). Fruit set ranged from 17 to 90%. Percentage fruit set was twice as high in subspecies *sororia* (73%) as in *argyrosperma* (36%). This same trend was observed by Merrick (3, 4) and in other work done in Puerto Rico by Thomas Andres and myself in manual cross, sib and self pollinations both within and between species. Domesticated *Cucurbita* often show a strong source/sink relationship where the presence of set fruit prevents or reduces set of later fruits. All plants in Experiment #2 produced fruits with at least some, and often many, partially to fully developed seeds. Again, differences were observed in subsp. *sororia* vs. *argyrosperma*: seed was often normal or nearly normal in subsp. *sororia* while no fruits of subsp. *argyrosperma* produced seed with fully developed embryos (cotyledons generally half-filled the seed coat).

Still to be tested is the viability of these seed as well as the fertility of the F_1 plants. My previous experience suggests that most of these partially developed embryos will germinate and that the F_1 plants will be fertile. continued studies will aid in determining what role introgressive hybridization has played or continues to play in the evolution of *C. argyrosperma* and *C. moschata.*

Table 1. Open pollination fruit set in *Cucurbita argyrosperma* subspecies *argyrosperma* (ARG) and subspecies *sororia* (SOR) following removal of staminate flowers. Plants flowered in a field *where C. moschata* was the only pollen source.

Taxon	Number of pistillate flowers opening during a 2-week period	Number of fruit set	Fruit set (%)

SOR-1	34	25	74
SOR-2	15	11	76
SOR-3	15	6	40
SOR-4	18	16	90
SOR-5	14	12	86
ARG-1	17	5	29
ARG-2	18	3	17
ARG-3	13	6	46
ARG-4	13	6	46
ARG-5	12	5	42

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Literature Cited

1. Decker-Walters, D.S., T.W. Walters, U. Posulszny and P.G. Kevan. 1990 Geneology and gene flow among annual domesticated species of Cucurbita. Can. J. Bot. 68:782-789.
2. Nabhan, G.P. 1984. Evidence of gene flow between cultivated *Cucurbita mixta* and a field edge population of wild *Cucurbita* at Onvas, Sonora.
3. Merrick, L.C. 1990. systematics and evolution of a domesticated squash, *Cucurbita argyrosperma*, and its wild and weeds relatives. In: D.M. Bates, R.W. Robinson and C. Jeffrey, eds., Biology and Utilization of the Cucurbitaceae. Cornell University Press, Ithica.
4. Merrick, L.C. 1991. systematics, evolution, and ethnobotany of a domesticated squash, *Cucurbita argyrosperma.* Ph.D. dissertation, Cornell University. 323 pp.
5. Merrick, L.C. and Bates. D.M. 1989. Classification and nomenclature of *Cucurbita argyrosperma* Baileya 23:94-102.
6. Merrick, L.C. and G.P. Nabhan. 1984. Natural hybridization of wild *Cucurbita sororia* Group and domesticated *C. mixta* in southern Sonora, Mexico. Cucurbit Genetics Coop Rpt. 7:73-75.
7. Whitaker, T.W. and W.P. Bemis. 1964. Evolution in the genus *Cucurbita.* Evolution 18:553-559.
8. Whitaker, T.W. and G.N. Davis. 1962. Cucurbits: Botany, cultivation, and utilization. Interscience, New York.
9. Whitaker, T.W. and R.J. Knight, Jr. 1980. Collecting cultivated and wild cucurbits in Mexico. Economic botany. 34:312-319.

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Relationship between Fruit Size and Seed Size in Cucurbits

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The seed yield of any fruit is a function of seed number and seed size. Seed number is determined early in fruit development by ovule fertilization. Seed size is determined later, and actually during the while period of fruit development, depending on a complex of environmental factors. Different types of relationships between fruit size and seed yield components have been reported. In *Capsicum* (3) and summer squash (6), a linear relationship between fruit weight and seed number was found. Less information is available concerning the relationship between fruit weight and individual seed weight. It is widely accepted that larger seeds of any crop have higher germinability and faster seedling development (1,4,5). This gives a potential advantage to the crop which is not realized in many cases (2). Herein are described the results of a cooperative study on the size of the fruit and the mean weight of its seeds, in four species of cucurbit crops.

Materials and Methods. Cucumber (*Cucumis sativus*): in one experiment, a plot of the U.S.A. slic9ing cultivar Poinsett 76 was grown under simulated commercial conditions in the field at Newe Ya'ar (northern Israel) in the summer of 1998. Each plant was allowed to set five fruits, at 2-3 day intervals between each fruit. Each fruit had been tagged on the day of anthesis and was harvested 42 days later. Each fruit was weighed at harvest and its seeds extracted and dried. Four samples of 25 seeds each were then weighed. There were four replicates of 10 plants each. Another experiment was conducted the subsequent winter in a greenhouse at Shefeye (northern Israel). Three cultivars differing in fruit size were grown" 'Jinchun No. 4', a hybrid from china having fruits 40-50 cm long; 'Poinset 76' a slicing-type from the U.S.A. having fruits ~25 cm long; and 'Triple Mech', a pickling-type from the U.S.A. having fruits ~15 cm long. Mature fruits were harvested 40-50 days past anthesis and weighed. Dry seeds were also weighed. There were four replicates of ten plants each.

Melon (*Cucumis melo*): One experiment, conducted in the field at Newe Ya'ar under commercial conditions in the summer of 1997, had two muskmelon cultivars, 'Noy Yizre'el' from Israel and 'Top Mark' from the U.S.A., grown in four replicates of 20 plants each. At maturation, the fruits and seeds were weighed. The second experiment on muskmelon was conducted the following year and next to the cucumber plot; it was conducted in the same fashion as the first experiment with cucumber, using 'Noy Yizre'el'.

Watermelon (*Citrullus lanatus*): 'Malai', an Israeli cultivar, and two breeding lines, nos. 203 and 239-4, were grown in a commercial dryland field for production of edible seeds near Daverat (northern Israel) in the summer of 1998. At harvest, the fruits were divided into five size group (<500 g, 500-1000 g, 1000-1500 g, 1500-2000 g and >2000 g) and the seed yield and yield components were observed for each group separately. There were four replicates for each accession, and the area of each replicate was 20 m^2.

Pumpkin/Gourd (*Cucurbita pepo*): At the Newe Ya'ar research Center in summer 1998, 15-20 plants of each two cultivars were grown: 'Tondo Schro di Piacenza', a pumpkin from Italy, and 'Flat Striped', an ornamental gourd from Canada. These two differ markedly (Approximately 7-fold) in fruit size. Ten mature fruits were harvested from each and weighted, and their dried seeds were weighted.

Results and discussion. Fruit order had a significant effect on fruit weight in 'Noy Yizre'el melon and 'Poinsett 76' cucumber (Table 1). However, the two species differed in the relationship between fruit and mean seed weight. In 'Noy Yizre'el' melon, there was a linear relation between these two variables while in 'Poinsett 76 cucumber these two were not correlated. A comparison among cultivars of these species enhanced this conclusion. The seed size of the two melon cultivars differed significantly, as did their fruit size. However, the seed size was the same in cucumbers, even though the three cultivars differed markedly in fruit size (Table 2).

The relationship between fruit size and seed size in 'Malali' watermelon and two related breeding lines resembled those of cucumber. There were no significant size differences in seeds extracted from large fruits or small fruits down to 500 g (Table 3). Only extremely small fruits (<500 g) produced smaller seeds. the three watermelon secessions differed significantly in mean fruit weight (Table 4) but nevertheless had the same mean seed weight.

The relationship between fruit size and seed size in *Cucurbita pepo was* nearly linear (Table 5). Thus, the relationship was more similar to that of melon than it was to cucumber or watermelon.

The results presented here are but a small part of a large unpublished investigation of the possible relationships between fruit and seed yield of cucurbits. These selected data show that, in cucurbits, two different relationships between fruit and seed size have developed over the course of evolution. Seemingly, cucumber and watermelon went in one path whilst melon and pumpkin/squash went in another.

Table 1. Effects of fruit order on fruit weight and individual seed weight in cucumber 'Poinsett '76' and melon 'Noy Yizre'el.

	Cucumber		Melon	
Fruit order	**Fruit weight (g)**	**Seed weight (mg)**	**Fruit weight (g)**	**Seed weight (mg)**
1	774 a	23.4 a	1647 a	37.4 a
2	719 a	23.8 a	1553 a	31.4 b
3	636b	21.4 ab	1216 b	25.4 c
4	636 b	20.5 b	981 c	2.9 c
5	558 c	22.1 ab	730 d	19.5 d

Table 2. Fruit weight and individual seed weight in different cultivars of cucumber (greenhouse) and melon (field).

Cucumber			Melon		
Cultivar	**Fruit wt. (g)**	**Seed weight (mg)**	**Cultivar**	**Fruit wt. (g)**	**Seed weight (mg)**
Jinchun No. 4	1121 a	28.2 a	Noy Yizre'el	1065 a	32.6 a
Poinsett 76	580 b	27.8 a	Top Mark	839 b	22.7 b
Triple Mech	420 c	28.3 a			

Table 3. Effect of fruit weight (g) on mean seed weight (mg) in 'Malali; and two breeding lines, 203 and 239-4, of watermelon.

Fruit weight	**Malali**	**203**	**239-4**
<500	-	149 b	145 b
500-1000	161 a	166 ab	161 a
1000-1500	164 a	170 ab	165 a
1500-2000	170 a	161 b	160 a
>2000	165 a	177 a	---

Table 4. Mean fruit weight and mean seed weight of 'Malali' and two breeding lines, 203 nd 239-4, of watermelon.

Cultivar/breeding line	**Fruit weight (kg)**	**Seed weight (mg)**
Malali	2.49 a	166 a
203	0.98 b	169 a
239-4	0.98 b	163 a

Table 5. Effect of fruit weight (g) on mean seed weight (mg) in an ornamental gourd and a pumpkin (both *Cucurbita pepo*)

Gourd		Pumpkin	
Fruit weight	**Seed weight**	**Fruit weight**	**Seed weight**
244	72	1720	---*
225	61	1670	145
1963	45	1560	155
191	47	1520	148
188	41	1465	128
175	42	1465	120
160	41	1430	110
160	36	1080	95
160	32	1035	118
153	30	1000	93

*no data

Acknowledgement: contribution No. 121/00 from the Institute of Field & Garden Crops, Agricultural REsearch Organization, Bet Dagan, Israel.

Literature Cited

1. DeMarco, D.G. 1990. Effect of seed weight and seed phosphorus and nitrogen concentrations on the early growth of wheat seedlings. Aust. J. Exper. Agri. 30:545-549.
2. Edelstein, M., H. Nerson, H.S. Paris, Z. Karchi, and Y. Burger. 1987. Is there any importance to seed size for growth of spaghetti squash? Hassadeh 67:688-689.
3. Marcelis, L.F.M. and L.R.B. Hofman-Eijar. 1997. Effects of seed number on competition and dominance among fruits of *Capsicum annum* L. Ann. Bot. 79: 687-693.
4. Naylor, R.E.L. 1993. The effect of parent plant nutrition on seed size, viability, and vigour and on germination of wheat and triticale at different temperatures. Ann. Appl. Biol. 123:379-390.
5. Sanches, V.M., F.J. Sundstrom, and N.S. Lang. 1993. Plant size influences bell pepper seed quality and yield. HortScience 28: 809-811.
6. Stephenson, A.G., B. Devlin, and J.B. Horton. 1988. The effects of seed number and prior fruit dominance on the pattern of fruit production in *cucurbita pepo* (zucchini squash). Ann. Bot. 62: 653-61.

Cucurbit Genetics Cooperative Report 23:68-68 (article 22) 2000

A Strain of Watermelon Mosaic Virus from Massachusetts Causes Prominent Symptoms on Squashes and Systematically Infects *Cucurbita equadorensis* and *C. maxima* PI 419081-1

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Watermelon mosaic virus (WMV), previously known as WMV-2, commonly occurs in cucurbits and wild legume species growing in the northeastern United States. Generally, the symptoms caused by this virus in cucurbits are less severe than those incited by other cucurbit viruses. However, they may vary with the species and viral strain involved. Symptoms include light and dark green mosaic, green veinbanding, chlorotic spots and some leaf rugosity. Infected plants are slightly stunted and fruits are not distorted, but some colons may be adversely affected (Provvidenti, 1986 and 1993.)

In July, 1999, Dr. Clark W. Nicklow (Ashland, Massachusetts) brought to our attention the occurrence of a widespread viral disease in pumpkins (*Cucurbita pepo* L) in a four acre field near Hudson, MA. Although plants appeared of quasi normal size, foliar light and dark green mosaic was rather prominent and fruit setting was considerably reduced. On July 14, samples of several infected plants were received directly from Mr. Manouel Ferjulian, the owner of the affected field. the foliar symptoms displayed by infected plants resembled those usually incited by the cucurbit strain of papaya ringspot virus (PRSV-W). However, using differential hosts, including bean plants of Black Turtle 2 (BT-2), and serology, the causal agent was identified as a strain of WMV. This strain henceforth is designated as WMV-MA.

In the greenhouse, BT-2 bean plants infected with WMV-AA were used as sources of inocula to test the following squashes: *Cucurbita pepo* 'Seneca Zucchini', 'Dark Green Zucchini', 'Butterbar', 'Table Green', and 'Delicata'; *Cucurbita maxima*: 'Zapalito Rotundo'; and *Cucurbita moschata*: 'Butternut'. All the plants of these cultivars developed prominent systemic foliar symptoms. Plants *C. moschata* "Nigerian Local' were systemically resistant, whereas those of *C. maxima* PI 419081-1 (China) and *cucurbita equadorensis* were systemically infected. Consequently, this strain of WMV differs from the others that we have found in the Northeast, since it is able to overcome the resistance to WMV in *C. eqiuadorensis* (Provvidenti et al, 1978) and *C. maxima* PI 419081-01 (China) (Provvidenti, 1982). However, in these two WMV-resistant species, WMV-Ma causes moderate foliar symptoms and limited plant stunting.

Literature Cited

1. Provvidenti, 1982. Sources of resistance and tolerance to viruses in accessions of *Cucurbita maxima.* Cucurbit Genet Coop. Rept. 5:46-47.
2. Provvidenti, R. 1993. Resistance to viral diseases of cucurbita. In: M.M. Kyle, ed. Resistance to viral diseases of vegetables. Timber Press, Protland, Oregon, pp. 8-43.
3. Provvident, R., R.W. Robinson and H.M. Munger. 1978. Resistance in feral species to six viruses infecting *Cucurbita.* Plant Dis. Rep. 62:326-329.

Cucurbit Genetics Cooperative Report 23:69-70 (article230) 2000

Searching for Molecular Markers Linked to ZYMV Resistance in Squash

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The purpose of this study was to find a molecular marker linked to zucchini yellow mosaic virus resistance from *Cucurbita moschata* 'Nigerian Local'. 'Nigerian Local' is a major source of virus resistance for both *Cucurbita pepo* and *C., moschata.* A linked marker could be very useful to breeders in that it would permit selection for virus resistance without actually inoculating plants with ZYMV. Heredity studies have determined that ZYMV resistance in C. moschata is primarily controlled by a single dominant gene (4,5).

We used two BC1 populations for this study: an interspecific population from the cross between a Sunseeds zucchini inbred and 'Nigerian Local', and an intraspecific cross between 'Waltham Butternut' and 'Nigerian Local'. In both cases the backcross was to the susceptible parent. the 'Waltham Butternut' x 'Nigerian Local' population fit a 1:1 segregation ratio as expected. The zucchini x 'Nigerian Local' fit a 1:1 ratio at the first scoring ten days after inoculation with ZYMV, but over the next several weeks most of the "resistant" plants developed virus symptoms of varying severity. These plants were classed as tolerant.

The marker search was conducted using bulk segregant analysis with both RAPDs and AFLPs. Primers were first screened on the parental lines; those which were polymorphic were then screened on the bulks., Markers which produced bands in the resistant bulks and in 'Nigerian Local' were screened on the individuals which comprised the bulks. For the 'Nigerian Local' x 'Waltham Butternut' population there were two resistant bulks and two susceptible bulks. the zucchini x 'Nigerian Local' population was represented by a resistant bulk, two tolerant bulks, and a susceptible bulk. DNA extractions were done using a previously published protocol (2). RAPDs were screened as described elsewhere (1). The AFLP protocol was one originally designed for meadowfoam (3); the restriction enzymes used were EcoRI and MseI.

Results: The 'Waltham Butternut' x 'Nigerian Local' population was screened with 943 RAPD primers, which yielded 4381 scoreable bands. Fourteen percent of the bands were polymorphic between 'Nigerian Local' and 'Waltham Butternut', with 43.3% of the primers giving at least one polymorphic band. This population was also screened with 14 AFLP primer pairs, which yielded 803 scoreable bands. All of the primer pairs gave at least one band which was polymorphic between the parental lines, with an average of 9.4 polymorphic bands per primer pair. Overall, 16.4% of the AFLP bands were polymorphic between the two parents.

The zucchini x 'Nigerian Local' population was screened with 220 RAPD primers. They yielded 1008 scoreable bands, 45% of which were polymorphic between 'Nigerian Local; and the zucchini. Fifty-five percent of the primers gave at least one polymorphic band. This population was not screened with AFLPs.

None of the RAPD primers or AFLP primer pairs amplified bands which were reliably linked to ZYMV resistance or tolerance. The levels of polymorphism between our parents would seem to be sufficient to identify markers linked to resistance. We are still unsure as to exactly why we were unable to find a marker linked to ZYMV resistance. Other efforts to find RAPD markers linked to introgressed disease resistance genes have produced similar results, including an attempt to find markers linked to ZYMV resistance from *Cucurbita ecuadorensis* introgressed into *Cucurbita maxima* (N. Weeden, personal communication). One possibility is that the gene for resistance is located near the telomere of a chromosome; such loci are known to be difficult to tag with RAPD markers (S. Knapp, personal communication). Another possibility is that the resistant and susceptible alleles are very similar in sequence, and thus could not be reliably differentiated by RAPDs, which only have an accuracy of 80-90% (6). The AFLP protocol we used was not optimized for *Cucurbita*. It is possible that further research might reveal AFLP markers linked to ZYMV resistance. In particular, MseI may not be the best restriction enzyme to use with *Cucurbita.*

We are currently working on a RAPD-based skeleton map of *cucurbita* using a yellow squash x 'Nigerian Local' BC1 population. One of the traits being mapped is ZYMV resistance. Preliminary data from the mapping population suggests that using ELISA as well as visual symptoms to differentiate resistant and susceptible plants may be key to finding a marker linked to ZYMV resistance. If the resistance gene is telomeric, it may be that other marker technologies such as simple sequence repeats (SSRs) may be more appropriate than RAPDs.

Literature Cited

1. Brown, R.N., A.B. Herrera, J.R. Myers, and M.K. Jahn. 2000. The Inheritance of Resistance to Four Cucurbit Viruses in *Cucurbita moschata* (Duch. ex Poir.) and the Search for Molecular Markers LInked to Resistance. ASHS Journal (in press).
2. Brown, R.N., J.R. Myers, M. Hutton, and P. Miller 1998. A simple protocol for isolating DNA from fresh *Cucurbita* leaves. Cucurbit Genetics Coop. Rpt. 21:46-47.
3. Karengam, S. 1999. PhD Thesis. Dept. of Crop and Soil Science, Oregon State University, Corvallis, Oregon.
4. Munger, H.M., and R. Provvidenti. 1987/ Inheritance of resistance to zucchini yellow mosaic virus in *Cucurbita moschata.* Cucurbit Genetics Coop. Rpt. 10:80-81.
5. Paris, H.S., S. Cohen, Y. Burger, and R. Yoseph. 1988. Single-gene resistance to zucchini yellow mosaic virus in *Cucurbita moschata.* Euphytica 37:27-29.
6. Weising, K., H. Nybom, K. Wolff, and W. Meyer. 1995. DNA Fingerprinting in Plants and Fungi. CRC Press, Boca-Raton, FL.

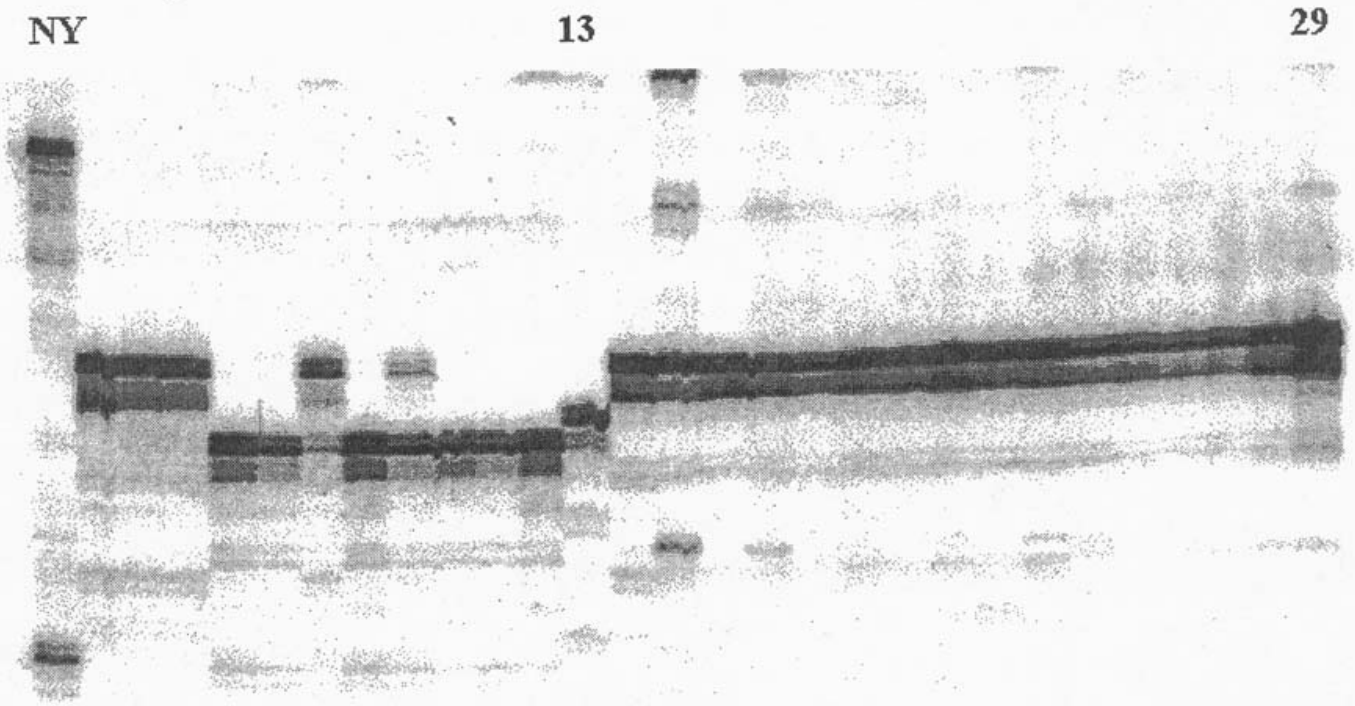

Figure 1. Amplification of *C. pepo* DNA using *Cucumis melo* CMAG59 SSR. *C. melo* 'Noy Yizre'el' appears on the left side of the autoradiogram, followed by *C. pepo*, left to right: 'Ebony', 'Table Queen', 'Royal Acorn', 'Golden Bush Scallop', 'Yellow Bush Scallop', 'Golden Girl', 'Early Summer Crookneck', 'Striped Pear', 'Wild Arkansas', 'Wild Texas', 'Benning's Green Tint', 'Wild Mexico', 'Cocozelle Tripolis', 'Long Cocozelle', 'Striato d'Italia', 'Verte Non-coureuse d'Italie', 'Lunga di Toscana', 'Tondo di Nizza', 'Connecticut Field', 'Small Sugar', 'Beirut', 'Porqueira', 'Yugoslavia 7', 'Verte Petite d'Alger', 'Vegetable Spaghetti', 'Fordhook Zucchini', 'Nero di Milano', and 'Orange Ball'. Seed sources for each of these accessions are presented in reference 4.

Cucurbit Genetics Cooperative Report 23:73-77 (article 25) 2000

Genetic Diversity Within and Between the Species *Cucurbita pepo, C. moschata* and *C. maxima* as Revealed by RAPD Markers

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Introduction. Genes not available in a species can often be obtained through interspecific hybridization. A good example is resistance against Zucchini Yellow Mosaic Virus (ZYMV), which is not found in *C. pepo* (6), but is available in *C. moschata* (3, 4, 2). Crossing the two species is difficult, though not impossible. Hybrids are often sterile or only set a few viable seeds (1). the reason for this is often the large genetic distance between the species.

Molecular markers provide an excellent means to quantify genetic differences between, but also within, the species. Molecular markers are neutral and independent of the genotype. In this study we used RAPD markers to estimate genetic diversity within and between the species*Cucurbita pepo, C. Maxima* and *C. Moschata.*

Materials and Methods. *Plant material*: Six *C. pepo* genotypes, three from each of two Austrian Breeding Companies, Saatzucht Gleisdorf (*pep1, pep2, pep3*) and VitroPlant (*pep4, pep5, pep6*), six *C. maxima* genotypes of different geographic origin (*max1*USA, *max2* China, *max3* Japan, *max4* Hungary, *max5* France, *max6* Mongolia) and six *C. moschata* genotypes (*mos1*"Nigerian Local" from Nigeria, *mos2* "Nicklow's Delight" from USA, *mos3* "Menina" from Portugal, *mos4, mos5,* and *mos6* from Puerto Rico) were selected for this study.

DNA isolation: DNA isolation was carried out using the QIAGEN Dneasy Plant Mini Kit (*http://www.qiagen.com*, Cat. Nr. 69103). *Primers:*26 10mer RAPD primers were used supplied by ABgene (*http://abgene.com*), and ROTH (*http://www.Carl-Roth.de*). *PCR conditions:*100 ng of genomoc DNA were used in 25 µl-volume amplification reactions containing 0.3 µM 10mer random primer, 1x reactions bugger, 1.5 mM $MgCl_2$, 200 µM dNTP and 1 Unit Taq polmerase. For amplification we used a TouchDown Thermocycler (Hybaid, *http://www.hybaid.co.uk*). *Temperature program:*Initial denaturation of 60 seconds at 94°C followed by 34 cycles of 60 seconds at 94°C, 45 seconds at 36°C, 30 seconds at 72°C, finished with a final extension step of 5 minutes at 72°. *Fragment separation:* Fragments were separated in 1.5% agarose gels stained with ethidium bromide and photographed with a Polaroid camera. *Data analysis*: Data were recoded to a present-absent scale (1/0) and then subjected to an UPGMA cluster analysis.

Results and Discussion. RAPDs are dominant markers. Polymorphism is indicated by the presence or absence of a fragment (Fig. 1). Only sharply delineated clear bands were accepted as marker loci. Altogether 379 such loci were recorded. From the total number of loci, ten were monomorphic throughout the three species. Monomorpic loci present in two species, i.e. in *C. pepo* and *C. maxima,* in *C. pepo* and *C. moschata*, and in *C. maxima* and *C. moschata* were 13, 12 and 6 respectively (Fig. 1).Number of evaluated and polymorphic loci in the three species is given in Table 1. Pairwise distances between the 18 genotypes were calculated (Table 2) and a cluster analysis was carried out. (Fig. 2).

The three species show almost equal distance to each other (Table 2). distance within the species is on average less than one fourth of the distance between the species. Comparing the three species clearly the least polymorphic one is *C. pepo* represented by six Austrian hull-less seeded oil pumpkin genotypes. This is shown by the lowest number of polymorphic loci (Table 1) and by the lowest average distance value (Table 2). This is most probably due to the narrow geographic distribution of the genotypes and a certain drift by selecting for the last hundred years the hull-less seeded types. In an earlier study (5) we detected 3,4 polymorphism per RAPD primer in 20 inbred lines of Styrian oil-pumpkin using 34 primers.

The clearly lower number found in this study (2.4) is probably due to the lower number of genotypes and markers and the fact that the genotypes of the present study were already selected for Austrian growing conditions.

The largest within species distance is exhibited by the six *C. moschata* genotypes. This group has the highest number of polymorphic loci, the highest number of markers per primer (Table 1), and the highest average distance value (Table 2). This seems to be due to their geographic origin. The three genotypes from Puerto Rico (kindly made available by Dr. Linda Wessel-Beaver) are obviously closely related like the oil pumpkin genotypes from Austria. "Nigerian Local" is a "runaway", a comparable difference can not be seen in the group of *C. maxima* with the widest geographic distribution. The large distance of "Nigerian Local" to "Menina" (67 in Table 2) is of special interest, because these genotypes are the two known sources of resistance genes against ZYMV in pumpkin. Because of the

large distance between the two genotypes it is possible that these resistance genes are different and therefore suitable for pyramiding in *C. pepo.*

Table 1. Number of evaluated and polymorphic loci in three *Cucurbita* species.

	C. pepo	***C.maxima***	***C. moschata***
Evaluated loci	146	152	170
Polymorphic loci	62	85	109
% Polymorphic loci	42.5	55.9	64.1
Polymorphic loci per primer	2.4	3.3	4.2

Table 2. Squared Euclidean dissimilarity between 18 *Cucurbita* genotypes based on binary data of individual RAPD loci.

	pep1	**pep2**	**pep3**	**pep4**	**pep5**	**pep6**	**max1**	**max2**	**max3**	**max4**	**max5**	**max6**	**mos1**	**mos2**	**mos3**	**mos4**	**mos5**
pep2	30																
pep3	30	20									**Average:**						
pep4	33	25	33								pep: 29						
pep5	34	28	30	31							max: 39						
pep6	33	19	33	28	21						mos: 48						
max1	167	173	173	178	171	162					pep/mos: 184						
max2	178	184	184	189	182	173	29				pep/max: 176						
max3	172	178	178	183	174	165	41	32			mos/max: 182						
max4	171	177	177	182	173	164	46	41	39								
max5	172	178	178	183	174	165	47	36	38	34							
max6	176	184	182	189	180	171	45	38	40	43	34						
mos1	182	186	184	187	188	183	169	172	168	173	172	172					
mos2	183	183	183	188	187	182	182	189	181	188	189	187	67				
mos3	179	177	177	184	179	176	174	183	175	180	179	179	67	36			
mos4	184	184	182	189	186	183	179	188	180	185	184	186	76	45	41		
mos5	184	184	184	189	188	185	183	190	184	189	188	192	78	43	39	20	
mos6	183	183	181	188	187	182	182	191	183	188	189	189	75	44	38	23	23

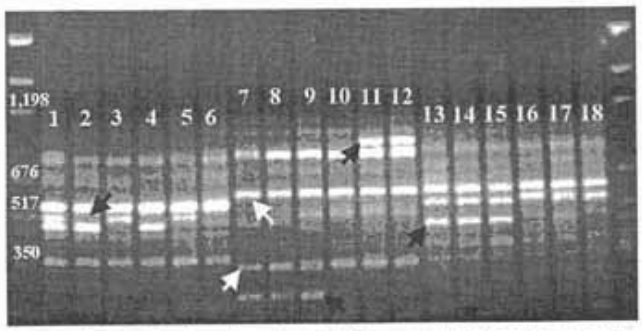

Fig. 1: Example for RAPD polymorphism, detected in 1.5% agarose gel, within and between species. Lane 1-6 represent the six *C. pepo*, 7-12, *C. maxima* and 13-18, *C. moschata* genotypes. Black arrows indicate within species, white arrows between species polymorphism. D-15 Pharmacia size marker was used. Most bands appeared between bp 1,198 and 222.

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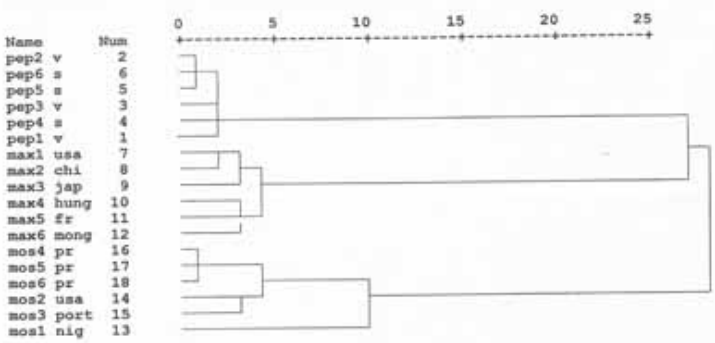

Figure 2. Dendrogram of 18 *Cucurbita* genotypes using the UPGMA clustering algorithm. Scaled distance at which clusters combine.

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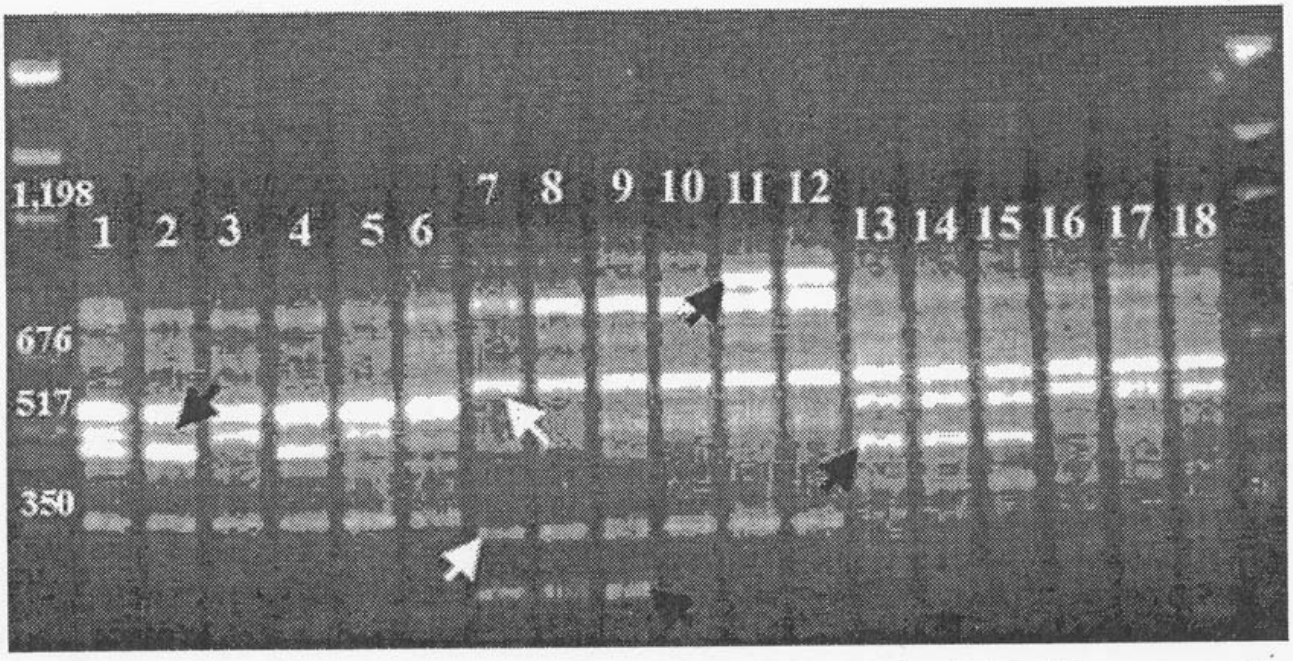
Literature Cited.

1. Baggett, J.R., 1979. Attempts to cross Cucurbita moschata (Duch.) Poir. 'Butternut' and C. pepo L. 'Delicata'. Cucurbit Genetics Coop. Rpt. 2:32-34.
2. Gilbert-Albertini, F., H. Lecoq, M. Pitrat, and J.L. Nicolet, 1993. Resistance of *Cucurbita moschata* to watermelon mosaic virus type 2 and its genetic relation to resistance to zucchini yellow mosaic virus. Euphytica 69:231-237.
3. Munger, H.M., and R. Provvidenti, 1987: Inheritance of resistance to zucchini yellow mosaic virus in *Cucurbita moschata.* Cucurbit Genetics Coop. Rpt. 10:80-81.
4. Paris, H.S., S. Cohen, Y. Burger, and R. Yoseph, 1988: Single-gene resistance to zucchini yellow mosaic virus in *Cucurbita moschata.* Euphytica 37:27-29.
5. Stachel, M., Gy. Csanadi, J. Vollmann, and T. Lelley, 1998: Genetic diversity in pumpkins (*Cucurbita pepo* L.) as revealed in inbred lines using RAPD markers. Cucurbit Genetics Coop. Rpt. 21:48-50.
6. Whitaker, T.W., and R.W. Robinson, 1986.: Squash breeding. In: Breeding vegetable crops. Ed.: M.J. Bassett. Avi, Westport, Ct.



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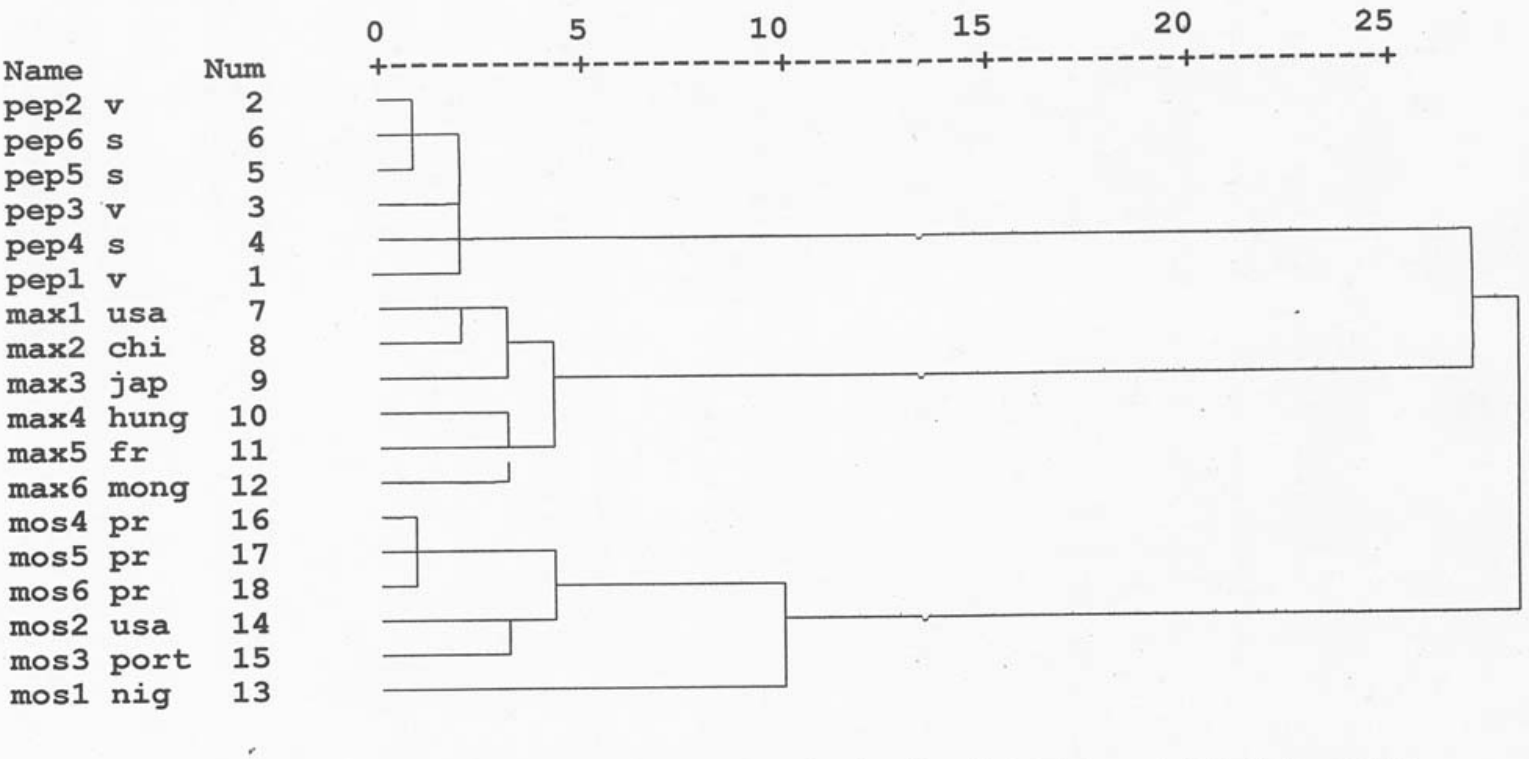

Figure 2. Dendrogram of 18 *Cucurbita* genotypes using the UPGMA clustering algorithm. Scaled distance at which clusters combine.

Cucurbit Genetics Cooperative Report 23:78-79 (article260) 2000

Genetic Variability in Bottlegourd, *Langenaria siceraria* (Molina) Standley

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India is one of the centres of diversity of bottlegourd (1), endowed with a variety of diverse germplasm. Hence a collection of bottlegourd from different parts of India was made and evaluated for their qualitative and quantitative characters. Twenty-eight such accessions were raised in an experimental plot using randomised block design with two replications.

Among various qualitative characters studied fruit shape and fruit colour exhibited high variation (Fig. 1). Fruit shape ranged from pyriform, dumb bell, curved, crooked neck to elongate forms and fruit color varied from light green to dark green with or without patches. Seed color also recorded variation from tan to dark brown.

Significant difference was observed in accessions for quantitative characters, viz. Vine length, number of primary branches, days to first female flower opening, nodes to first female flower opening, nodes to first female flower, sex ratio, number of fruits per plant, length of fruit, girth of fruit, 100 seed weight, number of seeds per fruit and crude fibre content. Existence of variability in quantitative characters in bottlegourd was also reported by earlier workers (2,3).

Range, phenotypic coefficient of variation (pcv) and genotypic coefficient of variation (gcv) for different characters are given in Table 1. Maximum range of variation was observed for number of seeds per fruit (250.25 - 821.75), followed by fruit set percent (20.00% - 70.,00%). The highest gcvs and pcvs were recorded for number of fruits per plant (38.05 and 50.71) and the lowest for internodal length (0.22 and 6.95). The gcv values were close to the pcv value for the following characters: vine length, number of primary branches, days to first female flower opening, nodes to first female flower, days to first harvest, number of fruits per plant, length of fruit, girth of fruit, 100 seed weight, number of seeds per fruit and crude fibre content.

The study shows that there exists a potential source of gene sanctuary for hottlegourd in the Indian sub continent, which can be effectively harnessed for selection of elite types.

Table 1. Range, mean, phenotypic coefficient of variation and genotype coefficient of variation of different characteristics of bottlegourd.

Characters	Range	Mean + se	14.73 + 1.27pcv	gcv
Vine length (m)	7.97 - 18.02	12.11 ± 1.32	23.45	20.76
Number of primary branches	5.25 - 9.75	7.66 ± 0.41	16.61	15.75
Internode length (cm)	13.25 - 15.66	14.40 + 1.00	6.95	0.22
Days to first female flower opening	39.00 - 54.75	46.30 ± 3.58	12.33	9.60
Nodes to first female flower	9.00 - 21.50	15.53 ± 2.03	23.06	18.99
Sex ratio	4.68 - 12.03	7.56 ± 1.62	29.10	19.72
Days to first harvest	52.25 - 68.50	61.98 ± 5.19	9.89	5.25
Fruit set (%)	20.00 - 70.00	40.18 ± 13.83	40.38	21.11
Number of fruits/plants	2.25 - 11.50	4.11 ± 1.38	50.71	38.05
Fruit yield/plant (kg)	2.91 - 13.37	7.47 ± 2.47	40.47	23.34
Average fruit weight (kg)	0.86 - 2.65	1.79 ± 0.44	29.21	15.87
Length of fruit (cm)	28.80 - 72.30	50.98 ± 5.27	25.31	23.10
Girth of fruit (cm)	25.50 - 51.50	34.86 ± 1.87	19.21	18.44
100 seed weight (g)	11.90-20.10	16.01 ± 1.27	13.16	10.51
Number of seeds/fruit	250.25 - 821.75	484.88 ± 40.48	28.15	26.88
Duration of crop	82.25 - 112.50	95.97 ± 7.53	8.77	3.91
Crude fibre content (%)	12.50 - 18.50	14.73 ± 1.27	13.52	10.40

Table 1. Range, mean, phenotypic coefficient of variation and genotypic coefficient of variation of different characteristics of bottlegourd.

Characters	Range	Mean±se	pcv	gcv
Vine length (m)	7.97 – 18.02	12.11 ± 1.32	23.45	20.76
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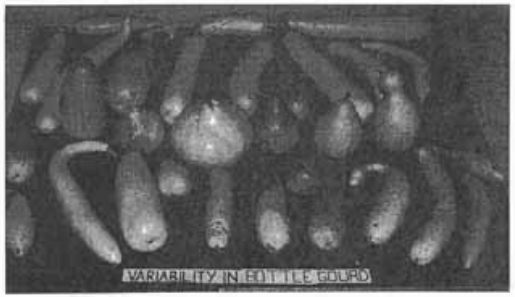

Figure 1. Variability in fruit shape of bottle gourd (*Lagenaria siceraria*).

Literature Cited

1. De Candolle, A. 1882. Origin of Cultivated Plants. Cited by Chadha, M.L. and T. Lal, 1993. Improvement of cucurbits, Adv. Horticulture. (Ed. Chadha, K.L. and G. Kalloo) Malhotra publishing house, New Delhi, p. 151-155.
2. Singh, S.P., N. K. Singh, I.B. Maurya. 1996. Genetic variability and correlation studies in bottlegourd. [Lagenaria siceraria (Mol,) Standl]. PKV Res. J. 20:88-89.
3. Tyagi,I.D. 1972. Variability and correlation studies in bottlegourd. Indian J. Horticulture. 29: 219-222.

Cucurbit Genetics Cooperative Report 23:78-79 (article260) 2000

Genetic Variability in Bottlegourd, *Langenaria siceraria* (Molina) Standley

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India is one of the centres of diversity of bottlegourd (1), endowed with a variety of diverse germplasm. Hence a collection of bottlegourd from different parts of India was made and evaluated for their qualitative and quantitative characters. Twenty-eight such accessions were raised in an experimental plot using randomised block design with two replications.

Among various qualitative characters studied fruit shape and fruit colour exhibited high variation (Fig. 1). Fruit shape ranged from pyriform, dumb bell, curved, crooked neck to elongate forms and fruit color varied from light green to dark green with or without patches. Seed color also recorded variation from tan to dark brown.

Significant difference was observed in accessions for quantitative characters, viz. Vine length, number of primary branches, days to first female flower opening, nodes to first female flower opening, nodes to first female flower, sex ratio, number of fruits per plant, length of fruit, girth of fruit, 100 seed weight, number of seeds per fruit and crude fibre content. Existence of variability in quantitative characters in bottlegourd was also reported by earlier workers (2,3).

Range, phenotypic coefficient of variation (pcv) and genotypic coefficient of variation (gcv) for different characters are given in Table 1. Maximum range of variation was observed for number of seeds per fruit (250.25 - 821.75), followed by fruit set percent (20.00% - 70.,00%). The highest gcvs and pcvs were recorded for number of fruits per plant (38.05 and 50.71) and the lowest for internodal length (0.22 and 6.95). The gcv values were close to the pcv value for the following characters: vine length, number of primary branches, days to first female flower opening, nodes to first female flower, days to first harvest, number of fruits per plant, length of fruit, girth of fruit, 100 seed weight, number of seeds per fruit and crude fibre content.

The study shows that there exists a potential source of gene sanctuary for hottlegourd in the Indian sub continent, which can be effectively harnessed for selection of elite types.

Table 1. Range, mean, phenotypic coefficient of variation and genotype coefficient of variation of different characteristics of bottlegourd.

Characters	Range	Mean + se	14.73 + 1.27pcv	gcv
Vine length (m)	7.97 - 18.02	12.11 ± 1.32	23.45	20.76
Number of primary branches	5.25 - 9.75	7.66 ± 0.41	16.61	15.75
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Number of seeds/fruit	250.25 - 821.75	484.88 ± 40.48	28.15	26.88
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Crude fibre content (%)	12.50 - 18.50	14.73 ± 1.27	13.52	10.40

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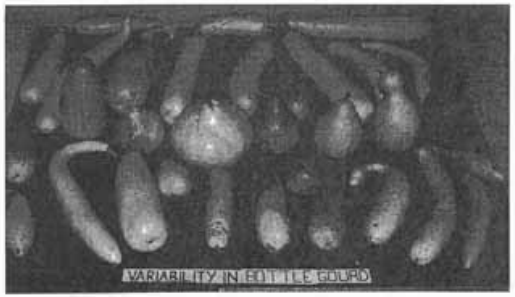

Figure 1. Variability in fruit shape of bottle gourd (*Lagenaria siceraria*).

Literature Cited

1. De Candolle, A. 1882. Origin of Cultivated Plants. Cited by Chadha, M.L. and T. Lal, 1993. Improvement of cucurbits, Adv. Horticulture. (Ed. Chadha, K.L. and G. Kalloo) Malhotra publishing house, New Delhi, p. 151-155.
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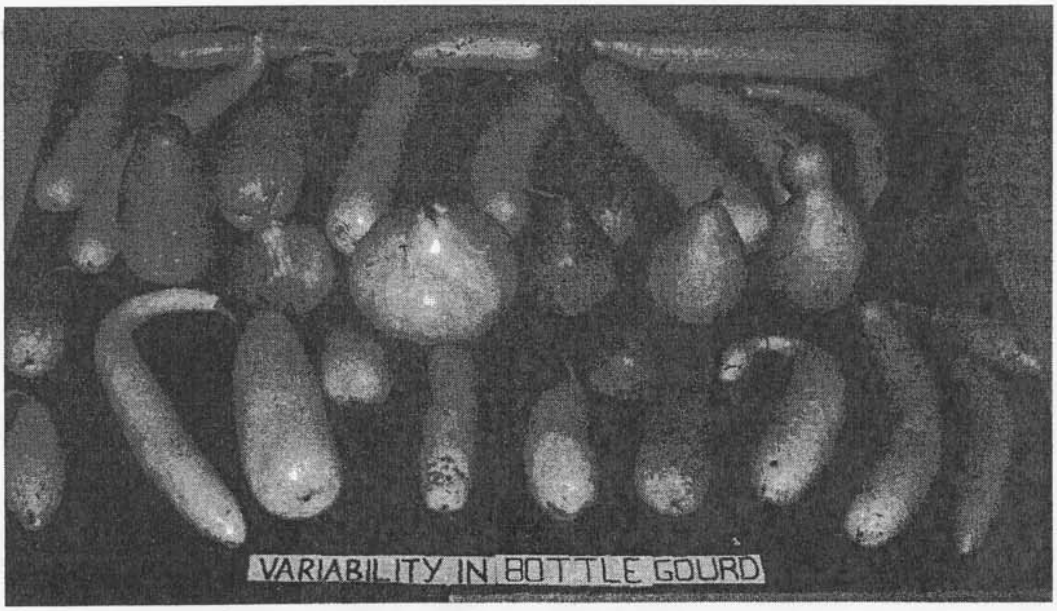

Figure 1. Variability in fruit shape of bottle gourd (*Lagenaria siceraria*).

Cucurbit Genetics Cooperative Report 23:80-82 (article 27) 2000

Bud Induction of Serpent Gourd (*Trichosanthes anguina* L.) In Vitro

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Introduction. Serpent gourd belongs to Cucurbitaceae and originates in the tropic area of Asia. The cultivation of serpent gourd occurs mainly in India and other countries of southeastern Asia. In china, little serpent gourd has been grown though, cultivation of this plant has been increasing. Serpent gourd is nutritional and has high value as a vegetable. there are only a few seeds in each fruit and the propagation coefficient is very low. The cost and shortage of seeds has been the limitation to the enlargement of serpent gourd production. Therefore, development of a fast propagation system is a significant aid to increasing the cultivation of this vegetable.

Material and Methods. The healthy seeds of the serpent gourd cultivar endemic to the Xi'an area were scalded at 60 C for 10 min and soaked at 20 C for 48 hr. The seeds having broken seed coats germinated at 27-28 C and 2 to 3 days later, planted into blocks filled with sterilized compost. The blocks were placed in a room at 27$\pm$ 1 C, and 12 to 14 hr of 1500 to 2000 Lux of light.

Apex buds and axillary buds of seedlings were used as explants. The explants were cut into pieces 0.5 to 0.8 cm long and sterilized with 70% alcohol for 30 seconds, rinsed 3 time with sterilized water, then sterilized with 0.1% $HgHCl_2$ for 3 min and rinsed 3 times again. The explants were planted on media following sterilization.

MS medium was used as the basal medium and supplemented with 2.5% sucrose and 0.6% agar.

Different concentrations of TDZ, KT, ZT, 6-BA, and various combinations of 6-BA with 2,3-D or IAA were supplemented to media for bud induction.

Results and Discussion. *Effects of cytokinins on bud induction:* Apex buds of 4-day-old seedlings were used as explants. Explants planted on the media supplemented with TDZ tended to produce callus. There was no bud initiation on the media supplemented with KT (0.1 or 2.0 mg/L) or TDZ (0.2 or 2.0 mg/L) or ZT at lower concentrations (0.05 or 0.2 mg/L). KT and ZT at lower concentrations only promoted explant elongation and root initiation, with the effect of KT greater than of ZT. Buds were initiated by ZT at higher concentrations (0.05 or 2.0 mg/L. However, the explants tended to elongate and the quality of initiated buds was poor. Buds were initiated at all concentrations of 6-BA tested (Table 1). The optimal 6-BA concentration was found to be 1 mg/L.

*Effects of combinations of 6-BA with 2,4,-D or IAA:*Apex buds and axillary buds of 3-week-old seedlings were used as explants. Buds were initiated in all treatments combining 6-BA and IAA. Similar bud initiation effects were found on the media supplemented with only 6-BA. However, greater IAA concentrations resulted in longer bud length (Table 2).

The explants tended to produce callus but no buds on media supplemented with 6-BA and 2,4-D at higher concentrations (0.1 or 0.2 mg/L) (Table 3). when lower concentrations of 2,3-D (0.05 or 0.02 mg/L) were combined with 6-BA, buds were induced. Differences in bud induction were also observed between explant sources. The apex and bud explants initiated buds only at the lowest concentration (0.02 mg/L) of 2,4-D in combination with either concentration of 6-BA. The axillary bud explants initiated buds at 2,4-D concentrations of 0.02 mg/L or 0.05 mg/L combined with either 6-BA concentration. Although buds were induced with combinations of 2,4-D and 6-BA, they were of poorer quality than those induced on the media supplemented with only 6-BA (Table 3).

Table 1. Effects of 6-BA on bud induction of Serpent gourd.

Concentration of 6-BA (mg/L)	Number of buds per explant	Bud length (cm)
0.5	4.3	2.1
1.0	5.3	1.1
2.0	too small to count	-

Table 2. Effects of combinations of 6-BA with IAA on bud induction of Serpent gourd.

Concentration of hormone (mg/L)		Explant	Number of bud per explant	Length of bud (cm)
6-BA	IAA			
0.5	0.1	apex bud	2.5	0.87
0.5	0.2	apex bud	2.7	1.13
1.0	0.1	apex bud	3.7	0.51
1.0	0.2	axillary bud	3.4	0.70
1.0	0.1	axillary bud	3.5	0.38
1.0	0.2	axillary bud	3.7	0.41
1.0	0.5	axillary bud	3.8	0.47
1.0	0.8	axillary bud	4.0	0.52
1.0	1.0	axillary bud	3.7	0.64

Table 3. Effects of combinations of 6-BA with 2,4D on bud induction of serpent gourd.

Concentration of hormone (mg/L)		Explant	Number of bud	Length of bud (cm
6-BA	2,4-D			
0.5	0.02	apex bud	2.0	0.3
		axillary bud	1.5	0.3
0.5	0.05	apex bud	*	*
		axillary bud	2.0	2.5
0.5	0.1	apex bud	*	*
		axillary bud	*	*
0.5	0.2	apex bud	*	*
		axillary bud	*	*
1.0	0.02	apex bud	5.5	3.4
		axillary bud	2.0	0.35
1.0	0.05	apex bud	*	*
		axillary bud	2.0	0.2
1.0	0.1	apex bud	*	*
		axillary bud	*	*
1.0 .	0.2 .	apex bud	*	*
		axillary bud	*	*

* Represents callus production of explants and no bud induced.

Literature Cited

1. Cao Huaxing, Gu Yongqiang, He Xiaohua. 1986. The effects of different cytokinine on the formation of adventitious buds in culture of watermelon. Journal of Shanghai Agricultural College. 4(4): 181-185.
2. Tang Dingtai, Zhang Jinglan, Xu Guifang et al, 1980. The effects of plant hormone on callus formation and regeneration of plant in *Cucumis melo* L.) Journal of botany 22(2): 132-135.
3. Tang Shaohu, Liao Yingfen, Xu Rongcan. 1994. Medium selection and tissue culture of seedless watermelon. Journal of Southwestern Agricultural University. 16(6):540-542.
4. Tao Yamin, Zhang Zhigang, Ye Jian, Pan Chongguang. 1994. Effect of several physical-chemical factors on somaclone propagation with seedless watermelon. Journal of Shanghai Agricultural College. 12(1):48-51.
5. Wang Xiaosu, LiBuxun, Wang Guangdong, 1997. In vitro plantlet regeneration from hypocotyl of *Sechium deule* Swartz. Journal of Northwestern Agricultural University. 25(1):83-87.
6. Kim., Y.H. and Janick, J. 1989. Somatic embryogenesis and organogenesis in cucumber. HortScience. 2:702.

Cucurbit Genetics Cooperative Report 23:83-85 (article 280) 2000

Research and Application of Seed Coating Agent for Cucurbit Crops in China

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Seed Coating Agents (SCAs) are compounds effective in protecting seed and young seedlings from attack by disease and insects. They may also supply nutrition for young seedling growth. SCAs are composed of active and inactive components. The former may include different kinds of insecticide, germicide, trace element fertilizer, plant growth regulator, etc. The latter includes membrane forming agents and warning color material. Since the 1980's, countries such as America, the former Soviet Union, Japan, and other western countries have obtained notable results in application of seed coating technology. Although seed coating agent research in China just started in the early 1980's, more than 30 kinds of SCAs have been developed, of which more than 24 have been put into commercial use. In China, SCAs are applied mainly to the most important crops such as rice, wheat, maize and cotton. The crop area sown with SCA treated seed reached 26,000,000 Ha by the end of 1998 and increases each year.

Generally, SCAs are designed according to region or to protect against the plant disease and insect pest expected to occur. They should have long efficacy (the effect can last for 40-60 days), and be absorbable, transferable and stable. The main type of SCAs in China are as following:

Pesticide and fertilizer compound type. China is a country with vast territory and greatly varied soil type. However, most of the soil is deficient in trace elements or infected by soil-borne disease. Therefore, most SCAs developed in china belong to pesticide-fertilizer compound type. For example, the eighteen SCAs developed by the China agriculture University from 1980 through 1985 are all this type.

> **Non-poisonous ecological type.** The current trend in China is the development of a series of non-poisonous ecological type SCAs to meet the needs of non-environment polluting crop production. This type of SCA utilizes microbes in its membrane as the active component. Therefore, they not only accelerate plant growth but also improve and protect the environment. "ZSB Ecological SCA" developed by the Seed Company of Zhejiang Province is an example of this type which has been applied to field crop production.

Region specialized type. Because China has a vast territory, varied ecotype, and different kinds of diseases and pests, the Chinese SCAs have a regional character. Generally, they are divided into two groups: southern type and northern type. There are also SCAs designed for specific regions.

Main components of Chinese SCA. The main chemical components of SCAs are considered to be either active or inactive components. Active chemical components refer to the material that can affect seed and seedlings such as pesticides, trace element fertilizers, plant growth regulators and microbes. the main active components used in chinese SCA at present are shown in Table 1.

Inactive chemical components are agents that maintain the physicochemical properties of the SCA, such as compounds related to membrane-formation, suspension, expander, acidity, adjustment adhesive, warning color, etc. Membrane-forming technology is critical for seed coating. The main component of the membrane-forming materials we are using is dissolvable dispersing-oseands and their derivatives such as acacia, substitution cellulose, etc. Some synthetic chemical compounds such as epoxyethane or poly-vinyl alcohol are also included. New types of components and technology such as hyper-moisture-absorbing resin SCA, seed embedded with mycose and ultra-micro powder seed coating technology are also being applied.

Main varieties of SCAs being used and produced in China at present. At present, more than 30 varieties of SCAs have

been registered in china. SCA applied to vegetable crops are shown in Table 2.

Other SCAs with good effect on vegetables now shown in Table 2 include: SCA Special for Gourd Vegetables and SCA Special for Cowpea, developed by the Vegetable Institute of Guangdong Province; Vegetable SEed Coating Agent No. 1, developed by the Tianjin Vegetable Institute; Eggplant SEed Coating Agent No. 1 and cucumber SEed Coating Agent No. 1, developed by the Northwest Sci-Tech Agriculture & Forestry University.

Effects of SCA on Gourd Vegetables. In gourd vegetables, SCAs are mainly used on watermelon, muskmelon and cucumber.The effects of these SCA on gourd vegetables are as follows.

On seed storage and germination. Seed longevity can be prolonged greatly by seed coating. Pu Hanli (1997) reported that the germination rate of coated seeds was 30% higher than that of the control after storage for 5 months and 66% higher after storage for 8 months. Germination rate, germination energy and germination index of cucumber seed coated with "Cucumber SEed Coating Agent No. 1", developed by Meng Huanwen (Horticulture Department of NSTUAF), were respectively, 5.78%, 1.27% and 6.63% higher than that of the control. The experiments on cucumber and tomato by Ma Wenhe (1999) showed that the germination rate was above the control level. Pu Hanli tested the effects of a gourd SCA on gourd vegetables and found that the field germination rate was increased on all tested crops and the best effect was shown on luffa (39/4% higher than that of the control.)

On growth energy and resistance of seedlings. Many researches have shown that seedlings from coated seed have higher growth energy and greater ability to withstand adverse ecology. Pu Hanli (1997) found that the ability to survive adverse ecology of luffa seedlings from coated seeds was 41.5% higher than that of the control. It was reported that root growth was improved and R/T ratio increased 150% and 180% respectively for cucumber and green pepper seed coated with Vegetable Seed Coating Agent No. 1.

On blossom and bearing of fruits. Due to better growth and increased ability to withstand adverse ecology, the plants from coated seeds bear fruits earlier. Pu Hanli's experiments demonstrated that plants grown from coated luffa seeds produced heavier fruit 10 days earlier than the control group. *Benincasa hispida* seedlings in nutritive cube field planted from coated seeds grew better and stronger. Their female flowers bloomed 5 to 6 days earlier and the fruits ripened 7 to 8 days earlier than that of the control.

Problems and Prospects. China is a country with large-scale vegetable production. The production area reached 11,270,000 ha by the end of 1998. Chinese cabbage seed production alone came up to 1140 tons. At present, seed production is being carried out all over China. Use of SCAs is increasing at an average rate of 30 percent each year. Therefore, there is a great market for SCAs in China. On the other hand, the vegetable seed coating industry is still at its beginning stages. Few SCAs are specially designed for vegetables, and development of SCAs for certain regional diseases are even fewer. Additionally, more attention should be paid to non-poisonous and non-polluting ecological types. Finally, the production ability should also be enlarged.

Table 1. The Main Active Components of Chinese SCA.

Kind of SCA	**Active component**
Pesticide	Carbofuran, Carbosulfen, Imidaproprid, Teflathrin, Frpronil, Lindane
Germicide	Carbendazim, Thiabendazole, Thiophanate, Triodimend Diniconazole, Carboxin, Captan, Thriam, Rhizolex, Imizalil Dimetachtalone, Metalaxyl, Mencozeb, Hymexazol, Zineb, Zinc, Methabearsibatem Myclobutanil, Fenpiclonil Tebuconazole, Cyproconazole
Plant growth Regulator	Pactobulrazole, Samiseren, Chlomequat, Triadimefon
Trace element	Zinc, Manganese, Iron, Molybdenum

Table 2. SCA Applied to Vegetables in China.

SCA	**Crops to be applied**	**Prevent objects**
No. 5	Muskmelon, watermelon and other vegetables	Vegetable Fusarium wilt, Vertcillium wilt, Damping off, Web blight, Wilt and Anthracnose

No. 7	Watermelon, rape and other vegetables	Vegetable and Watermelon Fusarium, Anthracnose, etc.
No. 9	Watermelon and other vegetables	Watermelon and Vegetable anthracnose, Fusarium, Web blight, Damping off, Elemental deficiency
No. 10	Watermelon, beet and other vegetables	Soil insects, Angular leaf spot, Fusarium wilt Anthracnose, Physiological disease, Increased yield
No. 32	Watermelon	Soil and foliar disease and insects

Cucurbit Genetics Cooperative Report 23:86-86 (article 29) 2000

Proceedings of the 1st International Oil Pumpkin Conference

9-13 August 1999

Styria & Lower Austria

Preface

Extracting oil from pumpkin seed is a century-old tradition centered in Styria, in the south-eastern part of Austria. Some 130 years ago, a recessive mutation occurred which prevented the coat of the pumpkin seed from lignifying. Generations of people who had spent their winter evenings removing the tough hulls from the seeds realized the benefit of this mutation. Subsequent selection of the green-seeded plants, homozygous for the mutation, led to the establishment of the ecotype *Cucurbita pepo* var. *styriaca.*

Over the centuries, the Styrians became aware of the healing qualities of pumpkin seeds and their oil and gave it an honored place in their folk medicine. Today, however, some of the curative effects - especially for prostate ailments and bladder irritations - have been scientifically proven, producing a growing market demand for pumpkin seed oil and other products made from pumpkin seed extract.

The prospect of combining age-old traditions with modern technologies for the benefit of growers and consumers motivated the organization of the "*First International Oil-Pumpkin Conference,*" which was held in Austria. It attracted people from countries like Israel, Russia and even New Zealand, and provided an excellent platform for discussing many aspects of the oil-seed pumpkin, such as history, breeding experiences, the virus question, technical aspects of pressing, introduction to new countries or regions, and marketing.

The proceedings of the conference present the essence of these four days of presentations, discussions, and information exchange in August 1999, held in different places in Austria. It forged a small community of people now dedicated to the "Styrian oil-seed pumpkin" who plan to meet again in 2003 to re-evaluate the problems, solutions, and new problems. We hope that reading these contributions will awaken interest and encourage discussion for the benefit of a natural product, which is tasty, healthy and produced in harmony with nature. For comments and further contributions we invite you to visit the website of the conference: *http://www.cucurbit.org/cukeoil.html*

Penny Lichtenecker and Tamas Lelley
March 2000, Vienna

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An Overview of the Oil Pumpkin

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The use of pumpkin (*Cucurbita pepo*) seed is not new. The wild gourd-like ancestors of pumpkins contain edible and highly nutritious seeds, and their consumption most likely constitutes the first use of this species. Archaeological evidence shows that soon after the end of the last ice age (over 10,000 years ago), *C. pepo* was being used and eventually became domesticated in North America. Since the species has multiple uses, selections from the genetically diverse wild populations went in various directions. Seed usage led to selection for larger seed size. This in turn led to larger fruit size with the seeds becoming more readily separable from the pulp. The discovery of wild plants with a mutation for non-bitter flesh led to selections for thicker, higher quality flesh at the expense of seed cavity size and the proportion of seeds per fruit. Pumpkins today are most commonly grown for their flesh with the seeds generally ignored. But there are regions in Mexico where the seeds are still considered the most desirable part of the plant. Landraces in these regions tend to have fruit that are thin-fleshed with proportionally large seed cavities containing seeds that are generally elongated, up to three times longer than wide and thick in cross-section. This seed shape facilitates hand dehulling along the raised margins to remove the indigestible testa. The seeds may be eaten raw or roasted as a snackseed, commonly called "pepitas," or ground into a "pipian" sauce. The testa of the seed varies in thickness and hardness, and thinner testa is tolerated when ground or roasted.

During the sixteenth century, pumpkin was introduced throughout Europe. Central Europe by then already had a well-established oilseed industry, including the pressing of flaxseed (*Linum usitatissimum*), rapeseed, and other mustard species (*Brassica* spp.) The high quantity and quality of the oil from pumpkin seeds along with other introduced North American domesticate, the sunflower (*Helianthius annus*), was soon realized. The oil mills needed to be only slightly modified to take advantage of these new introduced oilseeds. Thus a new use for an ancient domesticate was born.

In the Americas, pumpkin seed was also recognized in folk medicine to be an anthelmintic or vermifuge to expel intestinal worms. When pumpkin seed became popular in Germany, Poland, Austria, Hungary, the Balkans, and Ukraine, a pattern was observed of a low incidence of prostate disorders in this region. This has been attributed to the high consumption of pumpkin seed. A pharmacological study is still needed to substantiate whether there is any unique factor in pumpkin seeds beyond for example a high vitamin content to explain this (Wagner, this volume). The seeds and oil are often advertised as a remedy for urinary problems and benign prostate adenomas and are sometimes old in capsular form mixed with other medicinal herbs. The oil is highly polyunsaturated and rich in protein, and thus of high nutritional value.

Presently in Central and Eastern Europe, particularly in the Austrian province of Styria, a hull-less or so called "naked" seeded strain of pumpkin, sometimes called the Styrian pumpkin, is grown for the production of pumpkin oil. The seedcoat layers are morphologically still present as in regular pumpkin seeds but differ in being membranous rather than having thickened and lignified cell walls. Therefore they require less heat and pressure to extract the more concentrated oil. Furthermore, they may be eaten whole as snackseeds without the need for dehulling.

The origin of this unusual mutation is unclear. There are report at the end of the nineteenth century of this trait in European seeds (Teppner, this volume), although pressing for pumpkin seed oil dates back perhaps a couple of centuries prior to this. In the Americas, there are no reports of oil pumpkins earlier than the first European ones. The first naked seeded cultivar in the United States was 'Lady Godiva', released by the U.S. Department of Agriculture in 1972. It was a selection developed from European landraces. Subsequent cultivars in the U.S. with the naked seeded (or semi naked seeded) trait include 'Eat-It-All', 'Mini-Jack', 'Streaker', 'Trick or Treat', 'Trickster', 'Tricky Jack' and 'Triple Treat'.

Although the mutation is generally believed to have occurred spontaneously in Europe, thin-seeded plants of *C. pepo* are also found on rare occasion in northern Mexico.

Other countries are beginning to show an interest in the cultivation of oil pumpkin, including Canada, France, China, New Zealand, and Australia. Improvement in the economics of oil pumpkin production and its increased usage will depend on a

number of factors including:

1. increasing germinability of the easily breakable, rot-prone seeds;
2. improving cultural practices;
3. improving mechanical methods of harvesting the fruit and seeds;
4. optimizing the use of byproducts for animal feed, including: (a) the fruit after the seeds have been removed, and : (b) the pressed seed cakes after the oil has been extracted;
5. increasing usage of both seeds and fruit flesh for human consumption;
6. developing techniques to cure and extract the oil from the seeds to insure maximum yield and optimal flavor and nutrition;
7. improving crop protection and use of locally adapted cultivars through breeding for disease and pest resistance (especially important since pesticides are not generally used);
8. genetic improvements to obtain consistent high seed yields by producing a higher number of seeds per hectare and by increasing the oil content per seed, along with improvements in seed size and shape (particularly important for snackseeds), oil composition, color, and taste;
9. avoidance of rancidity to improve shelf life of the seed and oil;
10. increasing funding for marketing to present more attractive products and educate the consumer on the benefits and uses of pumpkin oil and pumpkin snackseed and promote the environmentally friendly manner in which it is traditionally produced on family-run farms.

All of these factors can help lower the cost and increase the profit of this otherwise high-priced food commodity.

Novel products derived from oil pumpkin seeds, such as the removal of the green endosperm layer of the seed to make a clear straw-colored oil that may be used in frying, need to be explored. The oil from oil pumpkin otherwise comes from pressing the entire seed, which produces a thick dark-colored oil with a low burning point that can not be used in frying. Other under-utilized potential marketable products include seed butter made from the grounded roasted seeds for use as a peanut butter-like sandwich spread and seed flower utilized as a thickener and seasoning. The snackseed may be used as an ingredient whenever other nuts are used, such as in granola or confectionery. Numerous flavors may be tried out on pumpkin snackseeds, such as a wasabi flavoring.

With the high degree of genetic variation in the genome, there is great potential for making breeding improvements in the oil pumpkin. If the recent history of other oilseed crops, such as soybean (*Glycine max*), sunflower, and specialty flavoring oils, such as walnut oil, provide any lesson, rapid changes in usage may take place.

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Seed Development in *Cucurbita pepo*: An Overview with Emphasis on Hull-less Seeded Genotypes of Pumpkin

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Abstract: Seeds of *Cucurbita pepo* L. are high in protein and oil, and a good source of potassium, phosphorous, some of the minor elements, and some of the B vitamins. The seeds are also large and abundant in fruits. Nonetheless, they are underutilized as a food crop, probably owing to the thick, leathery seed coat (hull) that cannot be removed easily be mechanical means. The existence in this species of a mutant seed phenotype characterized by an unlignified, thin seed coat has provided a genetic means by which breeders can develop new germplasm with essentially hull-less seeds that can be more easily exploited as a food and source of vegetable oil. To better exploit hull-less seeded cultivars as a food crop, we have been conducting research on seed development in this species. We subdivide seed development into three phases: (1) expansion and biomass accumulation in seed coats and differentiation of the embryo; (2) rapid embryo enlargement and depletion of seed coat reserves; and (3) Biomass accumulation (seed fill) in embryos. Biomass accumulation in seed coat peaks at 20 days post-anthesis (PA), coinciding with maximal fruit expansion. In both hulled and hull-less seeds extensive amounts of nonstructural constituents (starch, lipids, sugars, amino acids) accumulate in seed coats between 0 to 20 days PA. In normal seeds, secondary cell wall thickening begins early in seed coat development and is characterized by extensive lignification and cell wall thickening in hypodermal, sclerenchymatous and parenchymatous tissues by 20days PA. Secondary wall thickening continues as seeds mature. In hull-less genotypes there us a reduction in lignification of tissues, the difference being first detectible by lignin staining at 10 days PA, but becoming increasingly apparent during seed development. Cellulose accumulation is also reduced in hull-less as compared to hulled genotypes early in seed development, but unlike lignin, this difference is not accentuated in mature seeds. Rapid embryo enlargement occurs between 20 to 40 days PA. Seed fill increases more or less linearly between 25 to 60 days PA; however, rates of seed fill and the duration of seed fill may vary according to environmental conditions during fruit growth and seed development. In fruits harvested prematurely, assimilates stored in mesocarp tissue can be remobilized to the developing seed. Changes in color of the fruit exocarp normally associated with maturation of pumpkins do not necessarily coincide with the time of maximum seed fill.

Introduction: Seeds are a rich source of nutrients for human consumption, and as such, serve as the main food base for most of the world population. Seeds within the genus *Cucurbita* contain 32 to 37% protein and 42 to 50% oils (6), and also contain relatively high amounts of potassium, phosphorous and zinc (9) and significant quantities of niacin and thiamine (7). Thus, they represent a highly nutritious and potentially important seed crop for human use. However, because of the thick, leathery seed coat associated with most cultigens, and the difficulty in decorticating the seed, *Cucurbita* seeds have been underutilized as a human food. A recessive mutation (n) in *C. pepo*, in which secondary cell wall constituents are much reduced within the outer seed coat tissues (5, 14), provides a convenient genetic trait for developing hull-less seeded cultigens, the seeds of which can be more readily utilized as a human food source. These hull-less or 'naked seeded' cultigens of pumpkin may have been propagated for over 100 years in Austria (H. Teppner, personal communication) but the first published reference to genetically hull-less or 'schalenlosen' seed in *C. pepo* of which I am aware was in 1934 by an Austrian scientist, Tschermak-Seysenegg (16). Because pumpkin seed oil was highly valued in austria and other parts of Eastern Europe for use in salad dressings, the utility of hull-less seeded strains for more efficient extraction of the oil was immediately recognized.

Seed coat composition and function in hulled and hull-less cultigens of *C. pepo*: Morphologically, seed coats of *cucurbita pepo* are subdivided into two layers that can be easily separated in mature seed, a relatively thick, leathery outer layer and a thin, membranous-like, green inner layer. In 1909 Barber (3) reviewed the major nineteenth century contributions to cucurbit seed morphology, and provides an accurate illustration of the major seed coat tissues in *Cucurbita pepo.* Singh and Dathan (11) give a more detailed account of the derivation of the five zones or tissue types comprising the seed coat layers. The inner seed coat consists of several layers of thin-walled, relatively large, chlorenchyma cells. The outer seed coat consists of tangentially elongated, thin-walled, nonlignificed epidermal cells, 3 to 5 layers of small, extensively lignified hypodermal cells, a single layer of large, heavily lignified schlerenchymatous cells, and one or two layers of small, partially lignified parenchymatous and aerenchymatous cells. These tissue layers are formed by 10 days post-anthesis (PA), but the palisade epidermal cells are not fully elongated until 15 to 20 days PA (12). In mature desiccated seeds, the inner layer of chlorenchyma tissue compresses into a thin, membranous-like chlorophyllous layer (14). This layer usually remains intact when seeds are decorticated by hand. In the outer seed coat of mature, dried seed, the epidermis largely collapses, but the lignified hypodermal, sclerenchymatous, and arenchymatous tissues maintain their integrity and form the hard,m leathery hull characteristic of *cucurbita* seed.

Heinisch and Ruthenberg (5) made a detailed anatomical study of the seed coat in hulled and hull-less genotypes of pumpkin and reported that in 'naked seed' genotypes all seed coat layers were present, but thickening and lignification of cell walls were reduced,. They as well as other early investigators (8, 19, 19) noted genotypes in segregating populations with different degrees of cell wall thickening.

Stuart and Loy have conducted more detailed anatomical studies and have also analyzed seed coat composition in hulled and hull-less cultigens. At 10 days post-anthesis, prior to appreciable secondary wall development, hulled and hull-less genotypes are nearly indistinguishable (14), with the exception of slight staining with phloroglucinal (indicative of lignin) in normal genotypes. However, by 15 days post-anthesis, phlorogluycinal-positive staining within hypodermal, sclerenchymatous and aerenchymatous tissue is clearly much greater in hulled as compared to hull-less genotypes (12). The disparity in secondary cell wall thickening between hulled and hull-less genotypes continues to magnify as the seeds mature.

Quantitative estimates of the structural (cell wall) constituents of seed coats at 20 days PA and in mature seeds are given in Table 1. Henicelluloses, pectins and cellulose are the predominant cell wall constituents in normal seed coats at 20 days PA. Mutant seed coats exhibit normal amounts of pectins and hemicelluloses, but have much reduced amounts of cellulose and, especially, lignin as compared to hulled genotypes. Pectins and to a lesser extent hemicelluloses are largely degraded between 20 days PA and seed maturity. The reduction in hemicelluloses is much greater in hull-less as compared to hulled seeds. This may be because there is greater accessibility within secondary cell walls of hull-less than of hulled genotypes to hydrolytic enzymes that degrade cell wall polysaccharides (see 1, 2). In normal, hulled seed there is about a 3-fold increase in lignin and in some cases a slight increase in cellulose accumulation between 20 days PA and seed maturity. The most pronounced differences between seed coat genotypes at seed maturity are 60 to 70% reductions in cellulose and 79 to 88% reductions in lignin content in hull-less as compared to hulled seed coats (Table 1). The reduction in cellulose in hull-less as compared to normal seeds at maturity may well be at least partly due to greater degradation by hydrolytic enzymes as suggested for the hemicelluloses. It should be noted that the hull-less cultigens used in Stuart's studies (12, 13) exhibited some cell wall development in the outer seed coat, especially along the seed margins. This is in contrast to most of the Styrian cultigens I have observed, and to several of our own breeding lines, in which lignified tissues of the outer seed coat are virtually absent.

Nonstructural constituents, starch, lipids, sugars, free amino acids, and soluble proteins, are in great abundance in seed coats at 20 days PA (Table 2). Starch is especially abundant in chlorenchymatous tissues in both hulled and hull-less genotypes, but is also relatively abundant in hypodermal and parenchymatous cells of hull-less strains (14, 17). It is conceivable that in hulled as compared to hull-less genotypes, the starch in these latter two tissues may be more rapidly utilized for secondary wall formation. Between 20 days PA and seed maturity the nonstructural constituents are largely depleted. In hulled seeds it is presumed that some of these assimilates are utilized for lignin synthesis. In both genotypes, however, these constituents appear to serve as a major reservoir of assimilates for the developing embryo.

Stages of seed development:

Seeds exhibit a relatively linear increase in total biomass from 10 days post-anthesis (PA) until seed maturity at 55 to 60 days PA (Fig. 1.) However, different organs within the seed display different periods of development and rates in biomass accumulation (18). As such, seed development in *C. pepo* can be conveniently subdivided into 3 distinct, but overlapping stages: (1) 0 to 20 days PA - period of expansion and accumulation of reserves in seed coats, endosperm development and differentiation of embryos: (2) 20 to 40 days PA - period of rapid embryo enlargement, reduction in endosperm fresh weight and decrease in seed coat nutrient reserves; and (3) 25 to 60 days PA - period of accumulation of seed storage reserves.

Fresh weight and biomass of seed coats increases rapidly after fertilization and peaks at 20 days PA, after which the decrease slows until seeds reach maturity. Embryos remain relatively small and difficult to detect with the naked eye until about 20 days PA. Shortly thereafter, embryos enlarge rapidly and fill in the seed coat cavity between 35 to 40 days PA. The period of rapid cotyledon enlargement may vary considerably even within a single fruit, but according to our observations, rarely begins until after 20 days PA and cytologically appears complete by or before 40 days PA, but occasionally 3 mm long embryos were recovered at that stage (17). Quantitatively, embryos show nearly linear increases in fresh weight until about 45 days PA, after which growth usually slows until seed maturity at 60 days PA. Later stages of embryo expansion are often coincident with collapse of perisperm and inner seed coat tissues. We have on occasion, however, observed some fresh seeds at 60 days PA in which the perisperm and inner seed coat tissues are still largely intact, because of poor seed fill.

The patterns of biomass increase among seed organs can be expected to vary with differences in growing conditions. In our 1997 experiment seed biomass at 35 days PA was 40 to 42% of that in mature seed among three genotypes. But in a test of 11 experimental, hull-less seeded hybrids in 1999, seed biomass at 35 days PA averaged 55% of that of mature seed. The 1999 growing season was unusually warm and seed development occurred between about 15 July to 15 September. In 1997 fruit and seed development occurred between 28 July and 30 September under much cooler but fairly normal conditions for our region.

Fruit maturity and biomass changes in seeds from stored fruit: During crop growth plants ideally remain healthy and canopy photosynthesis remains active until fruit and seeds mature. Our observations on culture of pumpkins over the past 20 years suggest that it is quite common for plants to senesce prematurely before fruits are deemed mature. Relative to this phenomenon, we have been confronted with two problems when dealing with our culture and evaluation of hull-less seeded hybrids: (1) from visual observations of rind color, can we tell when fruit and seeds within the fruit are mature, and (2) if vines senesce prematurely, will fruit and seeds continue to develop and mature, and if so, to what extent.

The first problem was relatively easy to tackle, but resulted in a few surprises. We compared two inbred lines, NH285 and NH29-13, and their F_1 hybrid. We presumed from previous field observations that NH285 was late maturing. In contrast, we assumed that NJ29-13 matured early because of early changes in fruit color from green to dark orange. In our comparative studies in 1997, the skin or rind of NH29013 fruit indeed showed extensive changes from green to almost full orange coloration between 30 to 40 days PA (17, 18). On the other hand, NH285 fruit exhibited only subtle changes in skin color from the initial pattern of alternating light and dark green stripes. AT 45 days PA about half of the fruit exhibited some tinges of orange color light green portion of fruit, and after 60 days most fruit were light orange and green striped on the surfaces exposed to the sun. NH1003, the F_1 hybrid, were striped, but the striping was less distinct than that of NH285. By 40 days PA some fruits exhibited light orange and green stripes, and by 55 days PA most fruits were yellow-orange with no prominent striping. The different patterns of fruit coloration cited above were often not correlated with seed maturation. In all three cultigens embryo biomass increased significantly between 50 to 60 days PA, at a time when all fruits of NH29-13 and most fruits of NH1003 appeared mature. The changes in embryo biomass during this late period of maturation were astonishing: 43% increase in NH29-13, 47% increase in NH1003, and a 35% increase in NH285, the presumably late maturing cultigen.

Fruit mesocarp tissues reach peak biomass in *C. pepo* at about 30 days PA (4,18), at a time when seed fill is just beginning to accelerate. Thus, we hypothesized that fruit mesocarp reserves might serve as a source of assimilates for developing seeds under conditions in which leaf canopy production of photosynthates is limited. To test this, we excised fruit prematurely (35 to 50 days PA) from field-grown plants and stored them in a glasshouse for 10-day periods; changes in seed fill were compared between stored and intact fruits (18). In two sampling periods, 35 to 45 days PA and 40 to50 days PA, seed fill in stored fruits of NH29-13 was 12,7 and 20.5% lower, respectively, than that of intact fruit. In three sampling periods (35-45, 40-50, and 45-55 days PA) of NH1003, seed fill in stored fruit was 17.2, 16.6 and 58.3% lower than that in corresponding intact fruit. In two sampling periods (40-50, 45-55) for NH285, seed fill in stored fruit was essentially the same as in intact fruit. In more recent studies completed in 1999, we compared seed fill in intact fruit to those excised and stored in the field for at least 25 days, from 35 to 60-65

days PA. In this study, comparing 11 hybrids, seed fill in stored fruits averaged 20% less than that of intact fruit, even though seed biomass was 55% complete by 35 days PA. In 9 of the 11 hybrids for which we had reliable data, the increased seed fill (biomass) in stored fruits accounted for between 32 to 54% of the estimated loss in mesocarp biomass during the storage period. The results thus show that in absence of a supply of photosynthates to developing seeds during the seed fill stage, assimilates in fruit tissues can be remobilized to the seed.

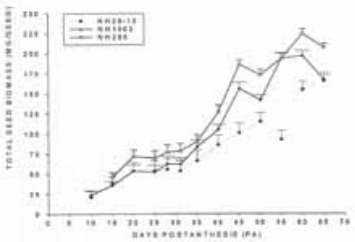

Fig. 1. Biomass accumulation in seeds of three hull-less seeded genotypes between 10 to 65 days post-anthesis. Bars denote SD. (taken from 17)

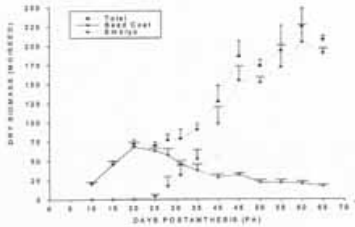

Fig. 2. Changes in seed coat, embryo and total biomass during seed development NH285, a hull-less seeded genotype (taken from 17).

Conclusions: The dynamics of fruit and seed development in *cucurbita pepo* pumpkins are not only important from a scientific viewpoint, but also for developing optimum cultural systems for maximizing and sustaining fruit and seed production. Fruit reach optimum size at about 20 days PA, and this also coincides with the period of peak fresh weight and biomass of seed coats (18). The function of seed coats is much broader than just a protective covering for seed embryos. All nutrients transported from the fruit tissues to the developing embryo enter the seed via the vasculature connecting the placental tissue to the micropylar end of the seed and running along the margins of the seed coat. Subsequent transport to the embryo must occur apoplastically and/or symplastically through the seed coat and perisperm tissues, and apoplasticaly from the perisperm to the endosperm enclosing the embryo. Starch, lipids, pectins, sugars and amino acids are abundant in seed coats during early seed development, so seed coats appear to serve as a major reservoir of nutrients for later embryo expansion and seed fill. It is also sometimes not appreciated that in *C. pepo* and other cucurbits seed size is largely maternally regulated or determined. Maximum seed coat size at 20 days PA largely delimits the degree of embryo expansion and final seed size. This phenomenon is true for both hull-less and hulled genotypes. Compared to normal hulled seed, the seed coat in hull-less genotypes does not offer a strong physical barrier to embryo expansion, so differences in water potentials between embryo tissue and adjacent maternal tissues (perisperm and inner seed coat) likely regulate final embryo size in developing seed.

Because seed coat the fruit expansion occur simultaneously, the degree of locule expansion influences seed coat expansion and final seed width and length dimensions. For example, in small fruit (0.5 to 1.2 kg) we rarely obtain seeds larger than 200 mg; whereas in all of our large-seeded lines, fruit size is greater than 2.0 kg. Nonetheless, breeding liens with small fruit but endowed with genes for large seed, will produce large seeds under conditions of low seed set.

A considerable portion of final seed biomass may accumulate during the last 10 days of seed development and maturation which, in some cases, occurs much later than when fruit appear mature according to changes in skin color. Therefore, determination of the extent of maximum seed fill is critical for maximizing seed yields in commercial production. furthermore, because nutrient reserves in fruit tissues reach a maximum at 30 days PA and can be remobilized to the developing seed, acceptable seed yields may be obtained in production fields under conditions in which plants senesce prematurely due to disease or other stress conditions.

Table 1. Structural (cell wall) constituents of seed coats in 20-day post-anthesis and mature seeds of normal and hull-less cultivars of pumpkin.[z]

	20-day PA seed coats		Mature seed coats	
	Hulled	**Hull-less**	**Hulled**	**Hull-less**

Constituent		*mg per seed coat*		
Protein[y]	5.07	4.24	6.51	2.94
Pectins	9.65	8.45	0.85	0.55
Hemicelluloses[x]	10.15	10.25	4.94	1.63
Cellulose	8.65	3.70	9.65	2.99
Lignin	3.20	0.60	8.80	1.37

[z] Values extrapolated from data of Stuart and Loy (13, 14) and based on averages from analysis of two hulled cultivars (Small Sugar and Jack o' lantern) and two hull-less cultivars (Tricky Jack and 293A).

[y] Proteins from mature seed estimated from N content of digested samples x 6.25, and considered to be largely those associated with the cell wall. Proteins from 20-day PA seeds were fractionated into soluble and bound fractions, with only the later given in Table 1.

[x] Values represent combined fractions of hemicelluloses, Hc_A and Hc_B.

Table 2. Nonstructural constituents in seed coats of 20-day post anthesis (PA) and mature seed of hulled and hull-less cultivars of pumpkin.[z]

	20-day PA seed coats		Mature seed coats	
	Hulled	**Hull-less**	**Hulled**	**Hull-less**
Constituent		***mg per seed coat***		
Soluble protein	2.25	2.19	-	-
Free amino acids	3.07	3.43	0.17	0.15
Reducing sugars	5.20	4.53	0.24	0.25
Phenolics	0.24	0.27	0.03	0.03
Starch	9.55	10.82	0.97	0.22
Lipids	6.65	5.11	0.93	1.21

[z] Values from data of Stuart (12), and based on averages from analysis of two hulled cultivars (Small Sugar and Jack o' lantern) and two hull-less cultivars (Tricky Jack and 293A).

Literature Cited

1. Bailey, R.W. and D.I.H. Jones. 1971. Pasture quality and ruminant nutrition. III. Hydrolysis of rye grass structural carbohydrates with carbohydrases in relation to rumen digestion. N.Z. J. Agr. Res. 14: 847-857.
2. Bailey, R.W. and S.E. Pickmere. 1975. Alkali solubility of hemicelluloses in relation to delignification. Phytochemistry 14:501-504.
3. Barber, K.G. 1909. Comparative histology of fruits and seeds of certain species of Cucurbitaceae. Bot. Gaxz. 47:263-310.
4. Culpepper, C.W. and H.H. Moon. 1945. Differences in the composition of the fruits of *Cucurbita* varieties at different ages in relation to culinary use. J. Agr. Res. 71:111-136.
5. Heinisch, O. and M. Ruthenberg..1950. Die Bedeutung der samenschale fur die Zuchtung des Okurbis. Z. Pflanzen. 29:159-174.
6. Jacks, T.J., T.P. Hensarling and L.Y. Yatsu. 1972. Cucurbit seeds: I. Characterizations and uses of oils and proteins. A review. Econ. Bot. 26:135-141.
7. Mansour, E.H., E. Dworschak, A. Lugase, E. Barna and A. Gerely, 1993. The nutritive value of pumpkin (*Cucurbita pepo* Kakai 35) seed products. J. Sci. Food Agr. 61:73-78.
8. Mudra, A. and D. Neumann, 1952. Probleme und Ergebnisse der Munchenberger Olkurbiszuchtung Zuchter 22:99-105.
9. Robinson, R.G. 1975 Amino acid composition of sunflower and pumpkin seeds. Agron. J. 67:541-544.
10. Schoeniger, G. 1950. Genetische Untersuchungen und *Cucurbita pepo*. Zucher 20:321-336.
11. Singh, D. and A.S.R. Dathan. 1972. Structure and development of seed coat in cucurbitaceae. VI. Seeds of *Cucurbita.* Phytomorphology 22:29-45.
12. Stuart, S.G. 1981. Comparative studies of testa development in normal and hull-less seed strains of *Cucurbita pepo* L. M.S. Thesis, University of New Hampshire, Durham, N.H.
13. Stuart, S.G. 1983. Comparative biochemical and genetic studies of testa development in normal and hull-less phenotypes of pumpkin (*Cucurbita pepo* L). Ph.D. thesis, University of New Hampshire, Durham, NH, U.S.A.
14. Stuart, S.G. and J.B. Loy. 1983. Comparison of testa development in normal and hull-less seeded strains of *cucurbita pepo* L. Bot. Gaz. 144:491-500.
15. Stuart, S.G. and J.B. Loy. 1988. Changes in testa composition during seed development in *cucurbita pepo* L. Plant Physiol. (Life Sci. Adv.) 7:191-195.
16. Techermak-Seysenegg, E. 1934. Der Kurbis mit schalenlosen samen, eine beachtenswerte Olfruct. Wiener Landwirts. Zeit. 84:7-15.
17. Vining, K.V. 1999. Seed development in hull-less seeded pumpkin (*Cucurbita pepo* L.), M.S. Thesis, University of New Hampshire, Durham, NH, U.S.A.

18. Vining, K.V. and J.B. Loy. 1998. Seed development and seed fill in hull-less seeded cultigens of pumpkin (*Cucurbita pepo* L.), p. 64-69. In: J.D. McCreight (ed.), Cucurbitaceae 98: Evaluation and enhancement of cucurbit germplasm. ASHS Press.
19. Weilung, F. and E. Prym von Becherer, 1950. Zur Factorenanalyse der Testaausbeldung beim Kurbis. Ber. Deut. Bot. Ges. 63:147-148.

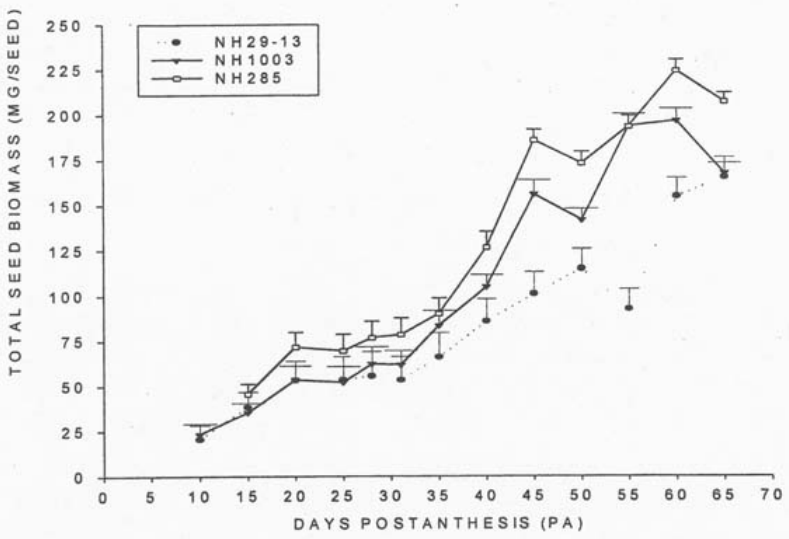

Fig. 1. Biomass accumulation in seeds of three hull-less seeded genotypes between 10 to 65 days post-anthesis. Bars denote SD. (taken from 17)

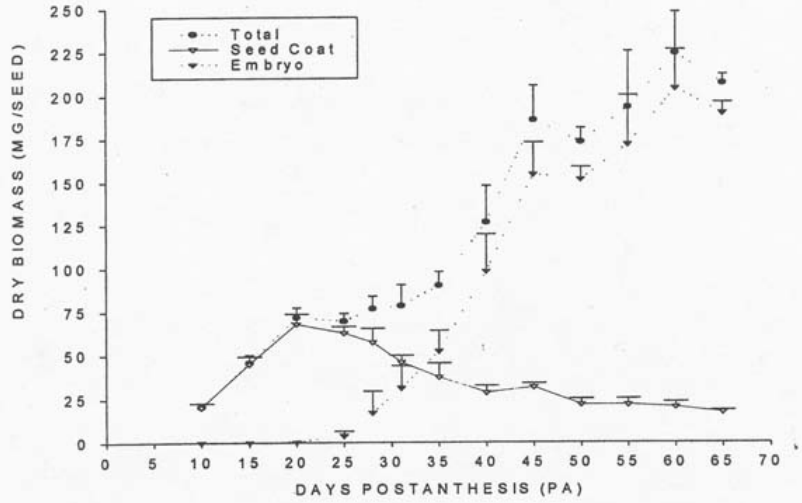

Fig. 2. Changes in seed coat, embryo and total biomass during seed development NH285, a hull-less seeded genotype (taken from 17).

Cucurbit Genetics Cooperative Report 23:96-98 (article32) 2000

A Preliminary Survey of Oilseeds in the Cucurbitaceae

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Earle and Jones (1962) rank the Cucurbitaceae as one of the highest plant families in terms of mean protein content as well as mean oil content of its seeds, and assert that it "appear(s) to offer outstanding promise as potential sources of new oilseeds." Over a dozen genera contain species, both domesticated and wild, that have been exploited for their oil (Table 1). Some are grown exclusively for their oilseeds, or at least contain cultivars that are grown for such purposes. But most have multiple uses, such as the fluted pumpkin, *Telfairia occidentalis*, which is cultivated in southern Nigeria primarily as a vegetable for its nutritious shoots and only secondarily for its oil-rich seeds. Even where the seed oil is the primary product, byproducts are often used as animal feed. Most cucurbit oilseeds are cultivated on a small scale for subsistence use and therefore are poorly known outside of local areas; thus the reason for this survey. None have attained the commercial importance of the hull-less oil pumpkin (*Cucurbita pepo*) in spite of their food potential. while oil pumpkin is unique in not having a hard seedcoat, which imparts certain advantages discussed in this volume's reports from the Oil Pumpkin Conference, other species contain different desirable traits that have not been fully exploited.

Worldwide the most popular edible seed in the family come from the genus *Cucurbita.*All the species in the genus contain edible seeds with both a high protein and oil content. In comparing the other species in the genus to the oil pumpkin, some have larger seeds while others bear fruit with a higher ratio of seeds to flesh. *cucurbita argyrosperma*, for example, was domesticated in pre-Colombian Mexico and Central America chiefly for its seeds. The silver-seeded form of *C. argyrosperma* has large seeds with pronounced seed margins that facilitate hand-dehulling. Some cultivars and landraces of *cucurbita maxima* have seeds containing the largest kernels in the genus, but these seeds lack a distinct margin. this later species, along with *C. pepo* , is used in southern Russia for seed oil. *Cucurbita ficifolia* tolerates cool temperatures and is cultivated at higher altitudes than any other *Cucurbita* . Its large, flat seeds are laboriously hand-dehulled by descendants of the Incas and sold at appropriately high prices in regional Andean markets. *Cucurbita moschata* , which grows best in hot humid lowlands, is also sometimes grown for its edible seeds. There is one report of a paid of recessive naked seed genes found in *C. moschata* in China (Zhou, 1987). It remains to be seen whether this is under the same genetic control as the oil pumpkin and whether this trait can be crossbred into other *Cucurbita* species.

Attempts have been made starting in the 1940s to cultivate some of the wild species of *Cucurbita* for use of their seeds, particularly the xerophytic species that thrive on marginal lands. The wild species contain much smaller seeds than the domesticates , but what they lack in size is made up for in quantity of seeds produced in the numerous, thinly fleshed gourd-like fruits borne on each vine. Therefore the net yield in kernels is high. The greatest effort at producing such a new crop has been invested in the buffalo gourd (*Cucurbita foetidissima*). This wild perennial species from the southwestern U.S. and northern Mexico grows particularly well in traditionally non-arable, semi-arid regions. Preliminary studies have shown great potential for this and other xerophytic species, including *Cucurbita palmata, C. digitata,* and *Apodanthera undulata.* But attempts at commercial production in the southwestern U.S. and adjacent northwestern Mexico, as well as Chile, Lebanon, Pakistan, and Australia, have not yet succeeded. Cost-effective cultural practices and commercial acceptance have not been realized. This demonstrates the difficulty of domesticating a wild plant.

Tropical Africa and India represent the richest regions for the use of Cucurbitaceae oilseeds. Different tribes often have various uses and names for the same species of cucurbit. Vernacular names such as "egusi" are confusingly applied in Nigeria to the oilseeds of both *Cucumeropsis mannii* and at least two species of *Citrullus*, all of which are used to make egusi soup. Manu local uses have not been adequately studied even among species that constitute important dietary sources of protein and oil. In regions where there is a shortage of animal protein in the human diet, cucurbit oilseeds are used as meat extenders or as an alternative source of protein.

The most widely grown species in Africa is watermelon (*Citrullus lanatus*). Its black, brown, red, tan or green colored seeds

have some of the most diverse uses in the family and are very popular in local areas, particularly in Nigeria, where the subspecies *colocynthoides* is cultivated. Since the flesh is bitter in this subspecies, the plants are grown solely for the seeds, which are larger than typical watermelon seeds. The bitter flesh helps protect the fruit from animal predation. *Citrullus* seeds may be ground into a substance like peanut butter, used as a condiment and thickener in soup and gravies, or used as flour to make bread. In West African cooking, a flavoring called ogiri is made from fermented *Citrullus* seeds, or sometimes from other cucurbit and leguminous seeds. The pale yellow oil extracted from *Citrullus* seed is sold commercially and used as a cooing and frying oil, and the residual protein-rich meal is sometimes friend in this oil. The oil is also used for making soap and as an illuminant. The snackseeds are popular in China where they are sometimes pickled in soya sauce or sugared. In the Central African Republic, the dry dehulled seeds are popped like puffed rice. A related wild species, *Citrullus colocynthis,* naturally produces an abundance of seeds in desert regions. While it has been exploited primarily for the laxative drug derived from its fruit pulp, it has also been known since Biblical times as a source of seed oil.

As in the genus *Citrullus*, there are some cultivars of *Cucumis* that are used solely for seed production. For example, *Cucumis melo* 'chate' is grown in Ethiopia in sorghum fields as an oil crop. Seeds of both *Cucumis sativus* and *C. melo* are used for food and medicine in parts of Africa and India. There are sketchy reports not listed in Table 1 of other species of *Cucumis* used for seed oil. For example, *C. metuliferus* in Niger, as well as *C. pustulatus* (syn. *C. figarei*) in Uganda may be used for seed oil.

Several genera, including *Fevillea, Hodgsonia* , and *Telfairia*, contain large oil-rich seeds. For example, *Fevillea cordifolia* has seeds 5-6 cm across with 10-15 seeds in a 10-12 cm round fruit. "Thus, on a weight per fruit basis, the seed-oil content...is apparently higher than in any other dicotyledon and among the highest ever reported for any plant" (Gentry and Wettach, 1986). With the exception of *Telefairia occidentalis* , these seeds also have thick, difficult to remove hulls.

There are significant discrepancies in the literature on the oil and protein composition and percentage oil content of many of these poorly known oilseeds. This is presumably due to cultivar and environmental differences or to actual misidentifications. But some species among the genera *Luffa* and *Fevillea*, are reported to contain either edible or poisonous seed oil. This confusion in the ethnobotanical literature may be due to different extraction methods and contaminants from the seed coat. The nutritional value and possible toxicity of cucurbit oilseeds need to be studied further, and the seeds need to be evaluated for suitability for edible oil versus drying oil for industrial purposes, such as paints and varnishes.

Table 1. Cucurbitaceae species that have been exploited for their seeds.

Scientific name	**Major seed production areas**	**Seed and oil uses**	**Notes (annual and monoecious unless otherwise stated)**
Acanthosicyos horridus	southwest coast of Africa	snackseed, almond substitute in confectionary, ground into flour	semi-domesticate, dioecious, xerophytic, perennial
Apodanthera undulata	Southwest U.S. and northern Mexico	potential industrial oil	wild, xerophytic, perennial
Benincasa hispida	Southeast Asia	edible seeds, anthelmintic	domesticate
Citrullus colocynthis	Northern Africa, Middle East, India	baking flour, soup thickener, cooking & frying oil, soap, illuminant, medicinal	wild & semi-domesticate, perennial
Citrullus lanatus	West Africa, Middle East. southern Asia	snackseed, baking flour, soup thickener, cooking & frying oil, soap, illuminant, medicinal	domesticate & wild
Cucurbita spp.	Americas, Eastern Europe, Africa, Asia	snackseed, edible oil, anthelmintic, potential diesel fuel in the xerophytic species	domesticate & wild, some xerophytic and perennial
Cucumeropsis mannii	tropical West Africa	roasted, fermented, powdered or pasted for soup thickener, cooking oil	domesticate
Cucumis melo	East Africa, southern Asia	roasted snackseed, fermented seedcakes, cooking oil, illuminant, medicinal	domesticate

Cucumis sativus	India, France	roasted snackseed, confectionary, cooking oil, illuminant, anthelmintic	domesticate
Ecballium elaterium	Mediterranean, Middle East	industrial oil	wild, monoecious & dioecious subspecies, perennial
Fevillea spp.	Neotropics	illuminant, industrial oil, medicinal oil	wild, dioecious
Hodgsonia macrocarpa	northern India and southern China	raw or roasted seed, edible oil, illuminant	recent domesticate, dioecious, perennial
Lagenaria siceraria	tropical Africa	cooking oil, medicinal oil	domesticate
Luffa spp.	tropical Africa & Asia, Brazil	cooking oil (may be poisonous), medicinal oil	domesticate
Momordica spp.	Paleotropics	cooking oil, illuminant, industrial oil	domesticate
Praecitrullus fistulosus	northern India, Pakistan	roasted seed	domesticate
Telefairia occidentalis	humid West Africa	raw roasted snackseed, fermented, or powdered seed as a high protein flavor enhancer and thickener in cooking, soap or roasted seed, edible oil, cosmetics	domesticate, dioecious, perennial
Telefairia pedata	East Africa	raw or roasted seed, edible oil, cosmetics	domesticate, dioecious, perennial
Thladiantha nudiflora	Southeast Asia	medicinal	wild, dioecious, perennial
Trichosanthes cucumerina	India, tropical Asia	industrial oil, anthelmintic	domesticate, usually dioecious

Literature Cited

1. Earle, F.R. and Q. Jones. 1962. Analyses of seed samples from 113 plant families. Econ. Bot. 16(4):221-250.
2. Gentry, A.H. and R.H. Wettach. 1986. *Fevillea* -- a new oil from Amazonian Peru. Econ. Bot.40(2): 177-185.
3. Zhou, X.L. 1987. A study on the breeding of named kernel pumpkin and its genetic behavior. Acta Hort. Sin. 14(2):115-118.

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Some Comments Concerning the Origin and Taxonomy of Old World Pumpkins

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Various forms of *cucurbita pepo*, many of them pumpkins, are recorded in botanical tomes of Europe beginning in the mid-16th century (6). Of the pumpkins, nearly all appear to be similar to one of two very distinct kinds: the orange-fruited, 20-grooved pumpkins of eastern Canada and U.S.A. and the green-striped or plain green-fruited, heavily 10-ribbed pumpkins of Mexico. There is also a large, slightly ribbed, broad dark green and orange striped pumpkin: the "Colocynthis oblonga variegata" of Fuchs (2) is similar phenotypically to the pumpkins of Europe and Asia Minor of today, including the oil pumpkins. A question concerning this last form is: was is derived directly from North America or was it derived through hybridization resulting from the two North American kinds being grown in close proximity with unchecked pollution, in the plots of plant collectors and/or herbalists?

There are other illustrations in that same work by Fuchs of *C. pepo* plants that appear to me to be the offspring of unchecked pollination among North american forms. Had the striped pumpkin of Fuchs been a relatively true-breeding stock, cold it alone have resulted in the tremendous phenotypic variability observed today among the striped pumpkins of Europe and Asia Minor? I believe that the answer is no, and that these pumpkins are ultimately derived through unchecked cross-pollinations between the two major kinds of North American pumpkins in the Old World. Other striped pumpkins, some quite large and some small, most with an orange background color, had already appeared in 16th century paintings of Europe (11).

Whilst the hull-less or "naked-seed" characteristic of pumpkin is sometimes considered to be a single-gene trait, the inheritance has also been seen as complex and under the control of several genes. I know of no earlier history of the isolation of this trait than the 19th century, as discussed by Teppner (8). A complex mode of inheritance would favor the idea that the hull-less seed trait was derived not so much through mutation as through genetic recombination. A great deal of recombination would be expected to have occurred, of course, if the two major kinds of North American pumpkins had hybridized.

The oil pumpkins are often referred to as *Cucurbita pepo* var. *styriaca* or *C. pepo* var. *citrullina.* Over the past 15 years or so, there has been a trend to differentiate between botanical taxonomy (10) and taxonomy of cultivated plants (9). Whilst the basic unit of botanical taxonomy is the species, the basic unit of cultivated-plant taxonomy is the cultivar.

Botanical taxonomy employs Latin terms and its lowest level is usually the subspecies, though at times is even lower, to botanical variety. The definition below the species level can be done with greatest confidence if based on genetic evidence. Thus, for *Cucurbita pepo*, Decker (1) has recognized three subspecies *C. pepo* spp. *pepo, C. pepo* spp. *ovifera,* and *C. pepo* spp. *fraterna.*

The highest level of horticultural plant taxonomy is usually the "cultivar group", which can be referred to simply as "group", and common terms, rather than Latin epithets, are preferred. The use of the term cultivar-group can be loose and refer to different characteristics. I believe that the greatest usefulness of this term is obtained when the characteristic(s) used for classification are easily recognizable to those who have any dealings with the species while at the same time reflect genetic relationships. Thus I proposed the use of fruit shape, a highly polygenic characteristic familiar to all who have anything to do with*Cucurbita pepo*, to classify the horticultural forms of this species (4,6). The edible-fruited cultivar-groups I named Cocozelle, Pumpkin, Vegetable marrow, Zucchini, Acorn, Crookneck, Scallop, and Straightneck.

The relationship between the botanical classification and the horticultural classification can be observed from genetic evidence (1,3,5,7): the first four groups named above belong to *C. pepo* spp. *pepo* whilst the later four groups belong to *C.*

pepo spp. *ovifera. C. pepo* spp. *fraterna* contains wild forms only.,

Using the botanical and horticultural classifications described above, the oil pumpkins of Europe and Asia Minor, the grooved pumpkins of Canada and the U.S.A. and the ribbed pumpkins of Mexico, could be considered as three sub-groups or market types within *C. pepo* spp. *pepo* Pumpkin Group.

Literature Cited

1. Decker, D.S. 1988/ Origin(s), evolution, and systematics of *Cucurbita pepo* (Cucurbitaceae). Econ. Bot. 42: 4-5.
2. Fuchs, L. 1542. Vienna Codex 11, 120: 479. Austrian National Library.
3. Katzir, N., Y. Tadmor, G. Tzuri, E. Leshzeshen, N. Mozes-Daube, Y. Danbin-Poleg, and H.S. Paris. 2000. Further ISSR and preliminary SSR analysis of relationships among accessions of *Cucurbita pepo.* In: N. Katzir and H.S. Paris, eds. Proceedings of Cucurbitaceae 2000, the 7th Eucarpia Meeting on Cucurbit Genetics and Breeding. Acta Hort. 510: 433-439.
4. Paris, H.S. 1986. A proposed subspecific classification for *Cucurbita pepo.* Phytologia 62: 133-138.
5. Paris, H.S. 1996. Summer squash: history, diversity, and distribution. HortTechnology 6:6-13.
6. Paris, H.S. History of the cultivar-groups of *Cucurbita pepo.* Hort. Revs. 25: (in press).
7. Robinson, R.W. and D.S. Decker-Walters. 1997. Cucurbita. CAB International, New York.
8. Teppner, H. 1999. Notizen zur Geschichte des Kurbisses. Obst, We in, Garten (Graz) 68(10): 36
9. Trehane, P.,. C.D. Brickell, B.R. Baum, W.L.A. Hetterscheid, A.C. Leslie, J. McNeill, S.A. Spongberg, and F. Vrugtman. 1995. International code of nomenclature for cultivated plants. Quaterjack, Wimborne, U.K.
10. Voss, E.G., H.M. Burdet, W.G. Chaloner, V. Demoulin, P.Hiepko, J. McNeill, R.D. Meikle, D.H. Nicolson, R.C. Rolina, P.C. Silva, and W. Greuter. 1983. International code of botanical nomenclature, in: F.A. Stafleu, ed. *Regnum vegetabile* 111. Utrecht.
11. Zeven, A.C. and W.A. Bradenburg. 1986. Use of paintings from the 16th to the 19th centuries to study the history of domesticated plants. Econ. Bot. 40: 397-408.

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The Origin and Breeding of the Hull-less Seeded Styrian Oil-Pumpkin Varieties in Austria

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Abstract: The cultivation of pumpkins (*C. pepo* L.) for the combined use of fruits and seeds in Styria is confirmed by records going back 200 years. This was the first and, at the beginning, the largest region in Europe that cultivated pumpkins for making seed oil.

Originally the hard seedcoat had to be removed to make good oil. A mutation having hull-less seeds seems to have appeared around 1880, but reports of cultivation first appeared around 1925. The quality of oil in the seeds and the vining form of growth did not change. For a long time farmers had selected for larger fruits, larger seeds and higher oil content and exchanged seed between regions. Based on local plant material systematic breeding activities and genetic studies were started in the 1930s.

Besides disease resistance and improvement of seed yield, reduction in the length of vines, an increase of the oil content in the seeds and - recently - an increase of tocopherols are the main goals in breeding work today.

History: Cultivation of vining Styrian-type oil pumpkins in Austria has been documented over the past 200 years. Because of the favourable climatatic conditions Styria was the main oil pumpkin producing and consuming area in Austria from the very beginning. Other countries that traditionally produced oil from pumpkin seeds are Hungary, Northern Yugoslavia, Romania nd the Ukraine.

Originally the same types of pumpkin were used for producing fruit flesh and seeds for pressing oil. Selection practised by farmers was influenced by both of these uses.

In the Styrian cultivation area, which was the oldest and largest in central Europe, farmers had ben practicing selection for at least 100 years (1). Usually farmers exchanged seeds or used seeds from the best production areas. Some of these selections could not adapt to other regions, but others proved suitable for various conditions and were sufficiently stable. For seed production the first fruits set, the largest fruits, and those containing the most and largest seeds were chosen. Attention was also paid to uniformity of shape and colouring of the fruit. fields for growing pumpkins were usually fertilised with manure. This practise favoured types for intensive cultivation. The experience gained from pressing oil from seed dehulled by hand enabled farmers to select seed for higher oil content as well. Thus landraces were continually being improved in the traditional oil pumpkin regions.

A mutation to pumpkins with hull-less seeds seems to have occurred in Styria around 1880. Reports of the cultivation of such hull-less-seeded pumpkins in Austria began to appear after the first World War. Cultivation of the hull-less-seeded pumpkins increased rapidly because it was now possible to produce more oil (Fig. 1 and 2) with less work and less expense. The oil from hull-less seeds was equal in quality to that of the hulled seeds (3).

To optimise cultivation techniques Professor Tschermak from the University of AGriculture in Vienna developed a bush-type hull-less seeded pumpkin, the so-called Tschermak's oil-pumpkin, by crossing the Styrian vining hull-less oil-seed pumpkin with the hulled-seeded bush-type squash 'Mark Marrow' (4). This variety, 'Tschermaks oil pumpkin' was registered at the Federal Institute in Vienna in 1955. Tschermak chose the bush form so that the fruits could mature more rapidly and uniformly and facilitated mechanical weed control. But his type produced smaller fruits and smaller seeds so the farmers still preferred the Styrian hullless vining pumpkin which they themselves had selected.

Breeding to increase the oil content of hull-less seeds of the Styrian vining oil pumpkin was begun in 1940 at the Lamberg Breeding station (2) near Ilz in the eastern part of Styria. In 1948 Prof. Gorbach of the Technical University in Graz initiated analyses of oil and protein content of seeds originating from elite plants.

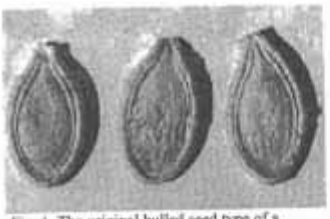

Fig. 1. The original hulled seed type of a landrace, cultivated in Styria years ago

Fig. 2. The typical hull-less seeds of the variety "Gleisdorfer Ölkürbis"

Fig. 3. Mature Fruits of "Gleisdorfer Ölkürbis"

Saatzucht Gleisdorf started its oil pumpkin breeding program on a small scale already in the first year after its foundation in 1947. Collections of landraces were made from different parts of Austria, mainly in the south-eastern Styria and in Slovenia. In 1960 the breeding program was enlarged. A number of lines were evaluated with respect to seed yields, excellent performance, and high oil content in the seeds. From this plant material the first vining hull-less oil-seed pumpkin variety of Austria based on local material was selected and registered under the name "Gleisdorfer Olkurbis" in 1970. Even today this is the most widespread oil-seed pumpkin variety in Austria and in 1995 the "Glei8sdorfer Olkurbis" was also registered in Hungary.

The typical "Gleisdorfer Olkurbis" has very long vines of 8-10 m in length, globular yellow fruits with green streaks weighing 3 to 7 kg. at maturity, and the oil content of its seeds is about 50%. Fig. 3 shows fruits at maturity.

The variety 'Weis 371' was developed at the Research Station for Special Crops in Wies, Styria and registered in 1976. This variety ripens a few days later than the Gleisdorfer Olkurbis and has a different shape of leaf. In 1998 the maintenance breeding was taken over by Saatzucht Gleisdorf.

Over the last 20 years breeding efforts have been concentrated on increasing the size, thickness and harvestibility of the seeds as well as improving the ratio of seed to fruit weight. In some selections an increase of dry seeds compared to fresh fruit weight from 1.5% to approximately 3.0% could be achieved.

In order to reduce the variable maturing time of fruits we started to develop bush-type strains with short vines, smaller fruits and more fruits per plant. Until now two oil pumpkin varieties of this type have been registered, the first is 'Sepp' (1994) with dark green seeds and marbled fruits and 'Markant' (1996) with light green seeds and striped fruits.

In a national research project financially supported by the Funds for Promotion of Research (FFF) with the title "Breeding of early maturing oil-seed pumpkin varieties of high quality" we started in 1995, to analyse, in addition to oil content, the fatty acids and tocopherol content in our breeding material (5, 6_. Table 1 shows the level of oil, linoleic acid and tocopherols content of 2 varieties at the location Gleisdorf over a period of 5 years. 'Sepp' is the variety with the higher oil and linoleic acid content. Due to the first serious virus outbreak in 1997 and also in 1998 the oil seed pumpkins showed strongly reduced seed size and as a consequence a reduced oil content in the seeds.

The years 19195 and 1996 with low temperatures in September affected linoleic acid contents for both varieties positively.

The first 3 years showed a constant level of tocopherols in both varieties. Since 1997 and 1998 were years with high virus infections in the trials, a substantial increase in tocopherol content was observed (Table 1), however, we did not find the negative correlation between tocopherol content and content of linoleic acid as described previously (7).

High virus infections 9ZYMV,WMV-2) occurred for the first time in Austria in 1997. We at Saatzucht Gleisdorf started a resistance breeding program, financially supported by the FFF and in cooperation with the Research Project for ZYMV-Resistant Oil-Seed Pumpkins for Austria co-ordinated by T. Lelley at IFA-Tulin.

Table 1. The level of oil, linoleic acid and tocopherols content of two varieties, Sepp and Gleisdorfer Olkurbis at the location Gleisdorf over a period of 5 years.

	Oil content (%)		Linoleic acid (%)r		a + ? Tocopherol (mg/kg)	
Year	**Sepp**	**Gleisd.**	**Sepp**	**Gleisd.**	**Sepp**	**Gleisd**
1994	50,5	46,5	43,4	42,3	136	149
1995	49,7	47,8	55,3	54,4	134	147
1996	43,0	41,1	55,4	50,2	119	144
1997	45,7	46,9	51,9	46,5	96	339
1998	42,6	46,9	47,6	44,8	202	327

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Literature Cited

1. Buchinger, A. 1948. Kurbiszuchtung. Die Bodenkultur, 2:10-27.
2. Buchinger, A. 1950. Der Steirische, schalenlose, langtriebige Olkurbis. Die Bodenkultur, 4:217-226.
3. Grebenscikov, I. 1954. Zur Vererbung der Dunnschaligkeit bei *Cucurbita pepo* L. Zuchter 24:162-166.
4. Schoninger, G. 1950. Genetische Untersuchungen an *Cucurbita pepo* L. Der Zuchter 20:321-336.
5. Murkovic, M., A. Hilebrand, J. Winkler, W. Pfannhauser, 1996. Variability of vitamin E content in pumpkin seeds. (*Cucurbita pepo* L.) Z. Lebensm. Unters. Forsch., 202:275-278,
6. Murkovic, M., A. Hillebrand, J. Winkler, E. Leitner, W. Pfannhauser, 1996. Variability of fatty acid content in pumpkin seeds (*Cucurbita pepo* L.) Z. L Kebensm. Unters. Forsch. 203:216-219.
7. Schuster, W., W. Zipse, R. Marquard, 1983. Der Einfluß von Genotyp und Anbauort auf verschiedene Inhaltsstoffe von Samen des Olkurbis (*Cucurbita pepo* L.) Fette, Seifen, Anstrichmittel 85:56-64.

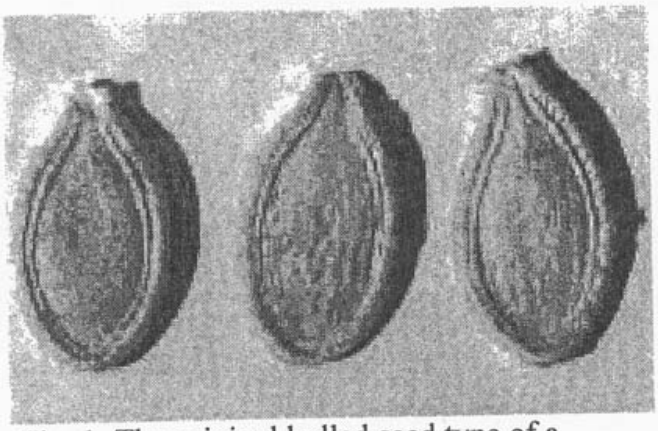

Fig. 1. The original hulled seed type of a landrace, cultivated in Styria years ago

Fig. 2. The typical hull-less seeds of the variety "Gleisdorfer Ölkürbis"

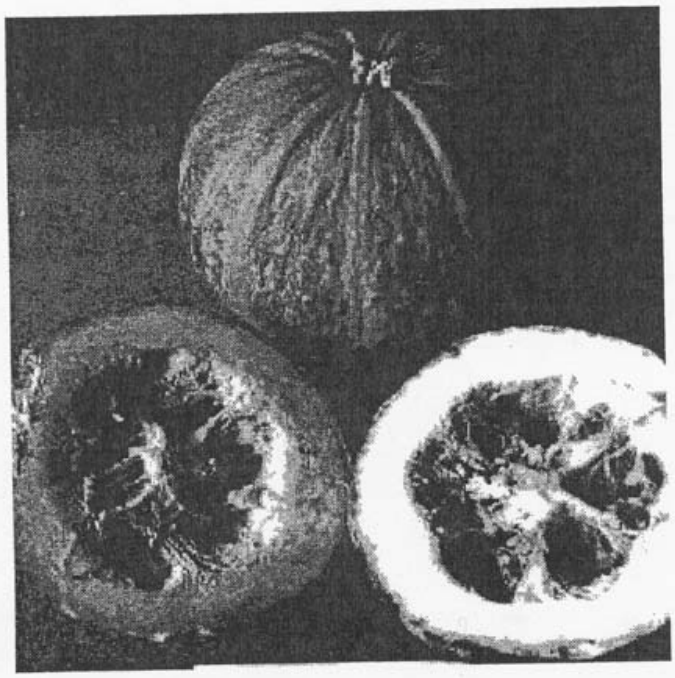

Fig. 3. Mature Fruits of "Gleisdorfer Ölkürbis"

Cucurbit Genetics Cooperative Report 23:105-? (article 35) 2000

Breeding, Production, and Utilization of Oil Pumpkin in Yugoslavia

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Abstract: The classification, breeding (history, methods and breeding goals), production (including the traditional practice of intercropping along with the present way of production in pure culture) and utilization (whole fruit or the flesh of the fruit for cattle feed and the seeds for snack, oil production or medicinal purposes) of oil pumpkin (*Cucurbita pepo* L.) in Yugoslavia are outlined in this paper. The breeding goals (general plant characteristics, seed coat type, seed characteristics, fruit characteristics, resistancy to diseases) as well as the pharmaceutical aspects (in relation to benign prostate hyperplasia) are elaborated.

Key words: *oil pumpkin, breeding, naked-seeded, hulled, production, utilization, snack, oil, benign prostate hyperplasia*

Classification: The most common cucurbits grown in Yugoslavia, in order of their decreasing economic importance, are: cucumber, watermelon, cantaloupe, *Cucurbita* species, mostly used as summer squash (*C. pepo*), or winter squash (*C. maxima, C. moschata, C. ficifolia* and *C. mixta*) and the gourd (*L. siceraria*) as a vegetable (7). The fruits of *C. pepo, C. maxima* and *L. siceraria* are popular decorations. The attention paid to chayote *Sechium edule* (Jacq.) Sw. as a new vegetable is increasing, and *Luffa* sp. is becoming also popular. Oil pumpkin, belonging to *C. pepo* is mainly grown or seed. The utilization of the seed is determined mostly by its oil content (9). Recently demand has been shown for *C. maxima* seeds for the same purpose as *C. pepo* oil pumpkin.

Breeding: *History of breeding.* Landraces of oil pumpkin, without a specific name, maintained by farmers and characterized by hulled seeds, elliptic, dark green fruits with an orange spot on the surface of the fruit touching the soil prevailed in production until recently. The naked-seeded oil pumpkin was almost completely unknown, except for the occasional spontaneous mutations resulting mostly in half-hulled seeds, considered more as an attraction without a recognized economic importance. It was not until the early 1980's that naked-seeded cultivars entered the production, and hulled-seeded cultivars did even later.

The only active oil pumpkin breeding program in the country is located at the Institute of Field and Vegetable Crops in Novi Sad. As the result of more than two decades of research and development in oil pumpkin genetics and breeding (4), during which several successful collaborations have also been established (i.e. with DUDU Bt. Debrecen, Hungary), the naked-seeded oil pumpkin cv. 'Olinka' has been officially registered in Yugoslavia in 1992 and the hulled-seeded cv. 'Olivia' in 1997. consequently both cultivars were registered in Hungary (1995) and in Slovenia (1999) as well.

Breeding methods: Mass selection and individual selection (occasionally with overstored seed) based on local populations or populations obtained by crossing have ben applied (Fig. 1). The investigation of heterosis showed positive hybrid vigor for most of the economically important traits of the hull-less seeded oil pumpkin (1). Ethephon treatment as a possible tool for commercial hybrid seed production was also successfully tested (2).

Breeding goals: The most important current and future breeding goals are: (a) general plant characteristics (the semi-bush growth habit characteristic for the cv. 'Olinka' favored over vining stem typical to 'Olivia' shown on Fig. 2, growth energy, number of fruits per plant), (b) seed coat type (naked-seeded without any seed coat development at the seed margins as well as the typical hulled-seed type are equally involved as shown on fig. 3, but the potential of a half-hulled or partially-hulled type is also under consideration), (c) seed characteristics (dimensions, mass, color, ease of dehulling, taste, chemical composition i.e. oil content, irregular seed development, ease of separating the seed form the fruit called harvestability), (d) fruit characteristics (fruit dimensions, shape and weight, thickness and mass of flesh, fruit color, fresh as well as dry seed mass per fruit), (e) resistance to diseases (anthracnose and viruses) (3).

Fig. 1. Part of a hulled-seeded oil pumpkin population for further selection obtained by crossing.

Fig. 2. The semi-bush growth habit of the naked-seeded cv. 'Olinka' as compared to vining type of the hulled-seeded cv. 'Olivia'.

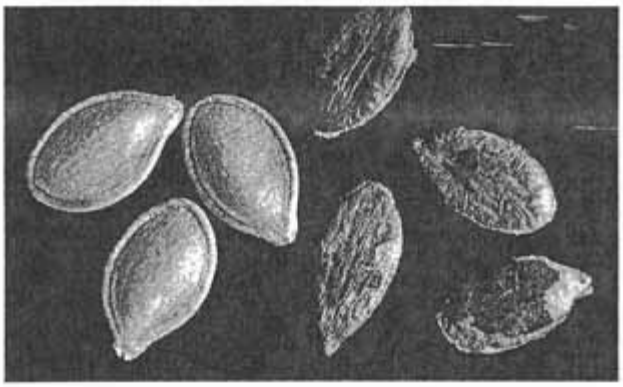


Fig. 3. Hulled- as compared to naked-seeded oil pumpkin seeds.

Detailed correlation analysis has been carried out between the more important fruit and seed characteristics from the breeder's point of view (to be presented at the VIIth EUCARPIA Meeting on cucurbit Genetics and Breeding "Cucurbitaceae 2000," Israel, March 19-23, 2000.

Production: *Intercropping.* The oil pumpkin has been traditionally intercropped with corn (11). Pumpkins were harvested at the same time as corn, carefully transported to the barnyard a d occasionally fed to the cattle long into the winter. This could be called forage, rather than oil pumpkin. Later more and more attention was paid to the seeds which were removed, dried and collected for further use. Intercropping is still practiced in some parts of Yugoslavia. It is hard to estimate the total area of corn intercropped with oil pumpkin but it is certainly still above 10.000 ha. Intercropping of oil pumpkin in corn is again gaining some interest from the point of view of sustainable agriculture (10).

Pure culture. The contemporary practice of oil pumpkin growing is in pure culture (5, 8). This kind of production could be estimated to an annual 1.500-2.000 ha. The growers are mostly small farmers usually with 0,5-1 ha of pumpkin. The crop is mostly planted by machine,. Instead of herbicides (usually Trifurcating) mostly mechanical weed control is practiced. Neither chemical treatment against diseases (fusarium root rot, powdery mildew, anthracnose, viruses) or insect pests (aphids) is applied, nor is mineral fertilization widespread, which fulfills completely the requirements of organic production. Around 3/4 of the production is hand harvested separating seeds from the previously halved fruits, but home-made harvesters are also used. The prototype of a new construction keeping the flesh clean after the removal of the seed, is a prerequisite for the further utilization of the flesh, is being tested, At present practically all the flesh is discarded after the harvest. The removed seed is usually dried in the sun. The dried seed is rarely stored, mostly sold immediately to the seed traders, roasters or oil mills.

Lack of complete fertilization could be a problem, resulting in "empty" seeds of the hulled-seeded oil pumpkin. Enhancement of fertilization by placing bee hives near the pumpkin fields is recommended. Seed set and seed fill was considerably improved by Boron (i.e. Solubor DF (R)) treatment. The effect of the Ethephon (proposed for commercial hybrid seed production) on the shortening of the internode length as well as improvement of fruit set was also observed.

The favorable effect of planting corn rows at some 5-10 m distance from each other along the oil pumpkin field has been observed. The resulting microclimate obviously stimulates the pumpkin plants and prevents the outbreak of diseases. Rodents are attracted by the corn cobs, thus preventing damage to the pumpkin fruits.

Utilization:Cattle feed. The type of the plant called hulled-seeded oil pumpkin in this paper was traditionally used for cattle feed (6). Often the whole fruit was fed to the cattle, but sometimes only the flesh after removal of the seeds was used for feeding. To provide fresh fruits (sometimes cut into pieces and cooked in water right before feeding) for the cattle (mainly dairy cattle but also for swine and horses), the fully ripe fruits were stored in a frost-free place where they remained basically undamaged during the whole winter. Studies showed that pumpkin,mixed with corn stalk, could be a valuable raw material for silage production.

At present, the seeds are the main product with very few cases of fresh utilization.

Snack. The seed of oil pumpkin is traditionally used as a homemade snack, especially the hulled-seeded oil pumpkin. Not only the oil, but also the protein content of the seed is important in this respect, giving the roasted seed a special, pleasant taste. There is a growing number of small companies specializing in roasting and packing seed for snacks. Dehulling the roasted pumpkin seed (with the teeth) is a very common pastime while watching a football game, TV or a movie, as well as traveling on public transportation vehicles.

Pumpkin seed oil. Oil production is a relatively new use of pumpkin seed,s hull-less seed being preferred to hulled in this case. The oil is produced by and is a popular item offered by natural food stores mainly as a delicious and healthy salad oil.

Medicinal use. The pharmaceutical aspect of the oil pumpkin seed and seed oil is related to the benign prostate hyperplasic (BPH). BPH is not a life-threatening disorder, but can substantially reduce the quality o0f the patient's life. The therapy includes herbal prostate drugs, phytomedicines. Unlike in the USA< in Europe the use of prostate gland drugs derived from plants including the oil pumpkin is very widespread. Among the herbal medicines *Cucurbitae peponis semen* have the longest tradition in successful BPH treatment in Europe going back to the 18th century. The pharmaceutical effect of the pumpkin-seed oil is attributed to the delta-7-sterols, amino acids, and selenium (13). The results of a clinical study of the effect of the seed oil deriving from the naked-seeded oil pumpkin cv. 'Olinka" clearly showed its prostatotropic activity (lowered amount of residual urine and enhance micturation speed) in comparison to an untreated control group of patients with BPH (12).

Literature Cited

1. Berenji, J. 1988. Hibridna snaga kod uljane tikvegolice, *Cucurbita pepo* L. Uljarstvo 23(3-4):79-85.
2. Berenji, J. 1988. Poznavanje tikava, Cucurbita sp. Zbornik referata "3. Poljedelski dnevi ABC Pomurka", Murska Sobota.
3. Berenji, J. 1989. Ciljevi oplemenjivanja uljane tikve, *Cucurbita pepo* L. Zbornik radova Savetovanje proizvodjaca Biljnih ulja i masti Jugoslavije, Beogrd, p. 134-135.
4. Berenji, J. 1992. Tikve. Bilten za Hmelj, sirak i lekovito bilje 23-24(64-65):86-89.
5. Berenje, J.1994. Uljana tikva. In.: Tehnologija proizvodnje lekovitog, aromaticnog i zacinskog bilja. Institut za ratarstvo i povrtarstvo Novi Sad i Agroseme-Panonija Subotica.

6. Berenji, J. 1995. Stocna tikva - zaboravjljene krmna kultura. Zbornik radova Instituta za ratarstvo i povrtarstvo Novi Sad 23:529-537.
7. Berenji, J. 1999a. Tikve - hrana, lek i ukras. Zbornik radova Instituta za ratarstvo i povrtarstvo Novi Sad, 31:63-75.
8. Berenji, J. 1999b. Proizvodnja i koriscenje uljane tikve (*Cucurbita pepo* L.). Zbornik radova 40. Savetovanja o proizvodnji i preradi uljarica, p. 303-308.
9. Berenji, J. and Gy. Karlovits. 1995. Az olajtok szarmazasa, rendszertana es alaktana. Olaj, szappan, kozmetika 44(2):54-59.
10. Momirovic, N., S. Oljaca, G. Vasic, D. Kovacevic and Z. Radosevic. 1998. Effects of intercropping pumpkins (*Cucurbita maxima* DUCH.) and maize (*Zea mays* L.) under different farming systems. Proceedings of the 2nd Balkan Symposium on field Crops, p. 251-254, Novi Sad.
11. Popovic, M. 1971. Prilog poznavanju kulture tikava (Cucurbita sp.) u nas. Savremena poljoprivreda 19(11-12):59-72.
12. Sabo, A., J. Berenji, J. Stojkov and J. Bogdanovic. 1999. Phamacodynamic effect of pumpkin seed oil (Oleum cucurbiaceae pepo) in patients with adenoma prostate. Abstracts of the 2nd European Congress of Pharmacology, Budapest, 3-7, July 1999, p. 360 s,
13. Schiulcher, H. 1986. *Cucurbita*-species, Zeitschrift fur Phytotherapie 7(1):19-23.

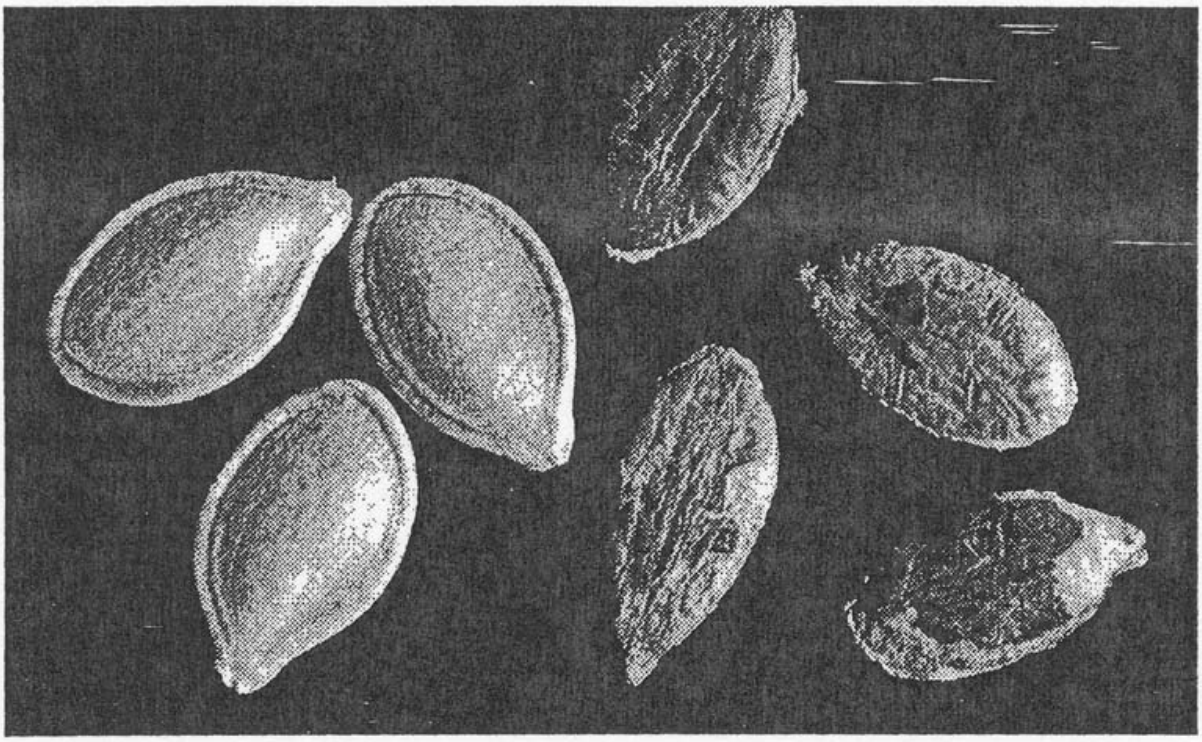
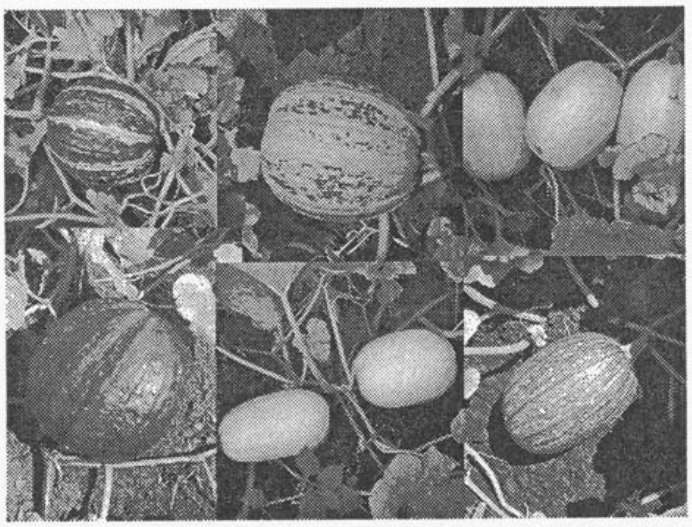


Fig. 3. Hulled- as compared to naked-seeded oil pumpkin seeds.



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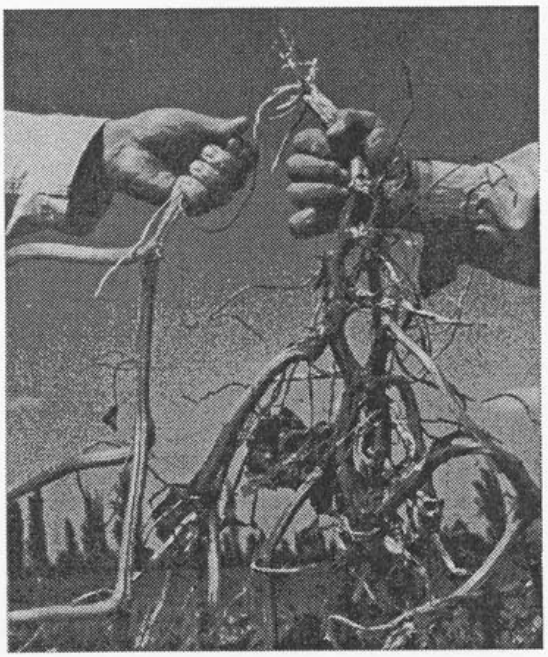

Fig, 2. The semi-bush growth habit of the naked-seeded cv. 'Olinka' as compared to vining type of the hulled-seeded cv. 'Olivia'.

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Oil Seed Pumpkins - A New Experience for New Zealand

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Abstract: Oil seed pumpkin, (*Cucurbita pepo* var. Styriaca), cv. Wies 371 was first evaluated in new Zealand in 1991 and subsequent trials indicated that the crop grew well in areas where traditional cucurbit crops are produced in both the North and South Islands. Commercial attempts to establish the crop failed initially through lack of interest and experience but were successful in 1999. Seed yields were very low but can be improved by using fresh seed stocks and by minimising the virus infections.

Introduction: For 30 years I did research for the Ministry of Agriculture and Fisheries, followed by 8 years working for the Crop & Food Research Institute concentrating on the introduction of new crops to New Zealand (1). Presently I work as a private consultant with an emphasis on new crop introduction.

New Zealand growing areas of squash and pumpkins are mainly in the northern and eastern parts of the North Island, with some squash produced also in the top and middle half of the South Island. Oil seed pumpkins could grow in any region where cucurbits are being grown successfully.

The oil seed pumpkin, (*Cucurbita pepo* var *Styriaca*), cv. Weis 371, was first evaluated as one of the 60 new crops introduced in 1991. As with numerous crops that are unfamiliar to the average New Zealander, it has taken many years for the oil seed pumpkin to become accepted. This slowness is often attributed to the patience required of growers while learning unfamiliar methods of growing or processing and the lack of consumer interest while the crop is being introduced. In addition, the harvesting and general processing methods for a new crop often prove to be very expensive and it is generally quicker to revert back to suppliers from well established areas so the costs can be kept down. Once these problems have been overcome, however, it is usually recognised that New Zealand is in an excellent position to offer a good quality products. Our market is also well suited for offering organically produced crops and well su8ited as a niche market situation, which is often the case for example in Japan,which has become more health conscious over recent years.

Last season was the first time that oil seed pumpkins were grown on a small commercial scale in Hawke's Bay, on the east coast of the North Island. It was quite clear that there are still many problems associated with the harvesting and drying of the seed. Despite these problems, the crop's products were very well received, which encourages ongoing development.

What was done with the crop? Crop & Food Research evaluated cv. Weis 371 for seed quality and spacing effects in Hawke's Bay, Waikato and south of Aukland, all areas in the North Island. In 1991 some commercial firms showed interest but development was slow until last year, when the first serious commercial cropping started. Some of the seed stock was nearly seven years old and this resulted in a low germination and very uneven plant growth and fruit maturity, which in turn resulted in low seed yield and quality.

Last year 'Weis 371' was used in trials designed to improve the seed yield. Some pilot trials were done with composted rock phosphate compounds and fish oil as a nutrient to increase the seed yields and the seed size.

Results. *Spacing trials.* Plant densities varied from 1 to 2.5 plants/m^2 , in rows 1.5 m apart. Mean fruit weight, seed weight/fruit and the individual seed size declined as the plant density was increased resulting in no increase in the seed yield/m^2 There was a positive relationship between fruit weight and seed yield. It is recommended that the oil seed pumpkin crops for seed be planted at 1 to 1.5 plants/m^2 rather than 1.5-2/2 plants/m^2 as recommended for squash fruit production. Estimated seed yields varied from 1 to 1.4 t/ha/ The mean oil content of the pumpkin seed was 40.4% with over 90% of the oil comprising of three fatty acids, linoleic (52.7%), oleic (28/3%) and palmitic (12.7%) acids (2).

Virus infection levels cause concern. The high levels of virus infection in the seeds, caused by WMV2, (204%) and ZYMV, (0.6-1.4%) may have caused higher than normal visual field symptoms of around 20% to 25% in the plants during the growing season, which is considered too high for our normal squash and pumpkin crops. As further plantings may be planned for next season, it is important that the seed stock is free from viruses and imported seed should be verified virus free. The reason for this concern is simple: considerable amounts of money and effort have been expended by the present squash exporting industry to minimise the virus infection levels in their crops. Thus it would be highly irresponsible to introduce a known source of virus infected seed crop.

Fertiliser trials. The initial trials using the composted rockphosphate and fish nutrient showed that both the seed size and seed quality could be increased but further tests will be carried out during the 1999/2000 season to confirm this.

Discussion: Until last 1999 the commercial acceptable of oil seed pumpkins was very slow as the harvesting and drying techniques were still inadequate. The market awareness of the product is not yet strong enough to make oil pumpkin production viable. This is changing from the New Zealand perspective, the oil seed pumpkin products are better known now and there is also a growing awareness of organically produced crops. the processing technology still remain a serious problem but this can be overcome.

The trial results indicated that oil seed pumpkins can grow in areas in New Zealand which successfully grow cucurbits. Whether or not oil production is commercially viable remains unanswered but the oil composition is similar to the overseas oils which have been used as a culinary or medicinally product.

The market potential for dry seeds is very good and there is a growing market in the USA and Asia not only for the traditional seed uses but also for new products that can be developed, such as confectionary lines. We have also found interest in new culinary and medicinal uses. New Zealand should be seen by the Austrian traditional industry as a complementary rather than a competing market and we would like to encourage more research and commercial cooperation.

The virus infection was more serious than originally thought and it was higher in the oil seed crops than in the other cucurbit crops during the 1999 season, which may be entirely a seasonal coincidence and should not be regarded as a trend. The seed yields were too low and for the crop to be viable these yields need to be increased substantially. The fertilizer treatments applied last season may help to achieve this goal.

In conclusion note the following points:

- Variety evaluation of new (virus free) types are needed in New Zealand
- Efficient mobile harvesting and drying equipment is needed
- Virus infection needs to be minimised
- Explore and promote new products from the seed

Literature Cited

1. Burgmans, J.L,, J.J.C. Scheffer and J. Follett. 1996. Naked oil seed pumpkin, Crop & Food Research broadsheet 70.
2. Douglas, J.A., J.L. Burgmans, J. Follett, J.J.C. Scheffer and R.A. Littler. 2000. The seed yield of oil seed pumpkin at four plant populations and the oil composition of the seed (prepared for publication).

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Virus Infections Levels of Oil Seed Pumpkins in New Zealand

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Abstract: Small commercial plantings of the oil seed pumpkin, 'Weis 371' and an unknown Hungarian variety, were made in Hawke's Bay, new Zealand during November and December of 1998. The first experiments with 'Weis 371' had started in 1991. The Hungarian seeds were tested in a glasshouse and showed 2-4% WMV2 and 0.6-1.4% ZYMV infection. Visual inspection of 'Weis 371' revealed a 20% infection early in the season which in February 1999 increased to 25%.. No virus symptoms were visible on the fruit but the foliage infection of oil seed pumpkins was often higher than in the neighbouring fields of pumpkin and squash.

Introduction: Seed of the variety Weis 371 was first introduced into New Zealand in 1991 and various plantings for research purposes have been carried out since that time. In 1998 small commercial plantings of 'Weis 371' and another variety, (of Hungarian origin) were carried out in Hawke's Bay.

Seed of the later variety was tested for virus presence in a glasshouse before planting out, but no 'Weis 371' seed has been tested. During trials since 1991 no visual virus infections were detected on the plants as has been the case with squash variety trials.

Results of virus checks: The seed of Hungarian oil seed pumpkins had a 2-4% WMV2 and 0.6-1.4% ZYMV in a glasshouse test. The crop had a 12% level of visual infection (on foliage) in January 1999. (estimate from 100 plants).

'Weis 371' had 20% visual infection in 1999. Close to the two experimental blocks were small areas of zucchini and watermelon, which showed no sign of virus infection. In February, 1999 these two blocks had an infection of 32% in the Hungarian line and 25% infection in 'Weis 371'. Only WMV2 was confirmed in February. No virus symptoms were noted on the fruit in the wy of distortion or blistering as is often seen on squash crops or other pumpkins. By February there was some virus infection in the zucchini crop.

Other squash crops were monitored in the district over the season. Apart from two crops, one early squash and a late crop which had 20% and 30% visual infection respectively, most other crops had a very low infection. the 1998/1999 season was a very mild one with respect to virus infections since most crops were sown after the main aphid activity which was cut short by high temperatures in November.

Over the years the virus had been not detected visually but it may occur during very early plant growth in squash, tomatoes, peppers and broad beans, especially when the virus is seed borne. It is possible that the plant is providing some kind of mechanism to mask or minimise virus infection as is the case with some other crops.

The high levels of virus infection of the seeds, caused by WMV2, (2-4%) and ZYMV, (0.6-1.4%) may have been the reason for the higher than normal visual field symptoms of around 20% to 25% in the plants during the growing season. This is considered too high for our normal squash and pumpkin crops. As further plantings may be planned for next season it is important that the seed stock be free from virus infection and imported seed should be certified virus free.

Some observations are:

- Virus infection is not always visible during early plant growth
- The high level of virus in oil seeds could be due to seed borne viruses
- Other virus sources are present in the surrounding weeds
- Damage to squash crops will result eventually if this high level is not reduced
- There is an urgent need for virus free foundation seed or resistant plants

Literature Cited

1. Fletcher, J.D. 1996. Zucchini yellow mosaic virus in buttercup squash - a new record in New Zealand, Australian Plant Path 25:142.
2. Fletcher, J.D., H.M. Nott, A.R. Wallace, B.T. Rogers and T.J.B. Herman 1998. Potyviruses in New Zealand Buttercup squash. (*Cucurbita maxima*). 9th Conference International working group on vegetable viruses, Turin, Italy.
3. Fletcher, JD (1995); Mosaic viruses of squash. Crop & Food Research broadsheet 66.

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Zucchini Yellow Mosaic Virus in *Cucurbita pepo* var. *styriaca*: Epidemiology, Strategies of Control

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In 1997 a severe outbreak of Zucchini Yellow Mosaic Virus (ZYMV) (3) caused serious damage and yield losses in oil pumpkin, *cucurbita pepo* var. *styriaca* all over austria. In 1998 and in 1999 the economic impact of the disease was lower, nevertheless the virus was againfound in oil pumpkin crops all over the country. From infected oil pumpkins the virus was transmitted to melons, cucumbers and zucchini and severely ffected the harvest.

In *Cucurbita pepo* var. *styriaca* symptoms usually become visible in the middle of June 3-4 weeks after germination. On leaves severe yellowing and mottling, mosaic symptoms, vein banding, or vein clearing, darker green blistersm deformations and more or less intense reductions of size can be observed (Fig. 1). Shoots are stunted and stand out from the crop. Early infected plants develop no or very small fruits, later infecttions lead to malformations and to reduced fruit size (Fig. 2). Infections before flowering period may diminish the number of pistillate flowers.

In concurrence with results obtained for *Cucurbita maxima* (2) preliminary studies by our institute indicate that oil pumpkin seed from old fruits does have the potential to act as a disease reservoir between seasons. Virus tests of weeds showed latent ZYMV infections in a few cases. Infected weeds, however, were only observed in or adjacent to ZYMV-infected cucurbit crops. No evidence for over-wintering of ZYMV in perennial weeds has been found. Thus it must be presumed that at present infected oil pumpkin seed plays the most important role for over-wintering of ZYMV in Austria.

From a few primary infection sites the virus spreads rapidly to the whole crop. During mechanical weed control plants are wounded thus transmitting the virus from plant to plant. Usually only a few aphids can be observed in oil pumpkins. Nevertheless it must be presumed that aphids from neighboring crops play an important role as virus vectors both within and between fields. Sometimes virus particles are also carried by vertebrates like deer or rabbits.

Several measure are necessary to control ZYMV in oil pumpkin crops and to reduce economic losses. The production and the exclusive use of healthy seed are imperative.

In order to reduce mechanical virus transmission mechanical weed control can only be carried out as long as the plants are not in contact with the working equipment.

In oil pumpkins virus sources can normally be found within the crop, the virus is not brought in from the outside. Thus, seed treatment with imidacloprid preventing aphid multiplication on the pumpkin plants has been tried through its influence on the non-persistent virus transmission is controversial (1). Its effect on spread of ZYMV in *cucurbita pepo* var. *styriaca* cannot yet be estimated. It is, however, obvious that a partial use of treated seed does not reduce virus spread. In this case virus contaminated aphids from neighboring fields transmit the virus into the insecticide-treated crop.

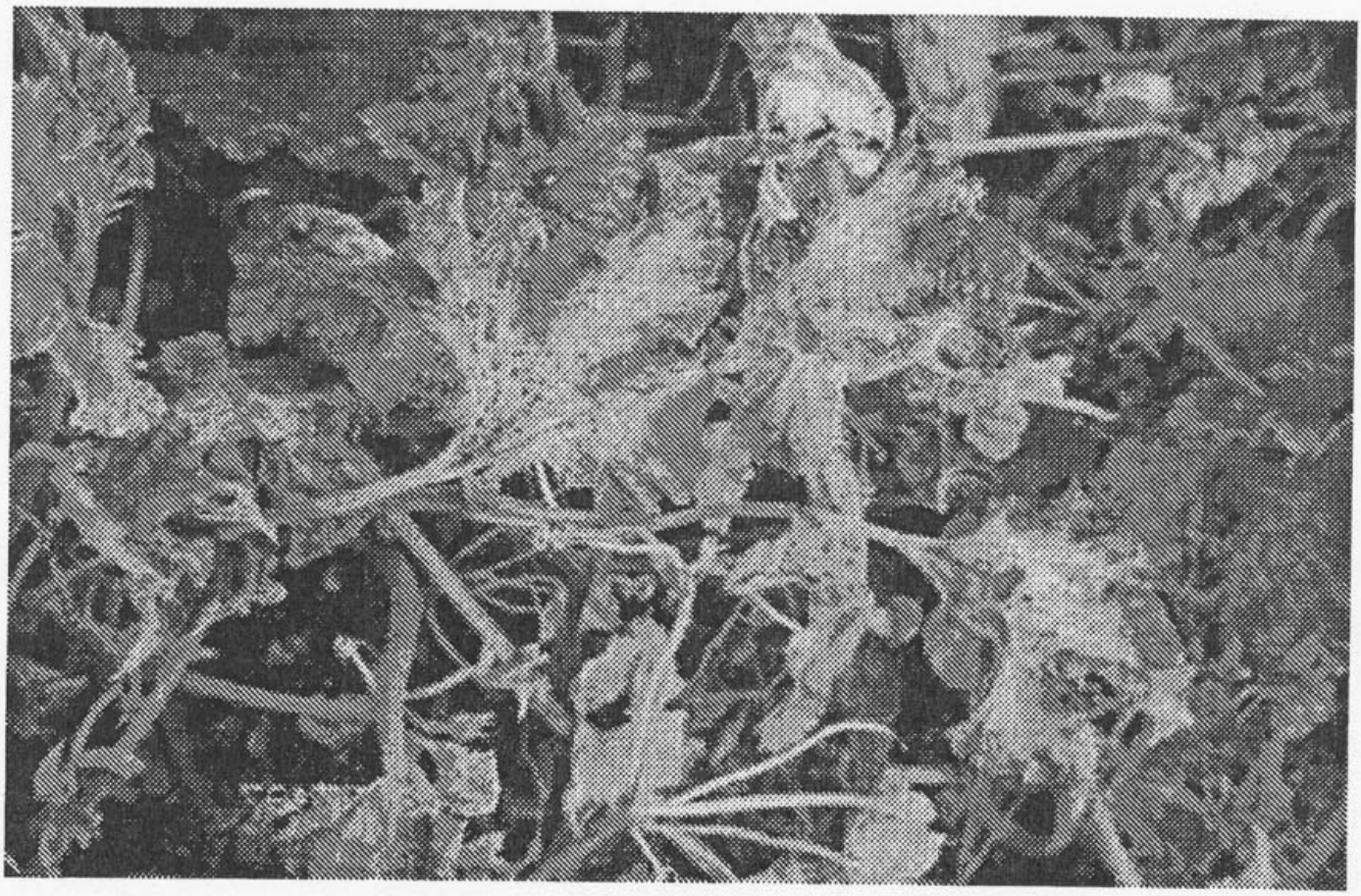
The mentioned measure, especially the improvement of seed quality might reduce the spread of ZYMV. In order to avoid yield losses like in 1997, however, they must be combined with the use of a ZYMV-tolerant oil pumpkin variety. In contrast to other varieties of *cucurbita pepo* , for oil pumpkins only one or two fruits per plant are needed to achieve a satisfactory harvest.Thus even a ZYMV-tolerance allowing the first fruits to develop without major damage should be sufficient to insure acceptable yields.

Fig. 1.: Symptoms of ZYMV on leaves of *Cucurbita pepo* var. *styriaca* approximately 2 months after germination.

Fig. 2.: Malformations of a young oil pumpkin fruit caused by ZYMV.

Literature Cited

1. Collar, J,L., Avilla, C., Duque, M. and A. Ferres, 1997. Behavioral response and virus vector ability of *Myzus persicae* probing on pepper plants treated with aphicides. J. Econ. Entomol. 9-0, 1628-1634.
2. Fletcher, J.D., Nott., H.M., Wallace, A.R., Rogers, B.T. and J.B.Herman, 1998. Potyviruses in New Zealand buttercup squash. 9th Conference of the ISHS vegetal be virus working group, --. 24-25, Turin 2-27, August 1998.
3. Lisa, V. and H. Lecoq, 1984. Zucchini yellow mosaic virus. CMI/AAB Descriptions of Plant Viruses no. 282.



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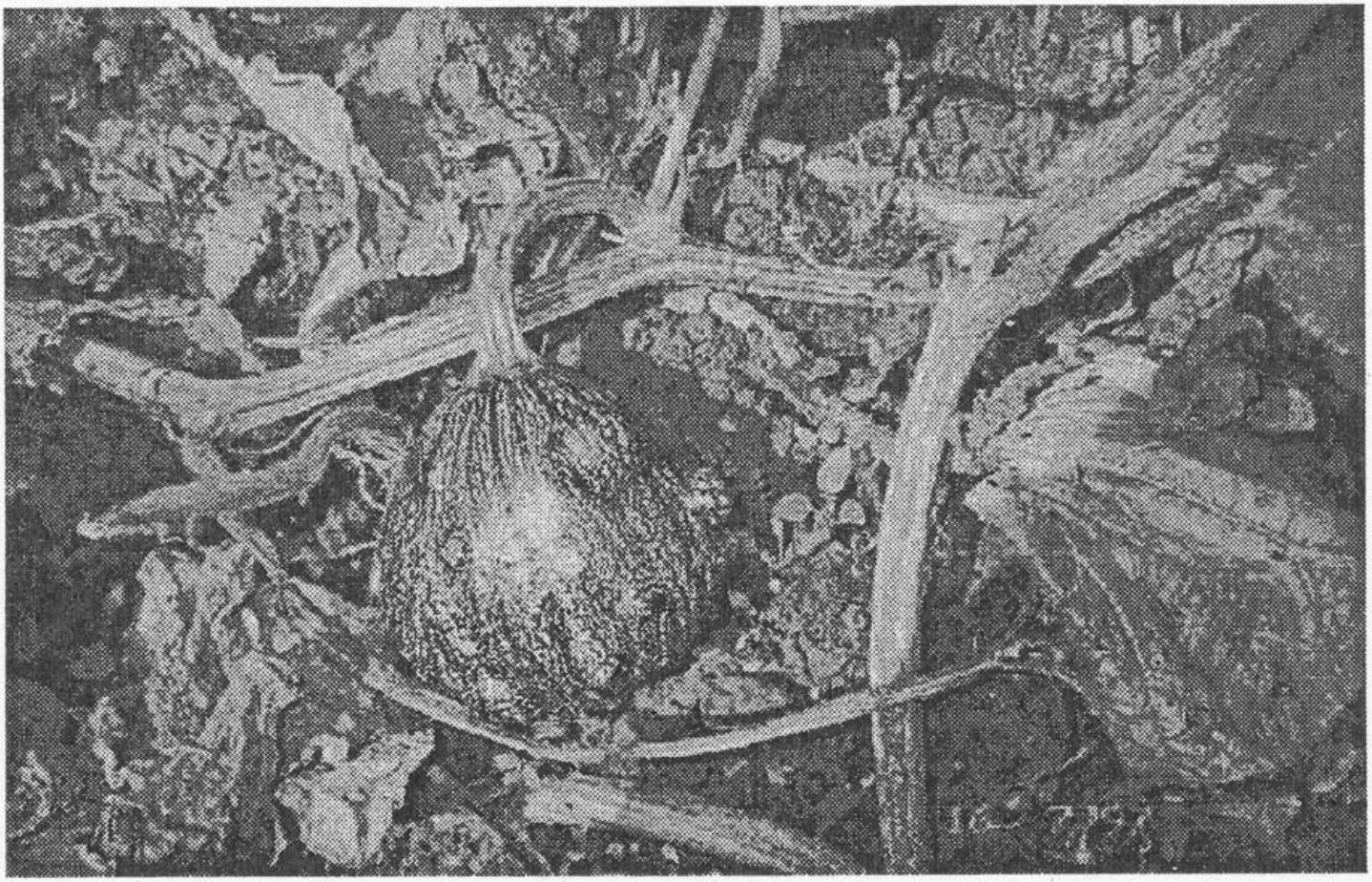

Fig. 2.: Malformations of a young oil pumpkin fruit caused by ZYMV.

Cucurbit Genetics Cooperative Report 23:117-119 (article 39) 2000

Breeding for ZYMV Tolerance of Seed-Oil Pumpkin (*Cucurbita pepo* var. *styriaca*) in Austria using Molecular Markers

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Abstract: For the first time in Austria, the year 1997 brought a severe infection of oil pumpkin by the Zucchini Yellow Mosaic Virus (ZYMV) causing a 50% loss in yield. an initiative to introduce resistance into Austrian germplasm by breeding found immediate acceptance and financial support by the Federal Ministry of Agriculture and Forestry and by the primarily affected states Styria, Burgenland and Lower-Austria.

Since, Hitherto, no resistance or tolerance genes were found in the species *cucurbita pepo* , genes of tolerance originally derived from *Cucurbita moschata* are being introduced in Austrian germplasm via crosses with Zucchini varieties possessing such tolerance. Sincere tolerance in this material appears to be a recessive trait, to aid selection attempts are being made to find a molecular marker linked to the tolerance gene, using bulked segregant analysis (BSA). F_2 derived F_3 plants were tested with the virus isolated in austria to determine the genetic constitution of the F_2 plants for creating the DNA bulks parallel to the BC_2F_1 plants were produced. If BSA will be successful in the winter 99/00, plants having the tolerance gene in heterozygous condition inBC_2F_1 plants can be selected for selfing or further backcrossing.

Keywords: *ZYMV, Zucchini Yellow Mosaik Virus, molecular marker, RAPD, BSA, Bulked Segregant Analysis*

Introduction: Zucchini yellow mosaic virus (ZYMV) is one of the most destructive pathogens infecting cucurbits including oilseed pumpkin. The virus was first described in Italy 1973 (1,2); later several ep9idemics were reported (3,4). In *C. pepo* or its close wild relatives no resistance to this virus has been found so far (5); however, resistance is available in some accessions of *C. moschata* from Nigeria and Portugal. It was described as a single incompletely dominant gene, *Zym* (6,7). The only wild species up to now identified as carrying resistance to ZYMV is *C. ecuadorensis.* This resistance is also conferred by a single, incompletely dominant gene, *Zym* (8). Direct transfer via interspecific crosses can only be expected to be successful in *C. maxima* (9).

Traditionally, a specific "hull-less" or "naked-seeded" pumpkin cultivar *C. pepo* var. *styriaca* has been grown in Austria for seed-oil production for well over 100years.In 1997, for the first time, a severe ZYMV epidemic destroyed about 50% of the pumpkin harvest. This happened at a time when increasing consumer awareness for taste, nutritive value and potential health effects of pumpkin seed oil caused an expansion of the growing area. An initiative to introduce resistance into austrian germplasm by breeding found immediate acceptance and financial support by the Federal Ministry of Agriculture and Forestry and by the most highly affected states Styria,Burgenland, and Lower-Austria.

This presentation describes the progress of this work, starting from its inception in the year 1998.

The breeding strategy: An extensive search for sources of resistance led to the identification of several zucchini varieties in the U.S.A. which possess a gene for tolerance against ZYMV. This gene was first reported in a genotype from Nigeria ("Nigerian Local") and, based on crossing experiments with other *C. moschata* cultivars, has been described as being partially dominant as a result of having some modifier genes for a single resistance gene (6). Transferring this gene to *C. pepo* summer squashes improved their tolerance to ZYMV, however, not to the same extent as in *C. moschata* (6). Nevertheless, a limited number of zucchini cultivars possessing this gene have been released by a few American seed companies.

Seeds of the varieties Jaguar, Tigress, and Puma were obtained from the Harris Moran Seed Company and of the varieties

Dividend and Revenue from Novartis U.S.A. They were described as highly tolerant against isolates of ZYMV from America and China (10). All these genotypes were tested, together with Austrian breeding lines, against the virus isolated in Austria. This experiment has proven the good tolerance of the zucchini varieties from America when compared to the high susceptibility of the Austrian material. An artificial infection of the two cotyledons and the first leaf with a high virus titer of the inoculum, while more or less killing the Austrian genotypes (strong mottling, deep foliar serration of young leaves, stunting or death), caused slight leaf symptoms, no stunting and good fruit formation, although protrusions could be observed on many of the fruits. Crosses of these varieties with Austrian germplasm were successful. The F_1 was again tested with the Austrian isolate of the virus. All F_1 combinations were partly selfed, partly back-crossed with the respective Austrian parents.

Later F_2 progenies of two combinations were selected and tested with the virus. One has shown a 37:11 susceptibility to tolerant segregation, the other combination segregated 41:7. Testing 10 to 15 F_3 progenies of 48 and 47 selfed F_2 plants of the two selected crosses identified highly susceptible and tolerant F_2 progenies. The fact that in the tolerant progenies a few highly susceptible plants regularly occurred, suggests that either tolerance is determined not only by one single gene, or that genetic background plays a role in the expression of the character.

DNA of tolerant plants of each of 12 different tolerant F_2 progenies and from susceptible single plants of each of 12 different highly susceptible F_2 progenies were collected. DNA will be pooled separately for a bulked segregant analysis.

The DNA approach: Bulked segregant analysis (11)m i.e. testing two DNA-pools with isogenic background differing only in one phenotypic character for polymorphism, is a very efficient method for finding a molecular market closely linked to a gene determining this character. Prerequisite is clear distinction of genotype classes, susceptible versus tolerant, and sufficient polymorphism between donor (*C. moschata*) and receptor (*C. pepo*) at the DNA level. The method is especially useful for dominant type markers i.e. RAPD or AFLP. Analyzing genetic relatedness of different pumpkin inbred lines revealed a high level of polymorphism within *C. pepo* using RAPD marker (12). Even more polymorphism can be expected between the two species (Fig. 1). Therefore, it can be expected that DNA polymorphism between these two species, close enoughto the gene for tolerance, will be found when a sufficient number of markers has been tested. At present, F_2 derived F_3 populations have already been tested for tolerance or high susceptibility and were identified. DNA from these plants has been isolated and isogenic pools established. Testing of markers for polymorphism on these two pools is in progress.

6. Munger, H.M., and R. Provvidenti. 1987. Inheritance of resistance to zucchini yellow mosaic virus in *Cucurbita moschata*. Cucurbit Genet. Coop. 10:80-81.

7. Paris, H.S., S. Cohen, Y. Burger, and R. Yoseph. 1988. Single gene resistance to zucchini yellow mosaic virus in *Cucurbita moschata*. Euphytica 37:27-29.

8. Robinson, R.W.,N.F. Weeden, and R. Provvidenti. 1988. Inheritance of resistance to zucchini yellow mosaic virus in the interspecific cross *Cucurbita maxima* x *C. ecuadorensis*. Cucurbit Genet. Coop. 11:74-75.

9. Robinson, R.W., and D.S. Decker-Walters. 1997. Cucurbits. Crop production science in horticulture series, Jeff Atherton (ed.) pp. 51-55.

10. Provvidenti, R. 1997. New American summer squash cultivars possessing a high level of resistance to a strain of Zucchini yellow mosaic virus from China. Cucurbit Genetic Coop. 20:57-58.

11. Michelmore, R.W., I. Paran, and R.V. Kesseli. 1991. Identification of markers linked to disease-resistance genes by bulked segregant analysis: A rapid method to detect markers in specific genomic regions by using segregating populations. Proc. Natl. Acad. Sci. USA 88:9828-9832.

12. Stachel, M., Gy, Csanadi, J. Vollmann and T. Lelley. 1998. Genetic diversity in pumpkin (*Cucurbita pepo* L.) as revealed in inbred lines using RAPD markers. Cucurbit Genet. Coop. 21:48-50.

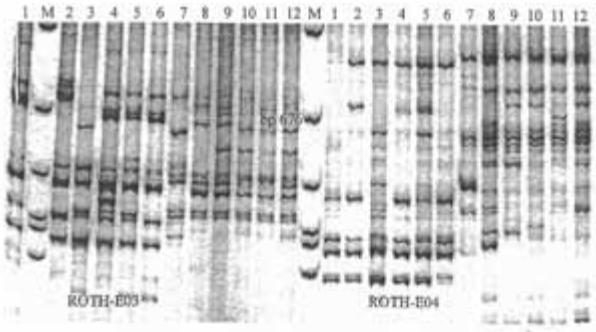

Fig. 1. Polymorphism within and between the two species *C. pepo* and *C. moschata* as found with two RAPD primer, ROTH-E04 and 04). Fragments are separated in 10% polyacrylamid gel, numbers 1 to 6 represent C. *pepo*, 7 to 12 C. *moschata* genotypes. M stands for the size marker: D15 Novex

Acknowledgement. All virus testing is being carried out in the Federal Office and Research Center for Agriculture under the supervision of DIU M. Riedle-Bauer, who also performs the grading of infection. In this connection special thanks are due to Ms. Betty Suarez for her outstanding technical assistance. This project is financially supported by the Federal Ministry of Agriculture and Forestry, by the States Styria, Burgenland and Lower-Austria and by the Breeding Company Gleisdorfer Saatzucht.

Literature Cited

1. Lecoq, H., M. Pitra, and M. Clement. 1981. Identification et caracterisation d'un potyvirus provoquant la maladie due rabougrissement jaune du melon. Agronomie 1:827-834.
2. Lisa, C., G. Boccardo, D'Agostino, G. Dellavalle, andM. D'Aquino. 1981. Characterization of a potyvirus that causes zucchini yellow mosaic. Phytopathology 71:668-672.
3. Provvidenti, R. H.M. Munger, and A.O. Paulus. 1984. Epidemics of zucchini yellow mosaic virus and other cucurbit viruses in Egypt in the spring of 1983. Cucurbit Genetic Coop. 7:78-79.
4. Provvidenti, R. 1986. Occurrence of Zucchini yellow mosaic virus in the United States in 1985. Cucurbit Genetic Coop. 9-96.
5. Provvidenti, R. 1993. Resistance to viral diseases of cucurbits. In: Resistance to viral diseases of vegetables: genetics and Breeding. Molly M. Kyle (ed). Timber Press, Portland Oregon. 8-43.
6. Munger, H.M., and R. Provvidenti. 1987. Inheritance of resistance to zucchini yellow mosaic virus in *Cucurbita moschata.* Cucurbit Genet. Coop. 10:80-81.
7. Paris, H.S., S. Cohen, Y. Burger, and R. Yoseph. 1988. Single gene resistance to zucchini yellow mosaic virus in *Cucurbita moschata.* Euphytica 37:27-29.
8. Robinson, R.W., N.F. Weeden, and R. Provvidenti. 1988. Inheritance of resistance to zucchini yellow mosaic virus in the interspecific cross *Cucurbita maxima* x *C. ecuadorensis.* Cucurbit Genet. Coop. 11:74-75.
9. Robinson, R.W., and D.S. Decker-Walters. 1997. Cucurbita. Crop production science in horticulture series, Jeff Atherton (ed.) pp. 51-55.
10. Provvidenti, R. 1997. New American summer squash cultivars possessing a high level of resistance to a strain of

Zucchini yellow mosaic virus from China. Cucurbit Genetic Coop. 20:57-58.
11. Michelmore, R.W., I. Paran, and R.V. Kesseli. 1991. Identification of markers linked to disease-resistance genes by bulked segregant analysis: A rapid method to detect markers in specific genomic regions by using segregating populations. Proc. Natl. Acad. Sci. USA 88:9828-9832.
12. Stachel, M., Gy, Csanadi, J. Vollman and T. Lelley. 1998. Genetic diversity in pumpkin (*Cucurbita pepo* L.) as revealed in inbred lines using RAPD markers. Cucurbit Genet. Coop. 21:48-50.

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11. Michelmore, R.W., I. Paran, and R.V. Kesseli. 1991. Identification of markers linked to disease-resistance genes by bulked segregant analysis: A rapid method to detect markers in specific genomic regions by using segregating populations. Proc. Natl. Acad. Sci. USA 88:9828-9832.

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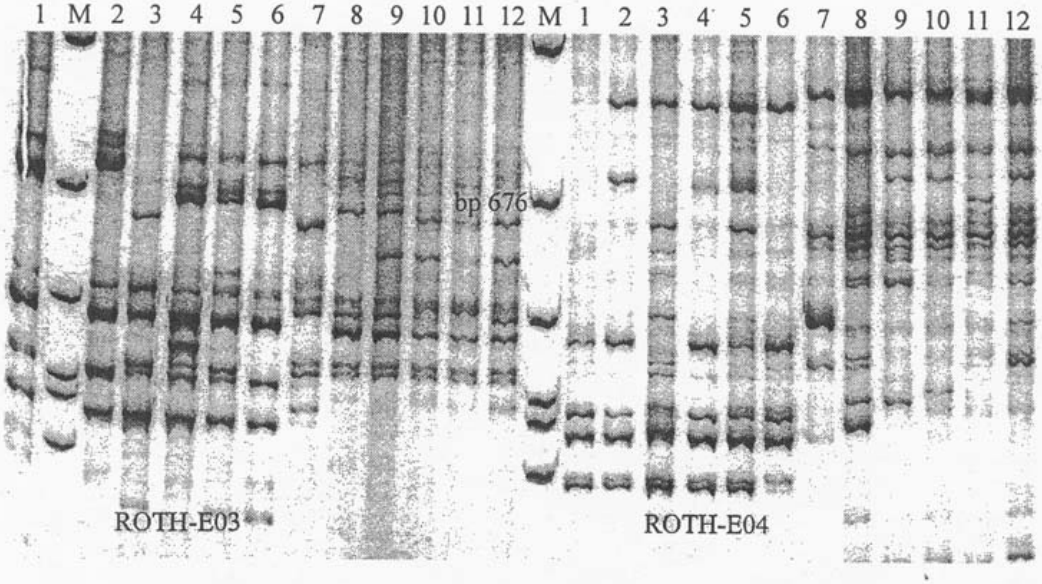

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Cucurbit Genetics Cooperative Report 23:120-121 (article 40) 2000

Production of Cucurbit Seed Oil by Cold Pressing Process in the "Farmaol" Company

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Abstract: The cold (actually low-temperature) process of pressing oil from pumpkin, water- and other melon seed for medicinal purposes (RF Pat. No. 2018514) is described. Construction of the press, as well as process temperature profiles employed, are explained. Analyses of the composition of oil pressed by different methods are given. A number of uses of seed-oil for medical, agricultural, and other purposes are discussed.

Key Words: *pumpkin (watermelon, melon), cold pressing, seed oil*

Introduction: For the production of seed oil of the highest quality for medicinal purposes, compared to oils manufactured for use as foodstuff, special processing technologies are required. These processes must first and foremost, aim at the conservation of all vitamins and biologically active substances or complexes in the oil, even at the expense of taste or the amount of oil yielded by pressing.

For this reason, when oil is to be used for medicinal instead of culinary purposes, consumers prefer oil produced by the so-called cold pressing technology. A special conveying screw press was designed and manufactured by our company, "Farmaol" for this purpose. To begin with, we should like to point out that formerly there were no pumpkins cultivated in Russia that produced :naked" or "hull-less" seeds. Consequently, a very sturdy and powerful oil pressing system had to be constructed since considerably more force is required for the extraction of oils from seeds in their hard shells than from hull-less seeds.

Furthermore, in a layer directly beneath the hull, there are a large number of useful substances, including Santoninum, which has a worm expelling effect or acts as a helminthicide. The initial shelling or dehulling and even the squeezing of the seed both reduce the prophylactic and curative properties of the oil. Thus, while in producing cucurbit oil for medicinal use, a certain loss in quantity is inevitable, yet the higher content of biologically active substances more than compensates for the lower yield.

The technique of "cold pressing": This is the cold-(actually "low-temperature") pressing process for the production of cucurbit-seed oil employed in our plant: selected seed (pumpkin, water- or other melon) previously dried to a moisture content of 7-9%, passes through a special vibrating screen equipped with magnetic "traps". The screen retains extraneous matter above seed size, while the magnetic traps catch small metal objects that inadvertently got into the seed. From the screen, the seed is discharged onto a large-surface tray for final sorting. This step is performed manually, since, in our opinion, this is the only way to assure the premium quality of our oils. Then, the bulk seed, while continuously being agitated and mixed, passes through the first heating chamber within 1 to 3 minutes. In this chamber, temperatures of 130°C to 140°C are maintained. This step kills the microflora on the surface of the seed hull, while the temperature of the seed itself in this short period remains below 50° or 60°C. Then, the seed passes through a second screw-conveyor section up to 80 m in length, where, within 40 to 50 minutes, the final drying phase is completed.

The temperature in this chamber is maintained at 60°C. Then the seed is pre-pressed (the hulls are cracked and crushed), followed by the final pressing in a screw press at low revolutions (n<l/sec), and at a pressure of more than 100 megapascal.

The output ratio varies from 20 up to 30%, depending on the kind of seed and quality of oil required. For culinary purposes the oil can be subjected to higher temperatures, and in this case the yield ratio may be as high as 45%.

Medicinal and biological applications of seed oils derived from the cucurbit family: A comparison of oil produced by the "cold pressing process" described above with oils obtained by the traditional method, which involves crushing and pan-toasting of the seed in order to increase the yield, shows that the cold-pressing method (RF Pat. No. 2018514) produces an oil with higher contents of the desirable components than obtained by the traditional method. The oil contains 1.5 to 2 times more palmitic acid and tocopherols, 3 to 4 times more carotins and 1.2 times more linoleic acid. The content of oleic and stearic acid is lowered 1.2 to 4 times.

Our oil is suitable for the treatment of diseases of the prostate gland, the cholepoietic system, colelithiasis, some renal diseases (there is a relevant Russian patent: "The stimulation of filtrational and excretory functions of the kidney," co-author of this patent is L. Chaban). The main uses of pumpkin-seed oil lie in the treatment of diseases of the liver, in particular toxic liver failure. The essence of the research in which we participated was finding agents constituting an alternative to the famous drug "ESSENTIALE." Experimental and clinical tests confirmed that the specific balance of the basic biological components (tocopherols, sterols, carotinols) as well as linoleic, oleic, palmitic acids, have a characteristic effect on cell structure in the case of toxic hepatitis. It is conceivable that the positive effect is achieved by a high concentration of tocopherols and unsaturated fatty acids canceling their failure to remedy fatty dystrophy. Fatty dystrophy in toxic hepatitis is a rapidly progressing accumulation of fat, caused by fat from storage tissues being transferred to the liver, concurrent with a reduction of triglycerides in this organ. An experiment was performed on white rats infected with toxic hepatitis. Hepatotropic tetrachlorocarbontoxin (CC14) was administered at a ratio of 620 mg/kg. Results of the experiment proved that for rats fed pumpkin seed oil all functions of the liver were restored. It is important to note that all changes occurred without any changes in weight and in the microstructure of the organ. These results led to the recognition of the liver-protecting properties of pumpkin-seed oil and to the granting of a patent (RF Pat. No.2001620) on the discovery of the medication "Hepatoprotector" (co-author L. Chaban), a product opening up new vistas.

Uses of Press Cake: As it is discharged from the press, the dry press cake is broken down by a special shovel attached to the screw. Subsequently, it is bagged and used as an additive to cattle and poultry feed. An increased rate of reproduction has been reported for hogs and poultry fed this kind of press cake. Moreover, cat and dog owners use this waste product to worm their pets. Such bio-additives are, in our opinion, also quite valuable for human consumption.

An unexpected use of this by-product was discovered by fisherman. They have been utilizing the press cake as bait for a long time, but kept this knowledge secret. We only know that they follow some roasting procedures and check the quality by smell. Then it will be pulverized and sieved. The bait is prepared immediately before use by adding water and stirring carefully. The mass should stick together slightly when squeezed. Finally a cereal and clay are added.

Acknowledgements: *We thank all of our workers for their patience and work. We should like to express gratitude to our main designers Gennadij Shipilov and Victor Rechecalov for their dedication to the idea of how to press oil from all kinds of seeds or nuts that can be found on our planet, and for their independent and original way of thinking, which has helped us to turn our ideas into patents. Our special gratitude is expressed to our lovely co-workers Tanya Ponomareva, Gayla and Nastya Artyomenko, Marina Gostuhina. Without them our work would be grey and monotonous.*

We feel special gratitude for Penny Lichtenecker, the American from Vienna, austria, who has encouraged and supported us in making international contacts. She is still continuing to patiently incorporate us into the international community of cucurbit workers.

Cucurbit Genetics Cooperative Report 23:122-123 (article 41) 2000

The Health Value of Styrian Pumpkin Seed Oil - Science and Fiction

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Styrian Pumpkin-seed oil is a natural vegetable oil

Vegetable oils are very important in human nutrition. They provide our system with essential polyunsaturated fatty acids, Vitamin E and phytosterins. Among vegetable oils, the oil obtained traditionally from the hull-less seeds of the specific pumpkin variety *cucurbita pepo* var. *styriaca* occupies a distinct position because, in addition to its high content of valuable nutrients, it possesses a unique taste. Its special dark color has brought it the name of "green gold" of Styria (Fig. 1).

Unlike industrially refined vegetable oils, Styrian pumpkin-seed oil is a pure natural oil. Industrially refined oils go through a series of technical processes designed to produce an oil with little taste or odor. According to the austrian Food Codex, natural oils have to be produced "only by mechanical and physical processes,"and "no additives" are permitted. Pure natural oils, such as Styrian pumpkin-seed oil, retain all their original nutritional substances which in combination give them their distinctive odor and taste and which can contribute significantly to human health.

The healing qualities of Styrian pumpkin-seed oil have been recorded over several centuries. At present numerous medicinal preparations made from Styrian pumpkin-seeds are on the market. Two fields of medical applications are of special interest: *prostate hyperplasia* and *bladder irritation.*

An enlargement of the prostate gland is a typical ailment of men over 50. the actual cause of this condition has not yet been completely explained. One distinct possibility is the ratio change in the production of sexual hormones (androgen/estrogen) that occurs with aging. The phytosterins which are present in Styrian pumpkin-seed oil, because of their structural relationship to some bodily substances involved in androgen metabolism, are capable of reducing hormonal stimulation of the prostate cells, thus effectively protracting the development of adenoma of the prostate. Women in this age group often suffer from the consequences of irritations of the bladder. This typically feminine condition is being successfully treated with medications containing the active ingredients of Styrian oil-pumpkin seeds.

Styrian pumpkin-seed oil and cardio-vascular diseases

Heart and circulatory diseases are Number 1 among civilization-related threats to health. A number of factors, such as stress, smoking, and bad eating habits, are cited as causes for the growing frequency of cardio-vascular complaints. Nutritional institutions react to this phenomenon by recommending the reduction of over-all fat consumption - particularly of animal fats. They further recommend the selective use of vegetable oils. For these purposes, Styrian pumpkin-seed oil is especially suited because of its advantageous combination of fatty acids, over 50% of which is linoleic acid, one of the essential polyunsaturated fatty acids, necessary for maintaining health. furthermore, pumpkin-seed oil is rich in Vitamin E, carotenoids and phytosterines. Absence of cholesterol makes this oil an ideal natural nutrient that can help prevent heart and circulatory diseases. In this respect, Styrian pumpkin-seed oil conforms perfectly to the recommendations of nutritional institutions (Table 1).

Table 1. Constituents of Styrian pumpkin-seed oil per 100 g oil.

Energy	**879 kcal/2682 kJ**
Vitamin E	3500 µg a-tocopherol 4000 µg tocopherol equivalents
Saturated fatty acids	20.5g

Mono-unsaturated fatty acids	23.1g
Poly-unsaturated fatty acids from this linoleic acid from this linoleic acid	51.6g50.9g

In addition to scientifically investigated and proven health benefits, there are of course many traditions found in folk medicine which claim a great variety of curative effects for pumpkin-seed oil, ranging from worming medicine to an aphrodisiac now fondly referred to as "Styrian Viagra". Many of these well-intended uses and recommendations have in common that they belong more to the realm of fantasy and fiction. At present they cannot be considered as a serious basis for scientific research.

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Fig. 1: The „green gold" of Styria

Cucurbit Genetics Cooperative Report 23:1214-125 (article 42) 2000

"Styrian Pumpkin-Seed Oil g.g.A" - Over One Million control Numbers Have Been Assigned

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The Stamp of Origin - "Styrian Pumpkin-Seed Oil g.g.A": The growing of pumpkins for the production of pumpkin-seed oil is an old tradition in Styria and now represents an interesting and important source of income for 10,000 farmers in Styria, Burgenland, and Lower Austria as well as for 60 commercial oil mills.

The european Union has introduced the Stamp of Origin as a means of protecting special regional products of high quality and of guarding them and the regions that produce them against imitations and unfair competition. In order to secure pumpkin-seed oil as a source of potential income for the future and to ensure an increase in volume, a request was filed in 1995 for its registration as a Protected Stamp of Origin according to Article 5 VO (EEC) Nr. 2081/92 as "Styrian Pumpkin-Seed Oil g.g.A" (Fig. 1).

The letters"g.g.A" mean "geschutze geographische Angabe" i.e., "protected designation of origin." About 2000 farmers joined together to produce quality pumpkin-seed oil according to the regulations for g.g.A products. This represents approximately 70% of the domestic land under cultivation for the production of marketable oil-pumpkin crops.

Complete record of production: After three years of basic preparations to meet the requirements of the European Economic community regulations, "Styrian Pumpkin-Seed Oil g.g.A" was officially recognized as a Stamp of Origin on November 12, 1998, ensuring protection to this product throughout Europe. The producers' Society, "Styrian Vegetables," has built up a system of continuous control for the production of pumpkin-seed oil bearing this stamp. Thus the origin of each bottle of Styrian pumpkin-seed oil can be traced completely from the field, to the harvested crop, to the oil mill where it is pressed, and finally to the point of sale. Just like Champagne, Prosciutto di Parma, Prosciutto di San Daniele or Greek Feta cheese, Styrian Pumpkin-Seed Oil g.g.A. is one of the best-controlled specialties of Europe.

The control system for Styrian Pumpkin-Seed Oil from specific protected geographic areas ensures:

- Oil-seed grown in designated areas of eastern Austria
- Oil pressed in mills located in the production region
- 100% pure pumpkin-seed oil from the first pressing

The consumer will recognize this special pumpkin-seed oil by the stamp "Styrian Pumpkin-Seed Oil protected geographic specification." Each stamp bears an individual control number which makes it possible to follow the pumpkin seed from the farmer's field to the display shelves in the store. The Styrian State Food Authority is responsible for these controls.

One million control numbers: From December 1998 to December 1, 1999, one million control numbers have been issued. Until mid-February 2000, 1.200.000 control numbers have been issued. This means that now much more than one million bottles of Styrian pumpkin-seed oil bear the protected-designation-of-origin stamp, indicating the great interest of the farmer in offering an exclusive product of high quality.

Styrian pumpkin-seed oil, with its unique taste, beneficial nutritional effects and now, with its official recognition by the European Union as a product with protected designation of origin, reflects in a certain manner the Styrian way of life and culture. In this sense "Styrian Pumpkin-Seed Oil" a visiting card for Styria which bears the message of sustainability an intelligent agriculture.

Fig. 1: The Stamp of Origin for the Styrian pumpkin seed oil issued by the EU.

Fig. 1: The Stamp of Origin for the Styrian pumpkin seed oil issued by the EU.

Cucurbit Genetics Cooperative Report 23:126-127 (article43) 2000

Cucurbita pepo - History and Thin Coated Seeds

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From the herbals of the 16th century it can be shown, that horticultural groups of *Cucurbita pepo* L. subsp. *pepo* (pumpkin, vegetable marrow) and of *C. pepo* L. subsp. *ovifera* (L.) Decker (scallop, acron, ornamental gourds) were already present at this time in europe, to allow rapid evolution of new cultivars (Fig. 1). Because of the scarcity of vegetable oils and fats in parts of Central Europe during these ages, the appearance of a pumpkin with its large, oil-rich seeds, was a welcome relief. So after this discovery of the New World, pumpkin spread quickly over the Old World as a vegetable and in some regions also as an oil plant. The first step in traditional processing of pumpkin seeds for oil production in Styria (Austria, Central europe) was peeling the seeds from the thick seed coat. The first reliable record for such peeled seeds dates back from the year 1735 (5).

The appearance of the thin coated mutant simplified the processing greatly (Fig. 2). The exact time of appearance is not known. In the penible report of the agricultural situation in Styria of Hlubek 1860 (3) only the peeling of seeds but no thin coated seeds, which can be processed without peeling, are mentioned. Even in the famous agricultural flora of Alefeld 1866 (1) from the 66 cultivars of *C. pepo* no thin coated type is mentioned. On the other hand, the search of the pumpkin breeders Tschermak-Seysenegg and Buchinger in the 30's and 40's lead them to the opinion that it was present at ca. 1880. thus we can estimate, that the Vinous Styrian Oil Pumpkin (*C. pepo* L. subsp. *pepo* var. *styriaca* Greb.) segregated between c. 1870 and 1880 in the region of Styria from the normal field pumpkin of that time (8, 2, 6, 7).

The most important characteristic of var. *styriaca* is the lack of any lignification in the seed coat, whereas in the normal thick coated *C. pepo* the four outer ones of the five seed coat layers are strongly lignified. In the modern literature the prevailing opinion is that, that only one gene (*N/n*) is responsible for this character, see e.g. the gene list of Hutton & al. (4). Under this circumstance it would be difficult to understand, why such a mutation occurred only once. The discovery of a mutant with a so-called semi-thin seed coat and the segregation in a thin x semi-thin cross lead the author to estimate that at least six genes much be responsible for the seed coat characteristics. In this case, an allele combination which gives rise to the very special thin seed coat phenotype of var. *styriaca* must be very understandable.

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Fig. 1. Fuchs (1543) "Tüerckisch Cucumer"

Fig. 2. "Auspatzel" - Styrian dialect meaning "removing the seeds."

Literature Cited

1. Alefeld, F. 1866. Landwirthschaftliche Flora oder die nutzbaren kultivirten Garten- und Feldgewachse Mitteleuropa's. Berlin.
2. Buchinger, A. 1944. Kurbiszuchtung. Zuchter. 16 (4-6): 75-85.
3. Hlubek, F.X. 1860. ein treues Bild des Herzogthumes Steiermark Graz.
4. Hutton, M.G. and Robinson, R.W. 1992. Gene List for *cucurbita* spp. Cucurbit Genetics Coop. Rpt. 15:102-109.
5. Kundegraber,M. 1988. Die volkstumliche Ernahrung im Lichte der Untertaneninventare. Am Beispiel der Herrschaft Stainz. Z. Histor. Verein. Steierm. 79:187-193.
6. Teppner, H. 1982. Der Steirische Olkurbis und cinge fruhe Quellen uber Kurbisanbau. In: Teppner, H. (ed.) Die Koralpe. Beitrage zur Botanik, Geologie, Klimatologie und Volkskunde. Graz. 57-63.
7. Teppner, H. 1999. Noitzen zur Geschichte des Kurbisses. Obst, Wein, Garten (Graz). 68(10):36.
8. Tschermak-Seysenegg, E. 1934. Der Kurbis mit schalenlosen Samen, eine beachtenswerte Oelfrucht. Wiener Landwirtschaftl. Zeitung. 1934 (7): 41-42, (8): 48-49.

Fig. 1. Fuchs (1543) "Tüerckisch Cucumer"

Fig. 2. "Auspatzel" - Styrian dialect meaning "removing the seeds."

Cucurbit Genetics Cooperative Report 23:128-128 (article 44) 2000

Styrian Pumpkin Oil: The Marketing Perspective

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For nearly 300 years, Styria in Austria has been the home of the well-kept secret of pumpkin seed oil. Not only did this oil evolve as the cornerstone of Styria's diet it was also recognized for its health benefits as early as 1773. Since its introduction to the North American market Styria's superior quality pumpkin seed oil has "stopped buyers and chefs in their tracks." The excitement has not been limited to only Epicurians and natural food enthusiasts. Researchers have also begun to take an interest in discovering the nearly limitless possibilities of Austria's pumpkin seed oil. This interest was underscored in August 1999 during the first Oil Pumpkin Conference appropriately held in the home of these unique pumpkins, Austria. Aside from increasing general knowledge of this special pumpkin variety, the researchers established a network to exchange information to help overcome the Zucchini Yellow Mosaic Virus (ZYMV) that is threatening pumpkins around the world. It is hoped that by their next meeting, a natural solution will have been found to eliminate this most serious threat to pumpkin crops.

A natural solution to this problem is critical to the continuation of successful marketing of the distinctive products made from the Styrian pumpkin. Not only natural-food enthusiasts, but also gourmet clients are skeptical of the long-term effects modern farming techniques (especially chemicals and genetic engineering) have on humans and expect that the pumpkin products from Austria's Styria are grown as 'naturally" as possible making them free of chemical contamination and artificial manipulation.

The increased cooperation between international researchers will also have additional benefits to market efforts of the seedless pumpkin products. The first will be the substantiation of long-accepted "farmer's remedies."

For generations, these remedies have been accepted without scientific documentation. Unfortunately, government scrutiny and other skeptics will not accept untested claims. Another benefit will be the discovery of new uses of these products to alleviate symptoms of some of the ailments plaguing modern man. Areas of special interest are the benefits to: men with enlarged prostate, anemic women, cardio-vascular health, and as a rich source of antioxidants.

The data documenting the findings of scientists must be the foundations for the articles written to enlighten medical experts and lay readers alike. from these "neutral" publications,marketers can design promotional materials targeted at trade buyers and end use customers regarding to the beneficial qualities of these fine products.

In 1998 the European Union awarded Styria "geographical protection" (similar to the *Vidalia Onion* of Georgia or french *Champaign*) for the pumpkin and its products. The corresponding seal is the consumers' assurance that the seeds and oil bearing it are pure and have their origin in Styria. The efforts of the Austrian agricultural community (farmers, biologists, botanists and nutritionists) will "raise the stakes" ensuring that the seal also identifies the quality level against which all other pumpkin products will be measured.

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A Bibliography of the Oil Pumpkin (*Cucurbita pepo*)

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This bibliographic survey covers the diverse publications in the breeding, biology, and usage of the oil pumpkin (*Cucurbita pepo*). It includes the uses of the oil pumpkin for its edible and medicinal oil and as a snackseed. Related species with similar uses (particularly the numerous reports of attempts at cultivating *C. foetidissima* as a new oilseed in arid regions) are not included in this bibliography. This list is intended to help research workers, growers, and the food industry keep up-to-date in their literature of this increasingly important world crop plant.

Bibliography

1. Abak, K., M. Sakin and S. Sakin-Kara Kulluku. 1990. Improvement of pumpkin for naked seeds. Int. Hort. Congr. 23:Abstract 3074.
2. Abak, K., N. Sari, B. Cetiner and S. Buyukalaca. 1999. Changes of protein, fat content and fatty acid composition in naked pumpkin seeds influenced by sowing time. proc,. First. Int. Symp. Cucurbits, Adana, Turkey, 20-23 May 1997. 492:187-192.
3. Abbott, P. 1999. Specialist Oil Seed Production and the Growing Potential for the United Kingdom. Nuffield Farming Scholarships Trust,. Uckfield. East Sussex. UK.
4. Akhtar, M.W., M.Z. Iqbal and M.N. Nawazish. 1980. Lipid class and fatty acid composition of pumpkin seed oil. Pakistan J. Sci. Res. 32(304):295-300.
5. Akritidis, C.B. and A.J. Siatras. 1979. Resistance of pumpkin seeds to air glow [during drying process]. Trans. Amer. Soc. Agric. Engin. 22(6):1414-1416.
6. Akritidis, C.B., C.A. Tsatsarelis and C.B. Bagiatis. 1998. Equilibrium moisture content of pumpkin seed. Trans. Amer. Soc. Agric. Engin. 31(6):1824-1827.
7. Al Khalifa, A.S. 1996. Physicochemical characteristics, fatty acid composition, and lipoxygenase activity of crude pumpkin and melon seed oils. J. Agric. Food Chem. 44(4):964-966.
8. Al-Zuhair, H., A.A. Abd-el-Fattah and H.A. Abdel-el-Latif. 1997. Efficacy of simvastastatin and pumpkin-seed oil in the management of dietary-induced hypercholesterolemia. Pharmacol. Res. 35(5):403-408.
9. Alekseeva, M.V. 1958. The seed proteins of *Cucurbita.* Trudy po Khim. Prirod. Soedin., Kishunev. Gos. Univ. 1958:103-108.
10. Alekseeva, M.V. 1965. Investigation of salt-soluble proteins of pumpkin seeds (*Cucurbita pepo* L.) by a column gradient extraction method. Biokhimya 30(1-:60-66.
11. Ali, S.M., H.H. Moghademn, D. Yazdani and P.A. Avval. 1999. Effect of plastic mulches, spacing and phosphorous and potassic fertilizer levels on the growth and yields of common pumpkin. *Cucurbita pepo* convar. *pepo* var. *styriaca.* J. Med. Aromat. Pl. Sci. 21(3):650-653.
12. Appendino, G., J. Jakupovic, E. Bellforo and A, Marchesini. 1999. Multiflorane triterpenoid esters from pumpkin. An unexpected extrafolic source of PABA. Phytochemistry 51(8):1021-1026.
13. Arevalo, J.J. 1957. Pumpkin seeds. Studia Rev. Univ. Atlantico, Barranquilla 2:138-209.
14. Asenjo, C.F. and J.A. Goyco. 1951. Preliminary note on the nutritional value of pumpkin seeds (from Puerto Rico). Bol. Colegio quim. Puerto Rico 8:14-16.
15. Axtell, B.L. and R.M. Fairman. 1992. Minor Oil Crops. FO Agricultural Services Bulletin 94. Food and Agriculture Organization of the United Nations. Rome.
16. Baratova, L.A., N.A. Bokova, A.K. Yus'Kovich and I.A. Samylina. 1983. Fatty-acid composting of pumpkinseed oil. Chem. Nat. Compounds 18(2):228-229.
17. Bastic, M., L. Bastic and J.A. Jovanovic. 1979. Triterpene alcohols in some vegetable oils. Glasnik Khemijskog Drushtva Beograd 44(9-10):619-626.
18. Bastic, M., L. Bastic and J.A. Jovanovic. 1980. 4-methylsterols and aliphatic alcohols in some vegetable oils. Glasnik Khemijskog Drushtva Beograd 45(6):251-260.
19. Basic, M., L. Bastic, J.A. Jovanovic and G. Spiteller. 1977. Sterols in pumpkin seed oil. J. Amer. Oil Chem. Soc.

54(11):525-527.
20. Bemis, W.P., J.W. Berry, M.J. Kennedy, D. Woods, M. Moran and A.J. Deutchman. 1967. Oil composition of *Cucurbita*. J. Amer. Oil Chem. Soc. 44:429-430.
21. Berenji, J. 1986. Hibridna snaga kod uljane tikve-golice *Cucurbita pepo* L. [Hybrid vigor of naked seeded oil pumpkin, *Cucurbita pepo* L.]. Uljarstvo23(3-4):79-85.
22. Bhatia, I.S., B/K. Gupta, Y. Paul, P.S. Sukjhija and P. Yash. 1977. Lipid composition of some cucurbit seeds Pl. Biochem J. 4(2):47-53.
23. Bodkowski, R., B. Patkowska Sokola, T. Szmanko and L. Jarosz. 1995. Lammfleischqualiat. Verbessrung durch Verfutterung praparierter Kurbissamen. [Lamb meat quality. Improvement by feeding processed pumpkin seed]. Fleischwirtschaft 75(5):722, 727-728.
24. von Boguslaawski, E. 1953. Olkurbis (*Cucurbita pepo* L.). In: Roemer, T., A. Scheibe, J. Schmidt and E. woermann (eds.). Handbuch der Landwirtschaft. 2:370-374. Parey, Berlin.
25. von Boguslawski E. and A. Taghizadeh. 1969. Uber die Dungung des Olkurbisses (*Cucurbita pepo* L.). [Fertilizing oil pumpkins (*cucurbita pepo* L.)]. Bodenkultur 20(4):381-394.
26. Bombardelli, E. and P. Morazzoni. 1997. *Cucurbita pepo* L. Fitoterapia 68(4):291-302.
27. Bracher, F. 1997. Phytotherapy in the treatment of benign prostatic hyperplasia. Urologe Ausgabe A. 36(1):10-17.Five garden plants for home-grown protein [Soybeans, sunflower and pumpkin seeds, comfrey and climbing beans]. Org. Gard. Farming 22(4):74-77.
28. Carbin, B.E., B. Larsson and O. Lindahl. 1990. Treatment of benign prostatic hyperplasia with phytosterols. Brit. J. Urol. 66(6):639-641.
29. Carle, R.B. and J.B. Loy. 1994. Heritability of seed size in hull-less seeded strains of *Cucurbita pepo* L. Cucurbit Genet. Coop. Rep. 17:125-127.
30. Carle, R.B. and J.B. Loy. 1995. Heritability of seed size and its association with fruit size in a hull-less seeded population of *Cucurbita pepo* L. In: Lester, G.E. and J.R. Dunlap (eds.). Cucurbitaceae '94: Evaluation and Enhancement of Cucurbit Germplasm. 221-223. Gateway, Edinburg, Texas.
31. Carreras, M.E., E. Fuentes and C.A. Guzman. 1989. chemotaxonomy of seed lipids of Cucurbitaceae grown in Argentina. Biochem. Syst, & Ecol. 17(4):287-291.
32. Catalano, M. Fatty acid distribution in glycerides of vegetable oils rich in linoleic acid. rivista Ital. Sotanze Grasse 48(8):398-403.
33. Chen, Y.C. 1988. Naked kernel pumpkin 65-1-8. Zuowu Pinzhong Ziyuan 3:45.
34. Craviota B. R.O. and M. Cervantes. 1965. Estudio sobre proteinas y aminoacidios de alimentos mexicanos. 1. Efficencia proteica de la semilla de calabaza y la mazcla de esta y harina de soja. Ciencia 24(1-2):83-88.
35. Curtis, L.C. 1948. the use of naked seed in *Cucurbita pepo* as a source of high quality liquid vegetable fat, as a high analysis protein, as a new confection, and as a sandwich spread. Proc. amer. Soc. Hort Sci. 52:403-406.
36. Datta, N. 1976. Vegetable oils from some nonconventional sources. Econ. Bot. 30(3):298.
37. Dreikorn, K. and P.S. Schoenhoefer. 1995. The place of phytotherapy in the treatment of benign prostatic hyperplasia. Urologe Ausgabe A 34(2):119-129.
38. Eagles, J.M. 1990. Treatment of depression with pumpkin seeds. Brit. J. Psychiatry 157:937-938.
39. Ecker, S. and O. Horak. 1994. Pathways of HCB-contamination to oil pumpkin seeds. Chemosphere 29(9-11):2135-2145.
40. Eckey, E.W. 1954. Vegetable fats and Oils. Reinhold, New York.
41. El Syiad, S.I., A.S. Abdel Gawad and M.A.H. El Geddawy. 1991. Lipid and phospholipid fractions of some gourd seeds. Assiut J. Agric. Sci. 22(2):297-306.
42. El- Gharbawi, M.I. 1977. Some chemical and physical characteristics of named pumpkin seed oil (*Cucurbita pepo*). Libian J. Agric. 6:199-203.
43. Ellsworth, R.K. 1971. Simple method for separation of milligram quantities of protochlorophyll a from seed oils in extracts of whole pumpkin seeds. Anal. Biochem. 39(2):540-542.
44. Ellsworth, R.K. and C.A. Nowak. 1974. A gas chromatographic-mass spectrometric analysis of the esterifying alcohols of pumpkin seed protochlorophylls. Anal. Biochem. 57(2):534-546.
45. English, J. 1985. Naked pumpkin seeds. Natl. Gardening Nov.:6-8.
46. Ermakov, A.I. and Z.D. Artyugina. 1982. Content and composition of seed oil in different pumpkin varieties. Fiziol. Biokhim. Kul't. Rast. 14(4):332-336.
47. Fahim, A.T., A.A. And-el-Fattah, A.M. Agha and M.Z. Gad. 1995. Effect of pumpkin-seed oil on the level of free radical scavengers induced during adjuvant-arthritis in rats, Pharmacol. Res. 31(1):73-79.
48. Fida, P. 1982. Rueckstandssituation in steirischen Kuerbiskernoelen [Pestzid Rueckstsaendem Chlorkohlenwasserstoffe]. [Situation of pesticide residues in Styrian Gourdkernal salad oil [chlorinated hydrocarbons; Austria]]. Ernachrung 6(2):56.

49. Franz, C. , S. Jager and B. Michel. 1997. Heil und Gewurzpflanzenanbau in Osterreich. [Medicinal and condiment plant cultivation in Austria]. Gemuse Munchen 33(1):12-14.
50. Fritsch, R., H. Ebner, D. Kraft and C. Ebner. 1997. Food allergy to pumpkinseed: characterization of allergens. Allergy Copenhagen 52:3):335-337.
51. Fulara, A. 1971. Effect of planting rates on the yield of fruits, seeds, and oil of 'Pulawska' oil pumpkin, [Wplyw rozstawy roslin na plon owocow nasion i tluszczu dyni oleistej Pulawskiej]. Biul. Inst. Hodowli Rosl 5(104):83-88.
52. Ganzera, M. E.M. Croom, Jr. and I.A. Khan. 1999. Determination of the fatty acid content of pumpkin seed, pygeum, and saw palmetto. J. Med. Food 2(1):21-27.
53. Garg, V.K. and W.R. Nes. 1986. Occurrence of DELTA-5-sterols in plants producing predominantly DELTA-7-sterols: Studies on the sterol compositions of six Cucurbitaceae seeds. Phytochemistry 25(11):2591-2598.
54. Ghaleb, M.L., M. Farines and J. Soulier. 1991. Composition chimique des huiles de graines de citrouille, courge, melon. [Chemical composition of oils of pumpkin, gourd, melon]. Rev. Franc. Corps Gras 38(1-2):17-22.
55. Grebenscikov, I. 1950. Zur Kenntnis der Kurbisart *Cucurbita pepo* L. nebst einigen Angaben uber Olkurbis. Zuchter 20:194-207.
56. Grebenscikov, I. 1954. Zur Vererbung der Dunnschaligkeit bei *Cucurbita pepo* L. Zuchter 24:162-166.
57. Haghdadi, M.R. 1973. Omzichwirkungen und Heterosiseffekte bei Olkurbis "*Cucurbita pepo* L." [Inbreeding effects and heterosis effects of the pumpkin "*Cucurbita pepo* L."] Inaugural Dissertation, no. 26, justus Liebig Universitat. Landwirtschartliche Fakulat.
58. Hamid, S., A.W. Sabhir, Salma and S.A. Khan. 1988. Agrochemical data on *Cucurbita pepo.* Pakistan J. Sci. Industr. Res. 31(7):516-517.
59. Javel, J. 1995. Alternativni olejniny. [Alternative oil crops]. Rosliny oleiste. XVII Ogolnopolska konferencja naukowa 16(1):83-90.
60. Havel, J. 1996. Hodowla roslin oleistych w Republice Czeskiej. [Oilseed breeding in the Czech Republic]. Rosliny Oleiste 17(1):107-111.
61. Heinisch, O. and M. Ruthenberg. 1950. Die Bedeutung der Samensdchale fur die Zuchtung des Olkurbis. Z. Pflanzen. 29:159-174.
62. Henderson, C.W., J.C. Scheerens and J.W. Berry. 1986. Antinutritional factors in *Cucurbita* seed meals. J. Agric. Food Chem. 34(3):434-436.
63. Hethelyi, E., P. Tetenyi, I. Zambo and P. Kaposi. 1989. GC/MS Investigation of different fatty acids from seeds of some medicinal plants. Herba Hungarica 28(3):69-78.
64. Hillebrand, A., M. Murkovic, J. Winkler and W. Pfannhauser. 1996. Ein hoher Gehalt an Vitamin E und ungesattigten Fettsauren als neues Zuchtziel des Kurhbiszuchters. [A high content of vitamin E and unsaturated fatty acids as a new aim of the pumpkin breeder]. Ernahrung 20(10):525-527.
65. Holod, M.H. and V.D. Semychaievs'kyi. 1971. State of protochlorophyll of the inner envelopes of pumpkin seeds in vivo. Ukrayins'k. Bot. Zhurn. 28(1):12-17.
66. Hopkins, C.Y. 1990. Fatty acids of Cucurbitaceae: seed oils in relation to taxonomy. In: Bates, D.M., R.W. Robinson and C. Jeffrey (eds.) Biology and Utilization of the cucurbitaceae. 38-50. Cornell Univ., Ithica, New York.
67. Idouraine, A., E.A. Kohlhepp, C.W. Weber, W.A. Warid and J.J. Martinez-Tellez. 1996. Nutrient constituents from eight lines of naked seed squash (*Cucurbita pepo* L.) J. Agric. Food Chem. 44(3):721-724.
68. Ignatov, N.V., L.B. Belyaeva, K.N. Tomofeev and F.F. Litvin. 1988. Photochemical reactions of protochlorophyll in the inner pumpkin seed coats. Biofixika 33(3):500-504.
69. Imbs, A.B. and L.Q. Pham. 1995. Lipid composition of ten edible seed species from North Vietnam. J. Amer. Oil Chem. Soc. 72(8):957-961.
70. Ito, S., S. Okada and Y. Fujino. 1974. Glyceroglycolipids in pumpkin. J. Agric. Chem. Soc. Japan 48(8):431-436.
71. Jacks, T.J. 1990. Cucurbit seeds: cytological, physiochemical, and nutritional characteristiizations. In: Bates, D.M., R.W. Robinson and C. Jeffrey (eds.). Biology and Utilization of the cucurbitaceae. 356-363. Cornell Univ., Ithica, New York.
72. Jacks, T.J., T.P. Hensarling and L.Y. Yatsu. 1972. Cucurbit seeds: 1. characterizations and uses of oil and proteins. A review. Econ. Bot. 26(2):135-141.
73. Joshi, d.C., S.K. Das and R.K. Mukherjee. 1993. Processing of pumpkin seed for oil. Agric. Engin. Today 17(5-6):83-89.
74. Joshi, S.S,., R.K. Shrivastava and S.S. Nigam. 1977. Calorie and amino acid composition of *cucurbita pepo* and *Cucumis melo* seeds. J. Indian Chem. Soc. 54(7):747-748.
75. Juillet, A., J. Susplugas and J. Courp. 1955. Les Oleagomeux et leurs Tourteaux: Botanique, Caracteres, Preparation, Emploid, Paul Lechevalier, Paris.
76. Kamel, B.S., J.M. De Man and B. Blackman. 1982. Nutritional, fatty acids and oil characteristics of different agricultural seeds Maple, pumpkin, citruses, apples. J. Food Technol. 17(2):263-269.

77. Kester, E.B. 1951. Minor oil-producing crops of United States. Econ. Bot. 5:38-59.
78. Kiendler, A. 1982. Der (Steirische) Olkurbis. [The Styrian oil pumpkin]. Fortschrittliche Landwirt 75(8):12-13.
79. Kinkela, T. and J. Bezard. 1993. Fat content and fatty acid composition of some food products from Congo. Sci. Ailments 13(3):567-575.
80. Kizirya, K.P. and G.N. Kaishauri. 1983. Technological characteristics of *Cucurbita* varieties. Kartofel' & Ovoshechi 1:37.
81. Kroll, J. and F.R. Hassanien. 1983. Studien zur Glceridstruktur von Fette. XVII. Zusammensetzung agyptischer Kurbis- und Melonensamenfetts. [Glyceride structure of fats. XVII. Composition of Egyptian pumpkin nd melon seed fats]. Nahrung 27(1):K1-K2.
82. Kuhlmann, H. U. Koetter, C. Theurer, K. Abak and S. Buyukalaca. 1999. Sterol contents in medicinal pumpkin (*Cucurbita pepo* convar. *citrullinina* var. *styriaca*) depending on genotype and location. Proc. First Int. Symp. Cucurbits, Adana, Turkey, 20-23 May 1997, 492:175-178.
83. Kusmenoglu, S. 1996. Fatty acid composition of oils in Cucurbitaceae seeds. J. Fac. Pharm. Gazi Univ. 13(2):167-170.
84. Lazos, E.S. 1986. A research note: Nutritional, fatty acid, and oil characteristics of pumpkin and melon seeds. J. Food Sci. Technol. (Mysore):51(5):1382-1383.
85. Lazos, E.S. 1992. Certain functional properties of defatted pumpkin seed flour. PI. Foods Human Nutr. 42(3):257-273.
86. Lazos, E.S., J. Tsaknis and M. Bente. 1995. Changes in pumpkin seed oil during heating. Grasas & Aceites 46(4-5):233-239.
87. Lee, J.I., S.N. Ryu, B.H. Lee and Y.M. Kim. 1994. Oil content and fatty acid composition of oil resource plants for edible oil products in Korea. RDA J. Agric. Sci. Upland Industr. Crops 36(1):135-143.
88. Liu, E.P. and S.Q. Wei. 1992. Luo Ren Jin Gua, a new pumpkin variety. Crop Genet Resources 4:47.
89. Loy, B. 1985. Improving seed yields in hull-less seeded strains of *Cucurbita pepo* pumpkins. HortScience 20(2):185.
90. Loy, J.B. 1988. Improving seed yield in hull-less seeded strains of *Cucurbita pepo.* cucurbit Genet. Coop. Rep. 11:72-73.
91. Loy, J.B. 1990. Hull-less seeded pumpkins: a new edible snackseed crop. In: Janick, J. and J.E. Simon (eds). advances in New Crops: Proceedings of the First National Symposium New Crops. 403-407. Timber, Portland, Oregon.
92. Mandl, A., G. Reich and W. Lindner. 1999. Detection of adulteration of pumpkin seed oil by analysis of content and composition of specific DELTA7-phytosterols. Eur. Food Res. Technol. 209(6):400-406.
93. Mansour, E.H., E. Dworschak, T. Huszka, J. Hovari and A. Gergely. 1996. Utilization of pumpkin seed and rapeseed proteins in the preparation of Bologna type sausages. Acta Ailmentaria 25(1):25-36.
94. Mansour, E.H., E. Dworschak, J. Peredi and A. Lugasi, 1993. Evaluation of pumpkin seed (*Cucurbita pepo.* Kaki 35) as a new source of protein. Acta Alimentaria 22(1):3-13.
95. Mansour, E.H., E. Dworschak, Z. Pollhamer. A. Gergely and J. Hovari. 1999. Pumpkin and canola seed proteins and bread quality. Acta Ailmentaria 28(1):59-70.
96. Marini, D.F. Balestrieri and A. Flori. 1991. Determination of the chemical composition of some varieties of Cucurbitaceae seeds, potential sources of oil and proteins. Rivista Della Societa Italisna Di Scienza Dell'Alimentazione 20(1-2):35-39.
97. Markovic, V.V. and L.V. Bastic. 1976. characteristics of pumpkin seed oil. Oils & Oilseeds. J. 29(1):39-42.
98. Markovic, V.V. and L.V. Bastic. 1976/ Characteristics of pumpkin seed oil. J. Amer. Oil Chem. Soc. 53(1):42-44.
99. Martinez, J.J., W.A. Warid and J.M. Loazia. 1996. Improvement of naked seed squash, *cucurbita pepo* L. in Sonora, Mexico. In: Gomez-Guilamon, M.L, C. Soria, J. Cuatero, J.A. Tores and R. Fernandez-Munoz (eds.). Cucurbits Towards 2000. Proc. 6th Eucarpia Meeting on Cucurbit Genetics and Breeding. 66-71. Estacion Experimental "La Mayora," C.S.I.C., Malaga, Spain.
100. Martonffy, B., 1986. Minositett fajta: Magtok. [A certified cultivar: seed pumpkin]. Kert. & Szolesz.35(26):5.
101. Martonffy, B., 1986. Minositett fajitak. [Certified cultivars, Oil pumpkin]. Kert & Sozolesz. 35(38):8-9.
102. Marzec, A. and R. Bulinski. 1997. Content of some trace elements in nuts and edible seeds. Bromatologia i chemia Toksykologiczna 30(2):125-128.
103. Matsui, T., H. Guth and W. Grosch. 1998. A comparative study of potent odorants in peanut, hazelnut, and pumpkin seeds oils in the basis of aroma extract dilution analysis (AEDA) and gas chromatography-olfactometry of headspace samples (GCOH). Fett 100(2):51-56.
104. Matus, Z., P. Molnar and L.G. Szabo. 1993. Olajtok (*Cucurbita pepo* convar, *pepo* var. *styriaca*) magjabol nyert presmaradek ossz-karotinoid-tartalmanak es karotinoid-osszetelenek maghatarozasa. [Main carotenoids in pressed seeds (Cucurbitae semen) of oil pumpkin (*Cucurbita pepo* convar. *pepo* var. *styriaca*)]. Acta Pharm Hung. 63(5):247-256/
105. Meeker, J. 1979. The $6 hill of pumpkins [Naked-seeded varieties]. Org. Gard. 26(5):7`-73. Mudra, A. and D.

Neumann. 1952. Promleme und Ergebnisse der Muncheberger Olkurbiszuchtung. Zuchter 22:99-105.
106. Murkovic,M., A. Hillebrand, S. Draxl, W. Pfannhauser, J. Winkler, K. Abak and S. Buyukalaca. 1999. Distribution of fatty acids and vitamin E content in pumpkin seeds (*Cucurbita pepo* L.) in breeding lines. Proc. First Int. Symp. Cucurbits, Adana, Turkey, 20-23 May 1997. 492:47-55.
107. Murkovic, M., A. Hillebrand, J. Winkler, E. Leitner and W. Pfannhauser. 1996. Variability of fatty acid content in pumpkin seeds (*Cucurbita pepo* :.). Z. Lebensm. Unters. Forsch. 203(3):216-219.
108. Murkovic, M., A. Hillebrand, J. Winkler and W. Pfannhauser. 1996. Development of pumpkin seed for production of edible oil: Distribution of tocopherols in breeding lines. (Meeting abstract). (Food Chem. 57(1):58.
109. Murkovic, M., A. Hillebrand, J. Winkler and W. Pfannhauser. 1996. Variability of vitamin E. content in pumpkin seeds (*Cucurbita pepo* L.). Z. Lebensm. Unters. Forsch. 202(4):275-278.
110. Murkovic, M., J. Winkler, W. Pfannhauser, K. Abak and S. Buyukalaca. 1999. Improvement of the quality of pumpkin seed (*Cucurbita pepo* L.) by use of cluster analysis. Proc. First. Int. Symp. Cucurbits, Adana, Turkey, 20-23 May 1997. 492:41-46.
111. Nesterova, O.V., V.Y Reshetnyak and I.A. Samylina. 1990. Improving the technology of drying in the production of cucurbin extract from pumpkin seeds. Farmatsiya 39(5):39-41.
112. Nesterova, O.V., I.A. Samylina and L.A. Bartova. 1989. Developing methods for standardizing cucurbin obtained from pumpkin seeds. Farmatsiya 38(6):36-38.
113. Neumann, D. 1952. Die Bluhverhaltnisse und der Frucht und Samenansatz beim Olkurbis (*Cucurbita pepo* L.) nach naturlicher und kunstlicher Bestaubung. Z. Pflanzenzucht. 31:513-544.
114. Nikiforov, A., M. Knapp, G. Buchbauer and L. Jorivetz. 1996. Zur Bestimmung der dominierenden Geruchskomponenten (character impact compounds) von steirischem Kirhiskernol. [Determination of dominating aroma compounds (character impact compounds) of pumpkin oil from Styria]. Ernahrung 20(12):643-644.
115. Osman, M.A., M.A.M. Kicka and M.B.A. El Samei. 1979. Chemical composition and hypotensive activity of *Cucurbita pepo* seeds. Res. Bull., Ain shams Univ. 1146:14pp.
116. Pangalo, K.I. 1929-1930. An attempt of studying pumpkins as oil plants. trudy Prikl. Bot. 23:267-275.
117. Pietsch, A. 1942. Beitrag zur photographischen darstellung. Farbbestimmung und bedeutung der olhaltigen samen von in Deutchland wachsenden pflanzen. Landw. Jahrb. 91:369-417.
118. Pleh, M. , I. Kolak, K.D. Dubravec and Z. Satovic. 1998. Sjemenarstvo bundeva. [Squash seed production]. Sjemenarstvo 15(1-2):43-75.
119. Reiterer, E. and R. Reiterer. 1994. Kurbis: von den Fruchten, den Kernen und ihrem Ol. [Pumpkins: of the Fruits, their Seeds and their Oil]. Verlag Brandstatter, Vienna.
120. Robinson, R.G. 1975. Amino acid and elemental composition of sunflower and pumpkin seeds. Agron J. 67(4):541-544.
121. Robinson, R.G. 1981.Production of naked-seeded pumpkin: a food crop for the family farm. Univ. Minn. Agric. Exp. Sta. Misc. Rep. 156:1-6.
122. Rossrucker, H. 1992. Doe Trocknung von Olkurbiskernen. [The drying of kernels of oil squash]. Bodenkultur 43(2):169-173.
123. Rybaltovsikii. O.V. 1966. Lotrytiu kukurbitina - amtiogel'mintonogo deistvuiushehego nachala semian tykvy. [On the discovery of cucurbitin - a component of pumpkin seed with anthelmintic action. Med. Parzitol. (Mosk.) 35(4):487-488.
124. Sadaqat, H. , A.W. Sabir, Salma and S.A. Khan. 1988. Agrochemical data on *cucurbita pepo.* Pakistan J. Sci. Industr. 31(7):516-517.
125. Samant, S.K. and D.V. Rege. 1989. Carbohydrate composition of some cucurbit seeds. J. Food Composition analysis 2(2):149-156.
126. Saura Calixto, F., J. Canellas and J. Garcia Raso. 1983. Determination of hemicellulose., cellulose and lignin contents of dietary fibre and crude fibre of several seed hulls. Data comparison. Z. Lebensm. Unters. Forsch. 177(3):200-202.
127. Schleketa, V.P. 1929-1930. The pumpkin in Ukraine. Trudy Prikl, Bot. 23(3):105-156.
128. Schilcher, H. 1979. Pharmacognostic characteristics and quality criterions of semen cucurbitae pumpkin seeds. Herba Hung. 18(3):151-154.
129. Schilcher, H. 1996. Starkung der Blasenfunktion durch Kurbiskerne? [Improving bladder function by pumpkin seeds?] Med. Monatsschr. Pharm. 19(6):178-179.
130. Schoeniger, G. 1950. Genetische Untersuchungen au *Cucurbita pepo* Zuchter 20-321-336.
131. Schoniger, G. 1952. Volaufige Mittelung uber das Verhalten der Testa-und Farbgene bei verschiedenen Kreuzungen innerhalb der Kurbisart *Cucurbita pepo* L. Zuchter 22:316-337.
132. Schoniger, G. 1955. Beobachtungen zur Vererbung gewisser Testaeigenschaften bei *Cucurbita pepo* L. Zuchter 25:86-89.
133. Schuster, W. 1977. Der Olkurbis (*Cucurbita pepo* L.). Vol. 4 Parey, Berlin
134. Schuster, W. 1987. Die entwicklung des Anbaues und der Zuechtung von oelpflanzen in Mitteleuropa. [Development

of cultivation and breeding of oil plants in central Europe]. Fett Wiss. Technol. 89(1):15-26.
135. Schuster, W., M.-R. Haghdadi and J. Michel. 1974. Inzucht und Heterosis bei Olkurbis (*Cucurbita pepo* L.) I. Inzuchtwirkung. [Inbreeding and heterosis in the oil gourd (*Cucurbita pepo* L.) I. The effects of inbreeding]. Z. Pflanzenzucht. 73:112-124.
136. Schuster, W.,M.-R. Haghdadi and J. Michel. 1974. Inzucht und Heterosis bei Olkurbis (*Cucurbita pepo* L.) II. Bastardwuchsigkeit. [Inbreeding and heterosis in the oil gourd (*Cucurbita pepo* L. II. Hybrid vigour]. Z. Pflanzenzucht. 73:233-248.
137. Schuster, W., W. Zipse and R. Marquard. 1983. Der Einfluß von Genotyp und Anbauort auf verschiedene Inhalsstoffe von Samen des Olkurbis (*Cucurbita pepo* L.). [Influence of genotype and cultivation place on various ingredients of oil-gourd seeds (*Cucurbita pepo* L.)] Fette Seifen anstrichmittel 85(2):56-64.
138. Schuster, W.H. 1992. Olkurbis (*Cucurbita pepo L.* var. *oleifera*). In: Schuster, W.H, and R.A. Marquard (eds.). Olpflanzen in Europa. [Oil Plants in Europe]. 133-138. DLG-Verlag, Frankfurt.
139. Seibel, W., J.-M. Brummer, G. Morgenstern and F.Bretschneider. 1983. Verwendung von Gelbleinsamen und Kurbiskernen bei Backwaren, Getreide Mehl & Brot 37(2):47-55.
140. Selmeczi, K.A. 1993. A magyaroszagi Olajnoveny-Kutultra. [The Culture of Oil Crops in Hungary]. 132-149. Akademiai Kiado, Budapest.
141. Shaller, A., H. Zenz, P. Liebhard and R. Jud. 1988. Studie uber den Einfluss von Stickstoffdungung, Standweite und Versuchsjahr auf die Zusammensetzung der Gesamtfettsauren von Kurbissamenol (*Cucurbita pepo* L. cv. Wies 371). [Study of the influence of nitrogen fertilization, plant spacing nd experimental year on the composition of total fatty acids of pumpkin seed oil (*Cucurbita pepo* L. cv. Wies 371)]. Lebensmittel & Biotechnol 5(4):211-212.
142. Sito, S., J. Barcic and S. Ivanvan. 1988. Influence of various air temperature on duration of drying pumpkin seed with higher water content after washing (*Cucurbita pepo* L.). Poljopr Znanst. Smotra 63(4):285-290.
143. Sperber, J., R. Barisch, E. Edinger and W. Weigl. 1988. Ol-und Eiweißpflanzen: Anbau-Kultur-Ernte, Osterreichischer Agrarverlag, Vienna.
144. Stuart, S.G. 1981. Comparative studies of testa development in normal and hull-less seed strains of *Cucurbita pepo* L. M.S. thesis, Univ. New Hampshire, Durham, N.H.
145. Stuart, S.G. 1983. Comparative biochemical and genetic studies of testa development in normal and hull-less phenotypes of pumpkin (*Cucurbita pepo* L.). Ph.D. thesis, Univ. New Hampshire, Durham, N.H.
146. Stuart, S.G. and J.B. Loy. 1982. Comparison of seed coat development and composition in normal and hull-less strains of pumpkin (*Cucurbita pepo* L.). Cucurbit Genet. Coop. Rep. 5:51-52.
147. Stuart, S.G. and J.B. Loy. 1983. Comparison of testa development in normal and hull-less seeded strains of *cucurbita pepo* L. Bot. Gaz. 144(4):491-500.
148. Stuart, S.G. and J.B. Loy. 1988. Changes in testa composition during seed development in *Cucurbita pepo* L. Plant Physiol. (LifeSci. Adv.) 7:191-195.
149. Suphakarn, V.S., C. Yarnon and P. Ngunboonsri. 1987. The effect of pumpkin seeds on oxalcrystalluria and urinary compositions of children in hyperendemiuc area. Amer. J. Clinical. Nutr. 45(1):115-121.
150. Suphiphat, V., N. Morjaroen, I. Pukboonme, P. Nginboonsri, T. Lowhnoo and S. Dhanamitta. 1993. The effect of pumpkin seeds snack on inhibitors and promotors of urolithaisis in Thai adolescents. J. Med. Assoc. Thai. 76(8):487-493.
151. Svirlith, E., Z. Djarmati, A. Dhordjevic, K. Svan. M. Djhordjevic Milic and N. Djordjevic. 1995. The quality of pumpkin seed oil (*Cucurbita pepo*) obtained by pressing, extraction with n-hexan and CO_2 under liquid and supercritical conditions. In:Berec, B. (ed.). 36th Conference about Production and Processing of Oil Crops, Proceedings, 211-218. Budva, Yugoslavia.
152. Szith, R. and H. Furlan. 1979. Neue Erfahrunggen bei der chemischen Behandlung von Unkrautern im steirischen Olkurbisanbau. [New experiences in the chemical treatment of weeds in Styrian oil-pumpkin growing]. Pfklanzenarzi 32(7-8)"79-80.
153. Teppner, H. 1999. Noitzen zur Gescichte des KJurbisses, Obst, Wein. Garten, (Graz) 68(10):36.
154. Tomoskozi, S., R. Lazity, J. Nagy., A. Bacsuri, E. Boros and L. Sarkadi. 1993. Nem konvencionalkis freherjeforrasoik az emberi talplakozasban. 3. Tokmag feherjeinek vizgalata. [Unconventional protein sources for man. 3. Study of pumpkin seed protein]. Elelmezesi Ipar 47(7):200-203.
155. Tsaknis, J., S. Lalas and E.S. Lazos. 1997. Characterization of crude and purified pumpkin seed oil, Grasas & Aceites 48(5):267-272.
156. von Tschermak-Seysnegg, E. 1934. Der Kurbis mit schalenlosen Samen, eine beachtenswerte Olfurcht. Wiener Landw. Zeitung 84:7-15.
157. Tsuyuki, H., s. Itoh and K. Yamagata. 1985. Lipid and triaclglycerol compositions of total lipids in pumpkin seeds. J. Jap. Soc. Food Sci. Tech. 32(1):7-15.
158. Undina, S., G. Neamtu, S. Apahidean and V. Pui. 1991. Continutul in acizi grasi uleiului extrasdin semintele unor

specii de Cucurbitaceae. [The fatty acid content of the oil extracted from the seeds of some species in the Cucurbitaceae]. Bul. Inst. Agron. Cluj-Napoca. Ser. Agric. Hort. 45(1):51-58.
159. Ventura, P., M. Girola, S. Contos, M. De Bernardi Di Valserra and S. Tripodi. 1996. Effects of a new dietetic integrator based on extracts of *Cucurbita pepo, Serenoa repens* and *Prunus africanus* on functional disturbances associated with prostatic hypertrophy. Rivista Patologia Clinica 51(2):53-66.
160. Vinning, K. and B. Loy. 1998. Seed development and seed fill in hull-less seeded cultigens of *Cucurbita pepo* L. Cucurbit Genet. Coop. Rep. 21:59-61.
161. Vining, K.J. and B. Loy. 1998. Seed development and seed fill in hull-less seeded cultigens of pumpkin (*Cucurbita pepo* L.). In: McCreight, J.D. (ed.). Cucurbitaceae '98: Evaluation and Enhancement of Cucurbit Germplasm 64-69. ASHS Press, Alexandria, Virginia.
162. Vining,K.J. and J.B. Loy. 1998. Born naked: The development of hull-less pumpkin seeds. (Meeting abstract). HortScience 33(2):208.
163. Vogel, P. 1978. Untersuchungen uber Kurbiskernol. [Studies on pumpkin seed oil]. Fette Seifen Anstrichmittel 80(8):315-317.
164. Wagner, C. 1997. Steirisches Kurbiskernol. [Styrian Pumpkin Seed Oil]. Pichler Verlag, Vienna.
165. Warid, W.A., J.J. Martinez and J.M. Loaiza. 1993. Productivity of named seed squash,. *Cucurbita pepo* L. Cucurbit Genet. Coop. Rep. 16:58-60.
166. Weiling, F. 1956. Die Ubertragung des Merkmals "Weichschaligkeit" com Olkurbis (*Cucurbita pepo* L.) in fertile Arbastarde aus der Kreuzung *Cucurbita maxima* Duch,. x *Cucurbita pepo* L. Zuchter 26:22-25.
167. Weiling, F. and E.P.V. Becherer. 1950. Zur Factorenanalyse der Testa aus bildung beim Kurbis. Ber. Deutsch. Bot. Ges. 63:147-148.
168. Wetscherek, S.G. W. Wetscherek and W. Zollitsch. 1991. Use of pumpkin seed cake in diets for fattening pigs. Bodenkultur 42(3):277-289.
169. Whitaker, T.W. 1962. Breeding squash and pumpkins. In: Kappert, H. and W. Rudorf (eds.). Breeding of Legumes and Fruits, Viniculture and Silviculture. Manual of Plant Breeding [Handbuch der Pflanzenzuchtung]. (2nd, ed.). 6:331-350. Parey, Berlin.
170. Whitaker, T.W. and G.W. Bohn. 1950. The taxonomy, genetics, production and uses of the cultivated species of *Cucurbita.* Econ. Bot. 4(1):52-81.
171. Wichtl, M. 1997. Cucurbitae semen,. In: Wichtl, V.M. (ed.). Teedrogen und Phytopharmaka: Ein Handbuch fur die Praxis auf wissenschaftlicher Grundlage. 178-180. Wissenschaftliche Verlagessellschaft, Stuffgart.
172. Wiezorek, C., A. Kowalski and S. Zalewski.1993. Evaluation of crude oils pressed and extracted from seds of *Cucurbita pepo.* Polish J. Food Nutr. Sci. 2(2): 17-24.
173. Wojciechowska, B. 1987. Search for alkaloids in the seeds of *Cucurbita pepo* L. and *Cucurbita maxima* Duch. Acta Biol. Siles. 932:127-143.
174. Wojciechowska, B. 1990. Paper chromotagraphic search for alkaloids compounds in the seeds of *Cucurbita pepo* L. and *cucurbita maxima* Duch. Acta Biol. Siles. 1078:11-28.
175. Yoshida, H., Y. Omura and G. Kajimoto. 1988. Effects of microwave cooking on the molecular species of pumpkin seed triacylglycerols. Nutr. Rep. Int. 37(2):259-268.
176. Zdunczyk, Z., D. Minakowski, S. Frejnagel and M. Flis. 1999. comparative study of the chemical composition and nutritional value of pumpkin seed cake, soybean meal and casein, Nahrung 43(6):392-395.
177. Zeiner, W.F. 1971. Der Olkurbis - eine beachtenswerte Nutzpflanze. [Pumpkins - a noteworthy economic plant]. Deutsche Landw. Ges. Mitt. 86:(15):403-404.
178. Zeitoun, M.A.M., W.E. Neff, E. Selke and T.L. Mounts. 1991. analyses of vegetable oil triglyceride molecular species by reversed phase high performance liquid chromatography. J. Liquid Chromatogr. 14(14):2685-2698.
179. Zhou, X. 1987. Study on the breeding of naked kernel pumpkin and its genetic behavior. Acta Hort. Sin. 14(2):115-118.
180. Zipse, W. 1983. Der Einfluß von Genotyp und Anbau auf die Sameinhalfsstofe von Olkurbis (*Cucurbita pepo* L.). Ph.D. thesis, Universitat Gießen, Germany.
181. Zucker, H., V.W. Hays, V.C. Speer and D.V. Catron. 1958. Evaluation of pumpkin seed meal as a source of protein using a depletion-replention technique. J. Nutr. 65:327-334.

Cucurbit Genetics Cooperative Report 23:137-138 (article 46) 2000

Cucurbita Gene List Update - 2000

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Previous lists of *Cucurbita* genes were published in CGC Rpt. 15:102-109 (1992) and in CGC Rpt. 19: 91-92 (1996). Before publishing a proposed new gene symbol for a *Cucurbita* gene, researchers are urged to consult this gene list and those in CGC 15 and 19 in order to avoid using a symbol already assigned to another gene.

The following list of new *Cucurbita* genes also includes a correction for the erroneous symbol for the D^S gene that was given in CGC Rpt. 19: 91-92 (1996).

Gene	Character	Species	Reference
D^x	*Dark* stem *only*, fruit color not affected	*pepo*	4
De	*determinate* plant habit; stem lacking tendrils and terminating with female flowers	*moschata*	2
mo-1	*mature orange-1*; complementary gene for loss of green fruit color prior to maturity	*pepo*	5
mo-2	*mature orange-2*; complementary gene for loss of green fruit color prior to maturity	*pepo*	5
*Slc**	*Squash leaf curl* virus resistance	*pepo*	3
uml	*umbrella-like*; leaves shaped like *partially opened umbrella*	*maxima x pepo*	6
wc	*white corolla*; petals white, tending to curl, less dentate than normal	*maxima*	1
wyc	*white-yellow corolla*; dentate, white-yellow petals	*maxima*	1

Literature Cited

1. Korzeniewska, A. 1996. Two independent loci for white and white-yellow corolla in *Cucurbita maxima Duch.* In: M.L. Gomez-Guillamon, C. Soria, J. Cuarto, Tores,and R. Fernandez-Munox, eds. Proc. Cucurbitaceae Toward 2000: 6th Eucarpia Meeting Cucurbit Genetics Breeding. Graficas Axarquia, Velez-Malaga, Spain. pp 78-81.
2. Kwack, S.N. 1995. Inheritance of determinate growth habit in *cucurbita moschata* Poir. J. Korean Soc. Hort. Sci. 36:780-784.
3. Montes-Garcia, C.E., S. Garza-Ortega, and J.K. Brown. 1998. Inheritance of the resistance to squash leaf curl virus in *Cucurbita pepo.* In: J.D. McCreight, ed. Cucurbitaceae '98. Evaluation and Enhancement of Cucurbit Germplasm. A.S.H.S., Alexandria, Virginia. pp. 328-330.
4. Paris, H.S. 1996. Multiple allelism at the *D* locus in squash. J. Hered. 87:391-395.
5. Paris, H.S. 1997. Genes for developmental fruit coloration of Acorn squash. J. Hered. 88: 52-56.
6. Rakoczy-Trojanowska, M. and S. Malepszy. 1999. Inheritance of umbrella-like leaf shape in materials derived from *cucurbita maxima x C. pepo* hybrids. Cucurbit Genet. Coop. Rpt. 22:50-52.

Cucurbit Genetics Cooperative Report 23:139-139 (article 47) 2000

Gene Nomenclature for the Cucurbitaceae

1. Names of genes should describe a characteristic feature of the mutant type in a minimum of adjectives and/or nouns in English or Latin.
2. Genes are symbolized by italicized Roman letters, the first letter of the symbol being the same as that for the name. A minimum number of additional letters are added to distinguish each symbol.
3. The first letter of the symbol and name is capitalized if the mutant gene is dominant. All letters of the symbol and name are in lower case if the mutant gene is recessive, with the first letter of the symbol capitalized for the dominant or normal allele. (Note: For CGC *research articles*, the normal allele of a mutant gene is represented by the symbol "+", or the symbol of he mutant gene followed by the superscript "+", if greater clarity is achieved for the manuscript.
4. A gene symbol shall not be assigned to a character unless supported by statistically valid segregation data for the gene.
5. Mimics, i.e. different mutants having similar phenotypes, may either have distinctive names and symbols or be assigned the same gene symbol, followed by a hyphen and distinguishing Arabic numeral or Roman letter printed at the same level as the symbol. The suffix "-1" is used, or may be understood and not used, for the original gene in a mimic series. It is recommended that allelism tests be made with a mimic before a new gene symbol is assigned to it.
6. Multiple alleles have the same symbol, followed by a Roman letter or Arabic number superscript. Similarities in phenotype are insufficient to establish multiple alleles; the allelism test must be made.
7. Indistinguishable alleles, i.e., alleles at the same locus with identical phenotypes, preferably should be given the same symbol. If distinctive symbols are assigned to alleles that are apparent reoccurrences of the same mutation, however, they shall have the same symbol with distinguishing numbers or letters in parentheses as superscripts.
8. Modifying genes may have a symbol for an appropriate name, such as intensifier, suppressor, or inhibitor, followed by a hyphen and the symbol of the allele affected. Alternatively, they may be given a distinctive name unaccompanied by the symbol of the gene modified.
9. In cases of the same symbol being assigned to different genes, or more than one symbol designated for the same gene, priority in publication will be the primary criterion for establishing the preferred symbol. Incorrectly assigned symbols will be enclosed in parentheses on the gene lists.
10. The same symbol shall not be used for nonallelic genes of different *cucurbita* species. Allelic genes of compatible species are designated with the same symbol for the locus.

Literature Cited

1. CGC Gene List Committee, 1982. Update of cucurbit gene list and nomenclature rules. CGC 5:62-66.
2. Rieger, R., A. Michaelis and M.M. Green. 1976. Glossary of Genetics and cytogenetics (4th ed.). Springer-Verlag.
3. Robinson, R.W., H.M. Munger, T.W. Whitaker and G.W. Bohn. 1976. Genes of the Cucurbitaceae. HortScience 11:554-568.

Cucurbit Genetics Cooperative Report 23:140-148 (article 48) 2000

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12. **Boorsma, P.A.** Vegetable Research, S&G Seeds BV, Westeinde 62/Postbus 26, 1600 AA Enkhuizen, The Netherlands. Ph: (0228) 36 6390; Fax: (0228) 36-6160.
13. **Bosma, Monique.** ENZA ZADEN, De Enkhuizer Zaadh. b.v. Postbox 7, 1600 AA Enkhuizen, The Netherlands. Ph: 02280-315844; Fax: 02280-315960.
14. **Boyhan, George E.** University of Georgia, P.O. Box 8112, GSU, Statesboro, GA 30460. Ph: (912) 681-5639; Fax: (912) 681-0376; Email: *gboyhan@uga.edu*. Melon and watermelon breeding.
15. **Brown, Rebecca**. Dept. Hort., Oregon St. Univ., Ag Life Sci Bldg 4017, Corvallis, OR 97331. Ph: (541) 737-5462; Email:*brownr@bcc.orst.edu.* Virus resistance, *Cucurbita* germplasm, squash breeding.
16. **Bruton, Benny.** U.S. Dept. Agriculture, Agricultural Research Service, Lane, OK 74555. Ph: (530) 889-7395; Fax: (530)668-0219. Pickling cucumber breeding.
17. **Burkett, Al**. PetoSluis Co. Inc.37437 State Highway 16, Woodland, CA 95695. Tel.: (916) 666-0931; Fax: (916) 668-0219. Pickling cucumber breeding.
18. **Caglar, Gulat**. KSU, Ziraat Fakultesi, Bahee Bitkileri Bolumu, 46060, Kahramanmaras, Turkey. Ph: 90-344-237666/284; Fax: 90-344-2230048; Email: *ghulat99@excite.com.* Cucumber breeding.
19. **Carey, Edward E**. International Potato Center (CIP), P.O. Box 25171, Nairobi, Kenya. Ph: 254-2-632054; Fax: 254-2-631499/630005; Email: *tcarey@cgnet.com.* Breeder with interest in cucurbits.
20. **Carle, R. Bruce.** UF Mid-Florida Res & Educ Ctr, 2725 Binion Road, Apopka, FL 32703-8504. Ph: (407) 884-2034; Email: *rbcwm@gnv.ifas.uf.edu*. Watermelon and squash breeding.
21. **Chen, Fure-Chyi.** Dept. Plant Industry, Natl. Pingtung Univ. Sci. & Techn., Neipy, Pingtung 91207, Taiwan. Rep. China. Ph: 886-8-774-0267; Fax: 886-8-774-0371; Email: *furechen@mail.npuyst.edu.tw.* Gene transfer, breeding, tissue culture and isozymes.
22. **Ching, Alejandro (Alex)**. Alternative Crops Res Ctr, NW MO St U, 106 Valk, 800 Univ Dr,Maryville, MO 64468.Ph: (660) 562-1126; Fax: (660) 562-1621; Email: *alching@mail.nwmissouri.edu.* Breeding & introduction of new cucurbits. Production & nutritional quality.
23. **Chung, Paul**. Seminis Vegetable Seeds, Inc. 37437 State Highway 16, Woodland, CA 95695. Ph: (530) 666-0931; Fax: (530) 668-0219.
24. **Coffey, Robyn.** Willhite Seed Inc., P.O. Box 23, Poolville, TX 76487. Ph: (817) 599-8656; Fax: (817) 599-5843; Email: *robyn@willhiteseed.com.*

25. **Cohen, Ron**. Newe Yaar Experiment Station, P.O. Box 1021, Ramat Yishayt 30095, Israel. Ph: 972-4-953-9516; Fax: 972-4-983-6936. Plant pathology; root and foliar diseases of cucurbits.
26. **Cohen, Yigal**. Department of Life Sciences, Bar-Ilan University, Ramat-Gan 52 100, Israel. Ph:: +9723-5318251; Fax: +9723-6771088. Melon.
27. **Cook, Kevin L.** Novartis Seeds, Inc., Veg-NAFTA, 10290 Greenway Road Naples, FL 33961. Ph: (941) 775-4090; Fax: (941) 774-6852. Email: *kevin.cook@seeds.novartis.com.* Breeding of summer squash.
28. **Corella, Pilar**. Asgrow Spain S.L., Paraje San Nicola s/n 04547 La Mojonera, Almeria, Spain. Ph: 34-51-580012; Fax: 34-51-581162.
29. **Coyne, Dermot P.** Department of Horticulture, University of Nebraska, Lincoln, NE 68583-0724 Ph: (402) 472-1126; Fax: (402) 472-8650. Breeding and genetics of squash.
30. **Cramer, Chris.** Dept. Agron. & Hort. NMSU, P.O. Box 30003, Dept. 3Q,Las Cruces, NM 88003-8003. Ph: (505) 646-3405; Email: *ccramer@nmsuvml.nmsu.edu.* Cucumber yield, yield components,combining ability, heterosis and recurrent selection.
31. **Crino, Paoli.** ENEA C.R. Casaccia, Biotech & Agr Div, Via Anguillarese 301, Roma, 00060, Italy.
32. **Cui, Hongwen.** Dept. Horticulture, Northwestern Agric. Univ., Yangling, Shaanxi 712100, P.R. China. Cucumber breeding.
33. **Dane, Fenny.** Dept. Horticulture, Auburn University, Auburn, AL 36849. Ph: (334) 844-3047; Fax: (334) 844-3131; E-mail: *fdane@acesag.auburn.edu. Citrullus* genomics.
34. **Danin-Poleg, Yael.** A.R.O., Newe Ya'ar Expt. Station, P.O. Box 1021, Ramat Yishay 30095, Israel. Ph: 972-4-9539553/4; Fax: 972-4-9836936; Email: *geneweya@netvision.net.il*
35. **Davis, Angela.** USDA, ARS, P.O. Box 159m Hwy 3 West, Lane, OK 7455. Ph: (580) 889-7395; Fax: (580) 889-5783; Email: *adavis-usda@lane-ag.org* Germplasm improvement.
36. **de Groot Erik.** Breeding, Sementi Nunhems S.R.L., Via Ghiarone, 2, 40019 S. Agata Bolognese, Italy
37. **DeLangen, Frank.** Mas St. Pierre, 13210 St Remy de Provence, France. Email: *frank.delangen@clause.fr.*
38. **de Ruiter, A.C.** de Ruiter Zonen CV, Postbus 1050, 2660 BB Bergschenhoek, The Netherlands. Ph: 010-5292204; Fax: 010-5216125. Breeding and seed production of cucumbers.
39. **Decker-Walters, Deena.** The Cucurbit Network. 11901 Old Cutler Road, Miami, FL 33156-4242. Ph: (305) 667-3800; Fax: (305) 661-5984; Email: *walters@servax.fiu.edu.* Communication via The Cucurbit Network; the whole family Cucurbitaceae.
40. **Della Vecchia, Paulo T.** Agroflora S/A, Caixa Postal 427, 12.900-000 Braganca, Paulista SP, Brazil. Ph: (011) 7871-0855; Fax: (011) 7843-6572. Breeding & genetics, seed production and disease resistance of melon and squash.
41. **Denlinger, Phil.** Mt. Olive Pickle Co., Inc., P.O. Box 609, Mount Olive, NC 28365. Ph: (919) 658-2535; Fax: (919) 658-6090; Email: *cn1713@coastalnet.com*.
42. **Dhaliwal, Major Singh.** Dept. of Vegetable Crops, L.S. & F. Punjab Agriculture University, Ludhiana-141001, Punjab, India.
43. **DiNitto, Louis**. Sunseeds, 8850 59th Ave., N.E., Brooks, OR, 97305. Ph: (503) 393-3243, Fax: (503) 390-0982. Melon (*Cucumis melo*).
44. **Dogimont, C.**,INRA, St. Maurice, BP 94, 84143 Montavet, France.
45. **Drowns, Glenn**. Sand Hill Preservation Center, 1878 230th Street, Calamus, IA 52729. Ph: (319) 246-2299; Email: *gdrowns@cal-wheat.k12.ia.us*. Genetic preservation of all cucurbits. Taxonomy of *Cucurbita moschata* and *Cucurbita argyrosperma.*
46. **Duangsong, Usa.** Limagrain Veg. Seeds Asia, 119/9 Moo 1, Baan Khao, Muang, Kanchanaburi 71000, Thailand. Ph: 66-2-636-2521-1; Fax: 66-2-636-2524; Email: *songusa@loxinfo.co.th.*
47. **Eigsti, Orie J.** 1602 Winsted, College Green, Goshen, ID 46526. Ph: (219) 533-4632. Fusarium wilt resistance in tetraploid *Citrullus lanatus* lines, to eliminate crop rotation.
48. **El Jack, Ali Elamin**. Dept. Horticulture, Fac. Agric. Sciences, University of Gezira, Wad-Medani, P.O. Box 20, Sudan. Email: *nahla_2elamin@yahoo.com*.
49. **Elmstrom, Gary.** c/o Sunseeds, 7078 E. Peltier Road, Acampo, CA 95220. Ph: (209) 367-1064; Fax: (209) 36701066; Email: *gary.elmstrom@sunseeds.com*. Triploid watermelon breeding.
50. **Ezura, Hiroshi**. Plant Biotech Inst, Ibaraki Agricl Ctr, Ago, Iwama, Nishi-ibaraki, 319-0292, Ibaraki, Japan. Ph: 0299-45-8330; Fax: 0299-8351; Email: *ezura@nocs.tsukuba-noc.affrc.go.jp*.
51. **Ficcadenti, Nadia.** Res Inst Veg Crops, Sect Ascoli Piceno, Via Salaria 1 - Monsampolo del Tronto, Ascoli Piceno, 6-3-3-, Italy. Ph: +39 735 701706; Fax: +39 735 703684; Email: *orticolt @insinet.it.*
52. **Fito, Laia.** Plant molec Mrker & Pathol Dept, Semillas Fito S.A., c/Selva de Mar, III, 08019 Barcelona, Spain. Ph: +34 (9)3 303 63 60; Fax: +34 (9)3 307 03 64; Email: *eulalia@fito.es.* Disease resistance and quality of melons (esp. Spanish) & cucumber; breeding schemes & genetic markers.
53. **Funakushi, Hisashi.** Mikado Seed Growers Co., Ltd., 1203 Hoshikuki, Chuo-Ku, Chiba City 260, Japan. Ph: 81-43-

265-4847; Fax: 81-43-266-6444.

54. **Gaba, Victor.** Dept. Virology, Inst. Plant Protection, A.R. O.,Volcani Center, P.O.B. 6, Bet Dagan 50250, Israel. Ph: 972-3-9683568/9; Fax: 972-3-9604180; E-mail:*vpgaba@agri.gov.il*. Tissue Culture & Transformation.
55. **Gabert, August C.** Sunseeds, 8850 59th Avenue NE, Brooks, OR 97305-9625. Ph: (503) 393-3243; Fax: (503) 390-0982; Email: *agabert%sunseeds@mcimail.com.* Cucumber and summer squash breeding and genetics.
56. **Ganapathi, A.** Dept. Biotechnology, Bhjarathidasan University, Tiruchirappalli - 620024, India. Ph: 91-0431-660386; Fax: 91-0431-660245; Email: *ganap@bdu.erhet.in*.
57. **Garza Ortega, Sergio.** Univ. Sonora, Dept Agric y Ganaderia, Iturbide #32 Jalisco/N. Heroes, Hermasillo, Sonora 83040, Mexico. Ph: (62) 13-80-06; Fax: (62) 13-80-06; Email: *sgarza@rtn.uson.mx.* Breeding of *Cucurbita* spp.: testing of new muskmelon lines.
58. **Gatto, Gianni.** Esasem Spa, via San Biagio 25, 37052 Casaleone (VR), Italy. Ph: 0442/331633; Fax: 0442/330834; Email: *GGatto@esasem.it.*
59. **Gomez-Guillamon, M. Luisa**. Estacion Experimental "La Mayora", 29750 Algarrobo-Costa, Malaga, Spain. Ph: (952) 51 10 00; Fax: (952) 51 12 52; E-mail: *guillamon@mayora.csic.es.*
60. **Green, C. Ed.** Seminis Vegetable Seeds, Inc. 37437 State Highway 16, Woodland CA, 95695. Ph: (530) 666-09031; Fax: (530) 668-0219.
61. **Groff, David.** Asgrow Seed Company, Rt. #1, Box 1907, Omega TyTy Road, Tifton, GA 31794. Ph: (912) 386-8701; Fax: (912) 386-8805. Breeding of squash, cucumber, melon and watermelon.
62. **Grumet, Rebecca.** Dept. Hort., Plant & Soils Building, Michigan State University, East Lansing, MI 48824-1325. Ph: (517) 353-5568; Fax: (517) 353-0890; E-mail: *grumet@pilot.msu.edu.* Disease resistance, gene flow, tissue culture and genetic engineering.
63. **Gupta, Satish. C.** Reitzel India Ltd., 220 Agil Campus, Whitefield Post, Bangalore 560066, India. Ph: 91-080=8452415; Fax: 91-080-8453063.
64. **Hagihara, Toshitsugu**. Hagihara Farm Co., Ltd., 984 Hokiji, Tawaramoto, Shjiki Nara, 636-0222, Japan. Ph: 07443-3-3233; Fax: 07443-3-4332; Email: *cucurbit@mahoiroba.ne.jp.*
65. **Haim, Davidi**. Hazera Quality Seed Ltd., Mivhor Farm Doar, Sede Gat 79570, Israel.
66. **Han, Sang Joo.** Novartis Seeds Co., Ltd. 8tyhfl. SungAm Bldg. #114, Nonihyun-don, Kangnam-ku, Seoul, Korea 135-010. Ph: +82 2 3218 5400; Fax: +82 2 516 2286; Email: *sangjool.han@seeds.Novartis.com.* Disease resistance.
67. **Hassan, Ahmed Abdel-Moneim**. Department of Vegetable Crops, Fac. Agriculture, Cairo University, Giza, Egypt. Ph: 724107 & 724966. Cucumber, melon, Squash & watermelon germplasm evaluat8ion and breeding for disease resistance, incl. viruses.
68. **Havey, Michael J.** USDA/ARS, Department of Horticulture, University of Wisconsin, Madison, WI, 53706. Ph: (608) 262-1830; Fax: (608) 262-4743; Email: *mjhavey@facstaff.wisc.edu.*
69. **Hentschel, Richard.** Pickle Packers International, Inc. One Pickle and Pepper Plaza, P.O. Box 606, St. Charles, IL 60174-0606. Ph: (630) 584-8950; Fax: (630) 584-0759; Email: *staff@ppii.org.* Trade Association for pickle vegetables, primarily cucumbers, peppers and cabbage.
70. **Herman, Ran**. "Zeraim" Seed Growers Company Ltd., Department of Breeding, Gedera 70 700, Israel. Ph: 08-59 27 60; Fax: 08-59 43 76.
71. **Hertogh, K.** Nickerson-Zwaan b.v., Postbus 28,4920 AA Made, The Netherlands. Ph: 31(0)62 690900; Fax: 31(0)162 680 970; Email: *seeds@nickerson-zwaan.nl.*
72. **Himmel, Phyllis.** Asgrow-Seminis Vegetable Seeds, 37437 State Highway 16, Woodland, Ca 95695. Ph: (530) 669-6182; Email: *phyllis.himmel@svseeds.com.* Viral diseases of cucurbits.
73. **Hirabayashi, Tetsu.** Nihon Horticultural Production Inst., 207 Kamishki, Matsudo-shi, Chiba-ken, Japan. Ph:: 0473-86-1455; Fax: 0473-86-1455. Varietal improvement of cucurbit crops, especially melon, cucumber and pumpkin.
74. **Hollar, Larry. A.** Hollar & Co., Inc., P. O. Box 106, Rocky Ford, CO 81067. Ph: (719) 254-7411; Fax: (719) 254-3539;.Email: *lahollar@iguana.ruralnet.net.* Cucurbit breeding and seed production.
75. **Holle, Miguel.** CALCE 2, #183 Urb. El Rancho, Miraflores - Lima 18, Peru. Ph: 51-14-383749; Fax: 51-14-351570; Email: *mholle@cgnet.com.* Plant genetic resources.
76. **Holman, Bohuslav.** Bzinska Str. 1420, Bznec, CZ-696 81, Czech Republic Ph: +420-631-384470; Fax: +420-631-384972; Email: *bholman@iol.cz.* Cucumber breeding and seed production.
77. **Humaydan, Hasib.** Ag Consulting International. 317 Red Maple Drive, Danville, CA 94506. Ph: (510) 736-1241; Fax: (510) 736-1241.
78. **Hutton, Mark.** Sakata Seed America, PlO. Box 1118, Lehigh Acres, FL 33970-1118. Ph: (941) 369-0032; Fax: (941) 369-7528; Email: *mhuttoin@sakata.com.* Squash breeding and cultivar development.
79. **Iamsangsri, Suphot.** Limagrain Veg. Seeds Asia, 119/9 Moo 1, Baan Khao, Muang, Kanchanaburi 71000, Thailand. Ph: 66-2-636-2521; Fax: 66-2-636-2524.
80. **Ibrahim, Aly M.** USDA-ARS, 1636 E. Alisal St. Salinas, CA 93905. Fax: (408) 753-2866. Cucumber, Melon,

Watermelon.

81. **Ignart, Frederic.** Centre de Recherche TEZIER, Route de Beaumont, Domaine de Maninet, Route de Beaumont, 26000 Valence, France. Ph: (33) 75575757; Fax: (33) 75552681. Squash and melon breeding.
82. **Ikegami, Talayuki.** Sakata Seed Corp., 1743-2 Yoshioka, Kakegawa, Shizuoka, 436-0537-26-1115,Japan. Ph: 81-0537-26-1111; Fax: 81-0537-26-1110. Cell biology.
83. **Ito, Kimio.** Vegetable Breeding Laboratory, Hokkaido Natl. Agric. Expt. Sta. Hitsujigaoka. Sapporo, Japan 062-8555. Ph: 011(851)9141; Fax: 011(859)2174; Email: *kito@cryo.affrc.go.jp.*
84. **Jhan, Molly Kyle.** Cornell Univ., Dept. Plant Brdng, 312 Bradfield Hall, Ithica, NY 14853-1902. Ph: (607) 255-8147; Fax: (607) 255-6683; Email: *mmk9@cornell.edu.* Melon and squash breeding and genetics.
85. **Jain, Jaagrati** B-149, M.P. Enclave, Pitampura, Delhi - 110034, India. Melon genetics and tissue culture.
86. **Jiang, Jiping.** Seminis Vegetable Seeds, 37437 State Highway 16, Woodland, CA 95695. Ph: (530) 666-0931; Fax: (530) 668-0219. Developing disease screens for fungal diseases of cucurbits.
87. **Johnston, Rob Jr.** Johnny's Selected Seeds. Foss Hill Road, Albion, ME 04910-9731. Ph: (207) 437-9294; Fax: (207) 437-2603; Email: *rob@johnnyseeds.com* Squash and pumpkins.
88. **Kampmann, Hans Henrick.** Breeding Station Danefield, Odensevej 82, 5290, Marslev, Denmark. Ph: 65 95 17 00; Fax: 65 95 12 93.
89. **Kanda, Minoru.** Kanda Seed Co., Ltd., 262 Shinga, Kashihara, Nara, 634-0006, Japan. Ph: 0744-22-2603; Fax: 0744-22-9073; Email: *minojazz@mue.biglobe.ne.jp.*
90. **Karchi, Zvi.**74 Hashkedem St. Kiryat-Tivon 36501, Israel.Ph: 04-9830107; Fax: 972-4-9836936. cucurbit breeding, cucurbit physiology.
91. **Kato, Kenji.** Fac, Agriculture, Okayama Univ., 1-1-1- Tsushima Naka, Okayama, 700 Japan. Ph: 81-86-251-8323; Fax: 81-86-254-0714; E-mail: *kenkato@ccws2.cc.okayama-u.ac.jp* Use of molecular markers for QTL mapping and cultivar identification in melon.
92. **Katzir, Nurit.** Newe Ya-ar Research Center, ARO, P.O. Box 1021, Ramat Yishay, 30095, Israel. Ph: 972-4-9539554; Fax: 972-4-9836936; Email: *nuritkat@netvision.net.il*
93. **Keita, Sugiyama.** Kurume Branch, Natl Res Inst. Veg/OrnPlnts/Tea,Kurume, Fukuoka 839-8503, Japan. Ph: +81-942-43-8271; Fax: 81-942-43-7014; Email: *keita@nivot-krm.affre.go.jp.* Watermelon.
94. **Kerje, Torbjorn.** IPGRI, c/o ICRAF, PO Box 30677, Nairobi,Kenya. Ph: 254-2-521514; Fax: 254-2-521209; Email: *t.kerje@cgiar.org.* Genetic diversity of *Cucurbita* and *Cucumis* in Southern Africa.
95. **King, Joseph J.** Seminis Vegetable Seeds, Inc. 37437 State Highway 16, Woodland, CA 95695.Ph: (530) 666-0931; Fax: (530) 666-5759; Email: *joe.king@svseeds.com.* Genetics and breeding of melon, cucumber and squash.
96. **King, Stephen R.** Seminis Vegetable Seeds, Inc. 37437 State Hwy. 16, Woodland, CA 95695. Ph: (530) 666-0931; Fax: (530) 668-0219; E-mail: *srking2570@aol.com.* Melon breeding.
97. **Kirkbride, Joseph H, Jr.** USDA-ARS. System. Bot. & Mycol. Lab. Rm 304 Bldg.011A, BARC-Wast, Beltsville, MD 20705. Ph: (301) 504-9447; Fax: (301) 504-5810; Email: :*jkirkbride@asrr.arsusda.gov.* Systematic taxonomy of the Cucurbitaceae.
98. **Klapwijk, Ad.** De Ruiter Zonen CV, Postbus 1050, 2660 BB Bergschenhoek, The Netherlands. Ph: 010-5292253; Fax: 010-5292410.
99. **Knerr, Larry D.** Shamrock Seed Company, Harris Place, Salinas, CA 93901-4586. Ph: (831) 771-1500; Fax: (831) 771-1517; E-mail: *ldknerr@aol.com.* Varietal development of honeydew.
100. **Konno, Yoshihiro.** Asahi Ind., Biol. Engin. Lab., 222 Wataruse, Kamikawa-machi,Kodama-gun, Saitama 367-0394, Japan. Ph: 81-274-52-6339; Fax: 81-274-52-4534; Email: *y.konno@asahi-kg.co.jp.* Watermelon breeding.
101. **Kouters, Jolanda.** East-West Seed, Farm Lert Phan, 7 Moo 9, Tambon Maefaek Mai, Amphur Sansai, Chiangmai, 50290 Thailand. Ph: (66) 53-848610; Fax: (66) 53-848611; Email: *research.tyh@eastwestseed.com.* Watermelon breeding.
102. **Kraakman, Peter.** DeRuiter Zohen, Torre Caribe 7D, Aguadulce, (Almeria) Spain. Email: *Peter.Kraakman@deruiterseeds.com*.
103. **Kristkova, Eva.** Res Inst Crop Prod, Praha Ruzyne, Workplace Olomouc, Slechtitelu 11, 738 71 Olomouc, Czech Republic. Ph: +420-68-5228355; Fax: +420-68-5228355; Email: *olgeba@ova.,pvtnet.vz.* Gene bank curating of cucurbitaceous vegetables; powdery mildew resistance in *Cucurbita.*
104. **Kuginuki, Yashuhisa.** National Institute Veg/Orn/Tea, Crop Research Station, Ano, Mie 514-2392, Japan. Ph: 0592-68-1331; Fax: 0592-68-1339. Breeding for resistance to disease.
105. **Kuhlmann, Hubert**. Fink GmbH, Benzstrasse 25, D-71083 Herrenberg, Germany. Ph: (07032) 922-122; Fax: (07032) 922-202.
106. **Kuti, Joseph O.** Dept. Agron & Res Sci, Hort Crops Lab, Texas A&M University, Kingsville, Kingsville, TX 78363. Ph: (361) 593-3978; Fax: (361) 593-3788; Email: *f-kuti@tamuk.edu.* Breeding and genetics; host-parasite interrelationships; postharvest physiology.

107. **Kwack, Soo-Nyeon**. Dept Hort Breeding, Mokpo Natl Univ., Dorimri, Chonggyemyun, Muangun, Chonnam 534-729, Korea.
108. **Kwon, Cheol-Sang.** Pusan Breeding Inst, Choong Ang Seed, 648-2 Kangdong-dong Kangseo-gu, Pusan, Korea 618-300. Ph: +82-51-972-8014; Fax: +82-51-972-3206; Email:*kl 3483@chollian.net.* Cucumber, watermelon, melon and squash breeding.
109. **Kwon, Young-Seok**. Natl. Alpine Agric. Expt. Sta., 1 Hoengkeri, Doam, Pyongchang, Kangwondo, Rep. Korea 232-950/ Ph: 82-374-330-7811; Fax: 82-374-330-7715; Email: *yskwon@naaes.go.kr.* Watermelon germplasm, evaluation and breeding for disease resistance.
110. **Lebeda, Ales.** Palacky University, Faculty of Science, Department of Botany, Slechtitelu 11, 783 71 Olomouc, Czech Republic. Ph: +420/68/5223325; Fax: +420/68/5241027; Email:*lebeda@risc.upol.cz.*
111. **Lecouvior, Michel.** Clause Semences, 1, Avenue L. Clause, 91221 Bretigny-sur-Orge, CEDEX France. Fax: (33) 04.90.92.21.55; Email: *Michel.le-couviour@Rhone-poulenc.com.* .
112. **Lehmann, Louis Carl.** Dept. Plant Breeding Research, Swedish Univ. Agricultural Sci., S-268 31 Svaloev, Sweden.. Ph: ++46-418-667200; Fax: ++46-418-67081; E-mail: *louis.lehmann@vf.slu.se. Cucurbita* - testing of squash and pumpkin for use in Southern Sweden.
113. **Lelly, Tamas.** Inst. Agrobiotech, Dept Plant Biotech, Konrad Lorenz Str 20, Tulln, Lower Austria, Austria 3430. Ph: +43 2272 66280 204; Fax: +43 2272 66280 77; Email: *lelly@ifa-tull.ac.at. Cucurbita* spp.
114. **Levi, Amnon.** U.S. Vegetable Laboratory, 2875 Savannah Highway, Charleston, SC 29414. Ph: (843) 556-0840; Fax: (834) 763-7013; Email: *alevi@awod.com*.
115. **Lin, Depei**. Sichuan Acdemic Agric. Science, Institute of Horticulture, Chengdu 61006, People's Rep. China. Ph: (028) 4791732; Fax: (028) 4442025. Watermelon, melon and *Cucurbita* breeding.
116. **Liu, Wenge.** Zhengzhou Fruit Research Inst, chinese Academy of Agric Sci, Zhengzhou, Henan, P.R. China 450009. Ph: (0371) 6815703; Fax: (0371) 6815771; Email: *wlirong@public2.zz.ha.cn.* Watermelon breeding,male sterility, tetraploids, triploids.
117. **Lopez, Anido, Fernando.** Catedra de Genetica, Fac. de Cs. Agrarias, UNR, CC 14, 2123 Zavalla, Argentina. Ph: 54--970080; Fax: 54-1-970085; Email: *felopez@fcagr.unr.edu.ar.* Breeding of *Cucurbita pepo* L. (caserta type).
118. **Love, Stephen Loyd.** Aberdeen R&E Center, P.O. Box AA, Aberdeen ID, 83210. Ph: (208) 397-4181; Fax: (208) 397-4311; E-mail: *slove@uidaho.edu.* Small scale private watermelon breeding with emphasis on adaptation to cold climates.
119. **Lower, Richard. L.** Coll. Agriculture, Univ. Wisconsin, 1450 Linden Drive, Room 240, Madison, WI 53706. Ph:: (608) 262-2349; Fax: (608) 265-6434; E-mail: *richard.lower@ccmail.adp.wisc.edu.* Effects of plant type genes on yield, sex-expression, growth parameters, pest resistance & adaptability.
120. **Loy, J, Brent.** Plant Biology Dept, Nesmith Hall, Univ. New Hampshire, Durham NH 03824. Ph: (603) 862-3216; Fax: (603) 862-4757; Email: *jbloy@cisunix.unh.edu.* Squash, melon, pumpkin. Genetics, breeding, plasticulture, mulch, rowcovers.
121. **Maggs, Gillian.** National Herbarium (WIND), NBRI, Private Bag 13184, Windhoek, Namiia. Ph: +264 61 3029111; Fax: +264 61 3022177; Email: *gillianm@lianam.lia.net.*
122. **Maluf, Wilson Roberto.** Dept. de Agricultra/UFLA, Caixa Postal 37, 37200-000, Lavras-MG, Brazil. Ph: (035) 8291301; Fax: (035) 829-1301; E-mail: *wrmaluf@ufla.br.* Cucumbers, melons, squashes.
123. **Markiewicz-Ladd, Krystina.** Polonica International, P.O. Box 2305, Gilroy, CA 95021. Ph: (408) 842-1022; Fax: (408) 675-1022; Email: *polonica@aol.com.* Melons - breeding, new germplasm, postharvest physiology, biotechnology, cultural practices, new diseases.
124. **Martyn, Ray D.** Dept. Botany & Plant Pathology, 1155 Lilly Hall, Purdue Univ., West Lafayette, IN 47907-1155. Ph: (765) 494-4615; Fax; (765) 494-0363; E-mail: *martyn@btny.purdue.edu*. Soilborne diseases of watermelon and melon, particularly the Fusarium wilts and vine declines.
125. **Matsuura, Seiji.** Kiyohara Breeding Sta. Tohoku Seed Co., 1625, Nishihara, Himuro, Utsunomiya, Japan. Ph: 0286-34-5428; Fax: 0286-35-6544.
126. **Maynard, Donald N.** University of Florida, 5007 60th Street East, Bradenton, FL 34203. Ph: (941) 751-7636; Fax: (941) 751-7639; Email: *bra@gnv.ifas.ufl.edu.*. Tropical *moschata* improvement; watermelon variety evaluation and production practices.
127. **Mazereeuw, J.P.** SETO A.S. Cebecoy Caddesi, Akasya Aopt. 45/1, 07100, Antalya, Turkey.
128. **McClurg. Charles A.** University of Maryland, Dept. Natural Resource Sci., College Park, MD 20742-4452.Ph: (301) 405-4342; Fax: (301) 314-9308; E-mail: *cm19@umail.umd.edu*. Production and culture of cucurbit crops.
129. **McCreight, J. D.** USDA-ARS, 1636 E. Alisal St., Salinas, CA 93915.Ph: (831) 755-2864; Fax: (831) 755-2814; E-mail: *jmcreig@pwa.ars.usda.gov.* Melon breeding and genetics.
130. **McGrath, Desmond John.** Dept. Primary Ind., Hortic. Res. Sta., P.O. Box 538, Bowen. Queensland,4805. Australia. Ph: + 61-7-4785 2255; Fax: + 61-7-4785 2427; Email: *mcgratdj@prose.dpi.qld.gov.au.* Disease resistance in *Cucumis*

melo, particularly gummy stem blight.

131. **Meadows, Mike.** Novartis Seeds, Inc. 10290 Greensway Road, Naples, FL 33961. Ph: (941) 775-4090; Fax: (941) 774-6852; Email: *Mike.Meadows@GWA.Sandoz.com.* Vegetable diseases.
132. **Melendez, Roberto Compean.** Heriberto Valdez 647 PTE., C.P. 81200, Los Mochis, Sinaloa, mexico. Ph: (68) 18-37-22.
133. **Merrick, Laura. C.** Dept. Agron., Iowa St. Univ., G207 Agronomy Hall, Ames, IA 50011-1010. Ph: (515) 294-7636; Fax: (515) 294-3613; Email: *lmerrick@agron.iastate.edu. Cucurbita* evolution; cucurbit germplasm, evaluation and conservation; ethnobotany and evolution.
134. **Milerue, Sompong**. Peto Thailand, P.O. Box 171, 99 Moo 2, Wiang-Y, Mae Gorn, A Muang Chiang Rai 57000, Thailand.
135. **Mochizuki, Tatsuya.** Kurume Br, Natl Res Inst Veg/Orn/Tea, 1823 Mii-machi, Kurume, Fukuoka 830, Japan. Ph: 00942-43-8271; Fax: 0942-43-7014.
136. **Mohamed, El Tahir Ibrahim.** PGR unit/Horticulture, Agr Res Corp, P.O. Box 126, Wad Medani, Sudan.
137. **Mohamed, Yousisf Fadlalla.** Dept. Plant Pathol, Fac. Agric Sci, University of Gezira, Wad Medani, P.O. Box 20, Sudan.
138. **Moraghan, Brian Joseph.** Asgrow Vegetable Seeds, P.O. Box 667, 13270 Rockpile Rd., Arvin, CA 93203. Ph: (805) 854-2360; Fax: (805) 854-4379; Email: *brian.moraghan@svseeds.com.* Melon and watermelon breeding and disease resistance.
139. **Morelock, Ted** Dept. Horticulture & Forestry, University of Arkansas, Fayetteville, AR, 72701. Ph: (501) 575-2603; Fax: (501) 575-8619; E-mail: *morelock@comp.uark.edu.* Cucumber breeding.
140. **Munger, H.M.** Cornell University, 252 Emerson Hall, Ithica NY 14853. Ph: (607) 255-7820; Fax: (607) 255-6683; Email: *hmm11@cornell.edu.*. Cucurbit breeding and disease resistance.
141. **Nadel, Michael.** 10851 Woodbine Street, Los Angeles, CA 90034. Ph: (310) 838-7675; Fax: (310) 202-7466; Email: *dansonseed@mediaone.net.* Breeding summer squash, cucumbers, melons and watermelons.
142. **Nannes, Jeroen**. Seminis Vegetable Seeds, P.O. Box 93, 2675 ZH Honselersdijk, The Netherlands. Email: *jnannes@scseeds.nl.* Breeding slicing cucumber.
143. **Navazio, John.** Chriseed, P.O. Box 98, Mount Vernon, WA 98273-0098. Ph: (360) 336-9727; Fax: (360) 424-9520. Breeding for increased pigments in cucurbits, carrots and beets.
144. **Nea, Larry**. Seminis Vegetable Seeds, 37437 State Highway 16, Woodland, CA 95695. Ph: (530) 666-0931; Fax: (530) 668-0219. Cucumbers, melons, squash, watermelon.
145. **Ng, Timothy J.** Dept. Natural Resource Sci., University of Maryland, College Park, MD 20742-4452 .Ph: (301) 405-4345; Fax: (301) 314-9308;. Email: *tn5@umail.umd.edu*. Melon breeding and genetics; postharvest physiology; seed germination.
146. **Niemirowicz-Szczytt, Katarzyna.** Warsaw Ag Univ, Dept Gen & Plt Brdng, ul Nowoursynowska 166, 02-766 Warsaw, Poland. Ph: (48-22) 843 09 82; Fax: (48-22) 843 90 61; Email: *niemirowicz@alpha.sggw.waw.pl.* Cucumber, melon, winter and summer squash, watermelon - genetics, breeding, tissue culture, biotechnology.
147. **Norton, Joseph D.** Dept. Horticulture, 101 Funchess Hall, Auburn Univ. Auburn, AL. 36849. Ph: (205) 844-3031; Fax: (205) 844-3131. Breeding and genetics of melon and watermelon.
148. **Nuez, Fernando.** Cat. de Genetica, ETS Ingen. Agron., Univ. Politenica, Camino de Vera, 14, 4620 Valencia, Spain. Ph: 34 (6) 387-74-21; Fax: 34 (6) 387-74-29; Email: *fnuez@btc.upv.es.* Genetics and plant breeding.
149. **Oliveira de Paiva, Waldelice.** EMBRAPA/CNPAT - Caixa Postal 3761, Rua Dra. Sara Kmesquita 2270, 60511-110-Fortaleza-Ceara, Brazil. Ph: (085) 299.18.01; Fax: (085) 299.18.03;Email: *Walde@cnpat.embrapa.br.* Research with cucurbit species, especially *Cucumis*, and particularly *Cucumis melo.*
150. **Om, Young-Hyuan**. National Horticultural Res Inst, 475 Imok-dong, Suwon 440-310, Republic of Korea. Ph: 82-0331-290-6171; Fax: 82-0331-295-9548; Email: *omyh@nhri.go.kr.* Breeding of cucurbit vegetables.
151. **Omara, Sadig Khdir.** Dept. Horticulture, Fac. Agric. Sci., University of Gezira, Wad-Medani, P.O. Box 20, Sudan.
152. **Ouyang, Wei**. United Genetics Seeds Co., 18 W. Hacienda Lane, Woodland, CA 95695. Ph: (707) 693-6815; Fax: (707) 693-6814; Email: *weiouyang1@yahoo.com.* Squash breeding.
153. **Owens, Ken**. United Genetics Seeds Co., 8000 fairview Road, Hollister, CA 95023. Ph: (831) 636-4882; Fax: (831) 636-4883; Email: *kobreeding@hotmail.com*. Cucumber breeding.
154. **Palomares, Gloria**. Dept Biotechnologia, Univ Politecnia, Camino de Vera, s/n., E-46022 Valencia, Spain. Ph: 34(6)387- 7420; Fax: 34(6)387- 7429;. Email: *gpaloma@btc.upv.es.* Genetic improvement in horticultural plants.
155. **Paris, Harry**. Dept. Vegetable Crops, A.R.O., Newe Ya'ar Research Ctr, PO Box 1021, Ramat Yishay 30-095, Israel . Ph: 972-4-894516; Fax: 972-4-836936; Email: *hsparis@netvision.net.il.* Breeding and genetics of squash and pumpkin.
156. **Pathak, Chandra.** c/o Nath Sluis Ltd., Nath House, Nath Road, Aurangabad, 431005, India.
157. **Piero Abril, Jose Luis.** Apartado de Correos no. 2,04720 Aguadulce, Almeria, Spain. Fax: 34-50 34 34 01.

158. **Perl-Treves, Rafel.** Dept. Life Science, Bar-Ilan University, Ramat-Gan, Israel 52900. Ph: 972-3-5318249; Fax: 972-3-5351824; Email: *perl@bhrosh.cc.biu.ac.il.*.
159. **Peter, K.V.** Natl. Ressearch Ctr for Spices, ICAR, Post Bag No. 1701, Marikunnu P.O. Calicut - 673 012, Kerala, India. Ph: 011-91-4925-258457.
160. **Peterson, Paul S.** Plant Pest Diagnostic Center, 3294 Meadowview Road, Sacramento, CA 9583201448. Ph: (916) 262-1139; Fax: (916) 262-1190; Email: *ppeterso@cdfa.ca.gov.* Laboratory germinationand seed quality assessment.
161. **Picard, Florence.** Vilmorin, Route du Mano9ir, 49-250 La Menitre, France. Email: v*cilmorin01@brettcomp.com.*
162. **Pitrat, Michel**. INRA, Domaine St. Maurice, BP 94, 84113 Montfavet cedex, France. Ph: (33) 90 31 63 30; Fax: (33) 90 31 63 98; Email: *Michel.Pitrat@avignon.inra.fr* Melon, disease resistance, mutants, genetic map.
163. **Pootstchi, Iraj** 97 St. Marks Road, Henley-on-Thames RG9 1LP, England. Ph: (0149) 574959; Fax: (10491) 574500. Breeding cantaloupes, melons and watermelons.
164. **Poulos, Jean M.** Asgrow Italia, Veg. Seeds Srl, Pontinia Research Station, C.P. 110-04014 Pontinia, Italy. Ph: 39(0)773 848549; Fax: 39(0)773 848548; Email: *jpo8los@svseeds.nl.*
165. **Price, E. Glen**. Sugar Creek Seed., Inc. P.O Box 508, Hinaton, OK 73047. Ph: (405) 542-3920; Fax: (405) 542-3921; Email: *SGRCRKSD@hintonet.net.* Seedless watermelon, polyploidy, genetics, breeding, cytogenetics.
166. **Provvidenti, Rosario.** Cornell Univ., Dept. Plant Pathology, NY State Agric. Experiment Sta., Geneva, NY, 14456-0462. Ph: (315) 787-2316, Fax: (315) 787-2389; Email: *rp13@cornell.edu.* Breeding & genetics of resistance to viral diseases of cucumber, squash, melon, watermelon & other cucurbits.
167. **Quisumbing, Alberto R. (Bert).** Delaware Valley Coll., Hort. Dept., 700 E. Butler Ave., doylestown,PA 18901-2404. Ph:333; Fax: (215) 489-2404; Email: *quisumbb@devalcol.edu.* IPM in cucumbers, melons and watermelons; host plant resistance to insects; marketing.
168. **Ramirez,, Pilar.** CIBM, Universidad de Costa Rica, San Jose, Costa Roca. Ph: 506-253-5661 x 3496; Fax: 506-224-6749; Email: *pramirez@cariari.ucr.ac.cr.* Viruses in cucurbits, production of resistant transgenic plants.
169. **Ray, Dennis**. Department of Plant Sciences, University of Arizona, Tucson, AZ 85721. Ph: (520) 621-7612; Fax: (520) 621-7186; E-mail: *dtray@u.arizona.edu.* Genetics and cytogenetics of *Cucumis melo* and *Citrullus* spp.
170. **Reiten, Joel.** Territorial Seed Co., P.O. Box 157, Cottage Grove, OR 97424. Ph: (541 942-9547; Fax: (541) 942-9881; Email: *tsc@ordata.com.* Bacterial wilt resistance, as well as virus resistance obtained through traditional breeding methods.
171. **Reuling, Gerhard T.M.** Nunhens Zaden B.V., P.O. Box 4005, 6080 AA Haelen, The Netherlands. Ph: 0475-599222; Fax: 0475-599223; Email: *bre@nunheims.nl.* Cucumber breeding.
172. **Rhodes, Bill B.** Clemson Univ./Horticulture, Poole Agricultural Center, Clemson, SC 29634-0375. Ph: (864) 656-0410; Fax: (803) 656-4960; Email: *BRhodes@clemson.edu.* Watermelon genetics, breeding, micropropagation, disease resistance, male sterility, triploids.
173. **Robinson, R. W.** Dept. Hort. Sci., New York State AES, Hedrick Hall, Geneva, NY 14456-0462 . Ph: (315) 787-2237; Fax: (315) 787-2397; Email: *rwr1@cornell.edu.* Breeding and genetics of cucurbits.
174. **Robledo, Claude.** Seminis-Recherch France, Mas de Rouzel-Chemin des Canaux, 30900 Nimes, France. Ph: 33 (0)4.66.38.79.80; Fax: 33(0)4.66.38.79.81. Melon breeding.
175. **Roig, Luis A.** Departamental Biotechnology, ETS. Ingen. Politec., Camino de Vera 14, 46022-Valencia, Spain. Ph: 34(6) 3877424; Fax: 34(6) 3877429.
176. **Saito, Takeo.** National Research Institute, Veg., Orn. Plants & Tea, Ano, Mie 514-2392, Japan. Ph: +81-59-268-1331; Fax: +81-59-268-1339; Email: *romario@nivot.affrc.go.jp.* Breeding melons resistant to diseases and insects; use of DNA markers for melon breeding.
177. **Sanghani, Amul.** Unicorn Agrotech, Ltd. 1-7-139/3, S.D. Road, Hyerabad, A.p., India 500 003. Ph: +91 40 7811554; Fax: +91 40 7842399; Email: *uniagro@hd1.vsnl.net.in.*
178. **Sarfatti, Matti.** Hazera Ltd., Research Dept., Mivhor, M.P., Lakhish Daram 79354, Israel.
179. **Schroeder, Robert Harold** Harris Moran Seed Co., 9241 Mace Blvd., Davis, CA 95616. Ph: (530) 756-1382; Fax: (530) 756-1016. Incorporating disease resistance into useful commercial cultivars.
180. **Schultheis, Jonathan R.** Dept. Horticulture, 264 Kilgore Hall, North Carolina St. University, Raleigh, NC 27695-7609. Ph: (919) 515-3131; Fax: (919) 515-7747; Email: *jonathan_schultheis@ncsu.edu.* Cultural management of cucurbits; plant spacing, establishment, nutrition, pollination & cultivar evaluation.
181. **Shetty, Nischit. V.** Asgrow Vegetable Seeds, 432 TyTy Omega Road, Tifton, GA 31794.
182. **Shiffris, Oved.** 21 Walter Avenue, Highland Park, NJ 08904. Precocious pigmentation in *Cucurbita.*
183. **Simon, Philipp. W.** USDA/ARS-Veg Crops, Dept. Hort., Univ. Wisconsin, 1575 Linden Dr., Madison, WI 53706. Ph: (608) 262-1269; Fax: (608) 262-4743; Email: *psimon@facstaff.wisc.edu.* Breeding and genetics.
184. **Sipeyre, Bruno.** Mas de Rouzel, Chemin des Canaux, 30900 Nimes, France. Ph:: 66.84.21.32; Fax: 66.38.09.42.
185. **Skirvin, Robert M.** Univ. Illinois, Dept. Horticulture, 258 PABL, 1201 Gregory Dr., Urbana, IL 6801. Ph: (217) 333-1530; Fax: (217) 333-4777; Email:*skirvin@uxl.cso.uiuc.edu.* Micropropagation; somaclonal variation.

186. **Snyder, James W.** 1231 Kirkwood Drive, Vineland, NJ 08360. Ph: (609) 794-3880; Fax: (609) 794-3881.
187. **Staub, Jack E.** USDA, ARS, Dept. Horticulture, Univ. Wisconsin, Madison, WI 53706-1590. Ph: (608) 262-0028; Fax: (608) 262-4743; Email: *jestaub@facstaff.wisc.edu.* Cucumber breeding & genetics, physiology, biochemical genetic markers, evolution, environmental stress.
188. **Stephenson, Andrew G.** 208 Mueller Lab, Penn State University, University Park, PA, 16802. Ph: (814) 863-1553; Fax: (814) 865-9131; Email: *as4@psu.edu.*
189. **Stevens, M. Allen**. Seminis Vegetable Seeds, Inc., 37437 State Highway 16, Woodland, CA, 95695.Ph: (530) 666-0931; Fax: (530) 668-0219. Direction of research.
190. **Stravato, Vittorio Mario.** via Carlo Levin. 18, 04022 Fondi - Latina, Italy. Ph: 771-510729; Email: *sumas@tiscalinet.it.* Disease resistance in Cucurbitaceae species.
191. **Summers, William L.** Iowa State University, Dept. Horticulture, Rm. 251, Ames, IA 50011-1100. Ph: (515)-294-1978; Fax: (515) 294-0730. Email: *summers@iastate.edu.* Genetic improvement of watermelon.
192. **Susic, Zoran.** Inst. "Srbija" - Ctr Vegetable Crops, Karadjorjeva St. 71, 11420 Smederevska Palanka, F.R. Yugoslavia. Ph: +381-26-323-170; Fax: +381-26-323-785; Email: *cfvsp@eunet.yu.* Genetics and breeding of Cucurbita species; cucumber breeding.
193. **Tatlioglu, Turan.** Institut of Applied Genetics, Univ. Hannover, Herrenhauser Str. 2, 3000 Hannover, Germany. Ph: (+49)511762-5675; Fax: (+49) 511762-3608; Email: *turan.tatlioglu@mbox.genetik.uni-hannover.de.*
194. **Taurick, Gary.** Harris Moran Seed Co., P.O. Box 392, Sun Prairie, WI 53590. Ph: (608) 837-6574; Fax: (608) 837-3758. Development of commercial hybrids of pickle, slicer and Beit Alpha cucumbers,
195. **Teppner, Herwig.** Inst. Botany, Karl-Franzens Univ., Holteigasse 6, A-8010 Graz, Austria. Ph: 316-380-5656; Fax: 316-380-9883; Email: *herwig.teppner@kfunigraz.ac.at.* Systemastics, morphology, ecology, crops & medicinal plants (teaching) and small scale breeding.
196. **Thomas, Claude E.** USDA-ARS, U.S. Vegetable Laboratory, 2875 Savannah Highway, Charleston, SC 29407. Ph: (803) 556-0840; Fax: (843) 763-7013; Email: *cthomas@awod.com.* Disease resistance in cucurbits.
197. **Thompson, Gary.** Dept. Plant Sciences, 303 Forbes Bldg., Univ. Arizona, Tucson, AZ 85721. Ph: (520) 621-9735; Fax: (520) 621-7816; Email: *garyt@u.arizona.edu.*
198. **Tolla, Greg.** Asgrow SVS, 432 TyTy Omega Rd., Tifton, GA 31794. Ph: (912) 386-8701; Fax: (912) 386-8805. Cucumber breeding and genetics.
199. **Tsaftaris, A.S.** Dept. Genetics & Breeding of Plants, Aristotelian Univ. of Thessaloniki, Thessaloniki, 54006, Greece.
200. **Vakalounakis, Demetrios J.** Plant Protection Inst. N.A.R.F., P.O. Box 1802, 711 10 Heraklio, Crete, Greece. Ph: +3081-240.986; Fax: +3081-245.858; Email: *vakaloun@nefeli.imbb.forth.gr*.
201. **Van Eijk, Manual.** Linda Vista Farm. East West Seed Co., P.O. Box 9073, 3006 Baliuag,Philippines. Ph: 63(0)44 7641370; Fax: 63 (0)44 7641250; Email: *research.ph@eastwestseed.com* Breeding of bitter gourd, squash, cucumber, melon, watermelon, sponge gourd, and bottle gourd.
202. **van Kooten, Henk. C.** Seminis Veg Seeds - Bruinsma, Wageningse Afweg 31, 6702 PD Wageningen, The Netherlands. Email: *hvkooten@svseeds.nl.* Breeding pickling cucumber.
203. **Vardi, Eyal.** Hazera Quality Seeds. Mivhor Farm, M.P. Lachish Daron 79354, Israel. Ph: +972-7-6813228; Fax: +972-7-6814057; Email: *vardi@hazera.com.*
204. **Walters, Terrence**. The Cucurbit Network, 11901 Old Cutler Road, Miami, FL 33156-4242. Ph: (305) 667-3800; Fax: (305) 661-5984; Email: *walters@servax.fiu.edu.* Communication via The Cucurbit Network; the whole family Cucurbitaceae.
205. **Wang, Gang.** #84 Orange Street, Woodbriudge, NJ 07095. Email: *w2140@hotmail.com.* Watermelon and melon breeding.
206. **Wang, Ming.** Department of Horticulture, Northwestern Agricultural University, Yangling, Shaanxi 712100, P.R. China. Ph: (0910) 709-3426; Fax: (0910) 701-2559. Watermelon genetics and breeding.
207. **Warid, Warid A.** 11 Cairo University Street, Apartment #4, Giza - 12211, Egypt. Breeding of cucurbits.
208. **Wasilwa, Lusike.** Rutgers Blueberry/Cranberry Res Ctr, 125A Lake OswegoRd., Chatsworth, NJ 08019. Ph: (609) 726-1590; Fax: (609) 726-1593; Email: *wasilwa@aesop.rutgers.edu.* Disease screening, fungal genetics, evaluation of fungal diversity of *Colletoirtrichum* spp.
209. **Watterson, Jon.** Seminis Vegetable Seeds, Inc., 37437 Highway 16, Woodland, CA 95616. Ph: (530) 669-6157; Fax: (530) 666-6791. Cucumber, melon, watermelon, squash, pumpkin, gourd germplasm and disease resistance.
210. **Wehner, Todd. C.** Dept. Horticultural Science, Box 7609, North Carolina St. Univ., Raleigh, NC 27695-7609. Ph: (919) 515-5363; Fax: 919-515-2505; E-mail: *todd_wehner@ncsu.edu.* Pickling/slicing cucumber, watermelon, luffa gourd; selection, disease resistance, yield, genetics & chilling.
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212. **Wessel-Beaver, Linda** Department of Agronomy & Soils Dept., Univ. Puerto Rico, P.O.Box 5000, Mayaguez, PR,

00681-5000. Ph: (809) 832-4040; Fax: (809) 265-0220; E-mail: *l_beaver@rumac.upr.clu.edu*, Pumpkin & squash breeding; disease resistance; insect resistance.

213. **Wiebe, Wayne.** Seminis Vegetable Seeds, Inc., 37437 State Highway 16, Woodland, CA 95695. Ph: (530) 666-0931; Fax: (530) 668-0219. Cucurbit diseases and disease resistance.
214. **Williams, Tom V.** Novartis Seeds, 10290 Greenway Road, Naples, FL 34114-3199. Ph: (941) 775-4090; Fax: (941) 774-6852; email: *tom.williams@seeds.novartis.com.*. Watermelon breeding.
215. **Winkler, Johanna.** Saatzucht Gleisdorf GesmbH, A-8200 Gleisdorf, Am Tieberhof 33, Austria. Ph: +43 (0) 3112 21050; Fax: +43 (0)3112 21050; email: *winkj.szgl@ccf.co.at.*
216. **Wolff, David W.** Wakata Seed America, Inc., P.O. Box 1118, Lehigh Acres, FL 33970-1118. Ph: (941) 369-0032 X 13; Fax: (941) 369-7528; Email: *Dwolff@sakata.com.* Watermelon breeding and genetics, molecular markers.
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218. **Wu, Wendy Y.** Known-You Seed Co., Ltd., 330, Kao Tan village, Jen Wu Hsing, Kaohsiung, 814, Taiwan, R.O.C., Ph: 886-7-3719725; Fax: 886-7-3718510. Breeding and growing cucurbits (all).
219. **Yamanaka, Hisako** Yamato-Noen Co., Ltd. 110, Byodobo-cho, Tenri-City Nara, Japan 632-0077. Ph: 07436-2-1182; Fax: 07436-3-3445.
220. **Yorty, Paul**. Qualiveg Seed Production, 3033 E. , 3400 N. Twin Falls, ID 83301. Ph: (208) 733-0077; Fax: (208) 733-0077. Cucurbit breeding.
221. **Zhang, Jiannong.** Melon Research Institute, Gansu University of Agriculture, Lanzhou, Gansu, 730070, P.R. China.
222. **Zhang, Xingping.** Novartis Seeds, Inc. Vegetables, 21435 Rd 98, Woodland, Ca 95695. Ph: (530) 666-0986; Fax: (530) 666-5273; Email: *Xingping.Zhang@seeds.Novartis.com.* Watermelon and melon genetics & breeding.
223. **Zitter, Thomas A.** Cornell Univ., Dept. Plant Pathology, 334 Plant Science Building, Ithica, NY 14853-5908. Ph: (607) 255-7857, Fax: (697) 255-4471; Fax: (607) 255-4471; E-mail: *tax1@cornell.edu.* Fungal and viral disease resistance.

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  - Gary Elmstrom
  - C.Ed Green
  - Phyllis Himmel
  - Hasib Humaydan
  - Aly M.. Ibrahim
  - Jiping Jiang
  - Joseph J. King
  - Stephen R. King
  - Larry D. Knerr
  - Krystina Markiewicz-Ladd
  - J.D. McCreight
  - Brian Joseph Moraghan
  - Michael Nadel
  - Larry Nea
  - Wei Ouyang
  - Ken Owens
  - Paul S. Peterson
  - Robert H. Schroeder
  - M. Allen Stevens
  - Jon Watterson
  - Wayne Wiebe
  - Xingping Zhang
- **Colorado**
  - Larry A. Hollar
- **Florida**
  - R. Bruce Carle
  - Kevin L. Cook
  - Deena Decker-Walters
  - Mark Hutton
  - Donald N. Maynard
  - Mike Meadows
  - Tom V. Williams
  - David W. Wolff
- **Georgia**
  - George E. Boyhan
  - David Groff

- Nischit V. Shetty
- Greg Tolla

- **Iowa**
  - Glenn Drowns
  - Laura C. Merrick
  - William L. Summers
- **Idaho**
  - Steven Loyd Love
  - Paul Yorty
- **Illinois**
  - Richard Hentschel
  - Robert M. Skirvin
- **Indiana**
  - Orie J. Eigsti
  - Ray D. Martyn
- **Maryland**
  - Joseph H. Kirkbride, Jr.
  - Charles A. McClurg
  - Timothy J. Ng
- **Maine**
  - Rob Johnston, Jr.
- **Michigan**
  - Rebecca Grumet
- **Missouri**
  - Alejandro (Alex) Ching
- **North Carolina**
  - Phil Denlinger
  - Jonathan R. Schultheis
  - Todd C. Wehner
- **Nebraska**
  - Dermot P. Coyne
- **New Hampshire**
  - J. Brent Loy
- **New Jersey**
  - Oved Shifriss
  - James W. Snyder
  - Gang Wang
  - Lusike Wasilwa
- **New Mexico**
  - Chris Cramer
- **New York**
  - Thomas C. Andres
  - Molly Kyle Jahn
  - H.M. Munger
  - Rosario Provvidenti
  - R.W. Robinson
  - Thomas A. Zitter
- **Oklahoma**
  - Bemmy Bruton
  - Angela Davis
  - E. Glen Price
- **Oregon**
  - Rebecca Brown
  - Louis Victor DiNitto
  - August C. Gabert
  - Joel Reiten
- **Pennsylvania**

- Alberto R. (Bert) Quisumbing
- Andrew G. Stephenson

- **Puerto Rico**
  - Linda Wessel-Beaver
- **South Carolina**
  - Amnon Levi
  - Bill B. Rhodes
  - Claude E. Thomas
- **Texas**
  - Robert Coffey
  - Joseph O. Kuti
- **Virginia**
  - Greg Welbaum
- **Washington**
  - John P. Navazio
- **Wisconsin**
  - Michael J. Havey
  - Richard L. Lower
  - Philipp W. Simon
  - Jack E. Staub
  - Gary Taurick

Cucurbit Genetics Cooperative Report 23:150-150 (article 50) 2000

International CGC Members

- **Argentina**
  - Fernando Lopez Anido
- **Australia**
  - Douglas A. Blazey
  - Desmond John McGrath
- **Austria**
  - Tamas Lelley
  - Herwig Teppner
  - Jojhanna Winkler
- **Brazil**
  - Paulo T. Della Vecchia
  - Wilson Roberto Maluf
  - Waldelice Oliveira de Paiva
- **China, P.R.**
  - HaiQing Bao
  - Hongwen Cui
  - Depei Lin
  - Wenge Liu
  - Ming Wang
  - Mingzhu Wu
- **Costa Rica**
  - Pilar Ramirez
- **Czech Republic**
  - Bohuslav Holman
  - Eva Kristkova
  - Alex Lebeda
- **Denmark**
  - Hans Henrik Kampmann
- **Egypt**
  - Ahmed Abdel-Moneim Hassan
  - Warid A. Warid
- **England**
  - Iraj Poostchi
- **France**
  - Sylvie Baudracco-Arnas
  - Frank De Langen
  - C. Dogimont
  - Graines Gautier
  - Frederic Ignart
  - Michel Lecouviour
  - Florence Picard
  - Michael Pitrat
  - Claude Robledo
  - Bruno Sipeyre
- **Germany**
  - Hubert Kuhlmann
  - Turan Tatlioglu
- **Greece**
  - A.S. Tsaftaris
  - Demetrios J. Vakalounakis

Guadeloupe (F.W.I.)
- Nathalie Boissot

- **India**
  - Major Singh Dhaliwal
  - A. Ganapathi
  - Satish C. gupta
  - Jaagrati Jain
  - Chandra Pathak
  - K.V. Peter
  - Amul Sanghani
- **Indonesia**
  - Doretta Akkermans
- **Israel**
  - Ron Cohen
  - Yigal Cohen
  - Yael Danin-Poleg
  - Victor Gaba
  - Davidi Haim
  - Ran Herman
  - Zvi Karchi
  - Nurit Katzir
  - Harry Paris
  - Rafael Perl-Treves
  - Matti Sarfatti
  - Eyal Vardi
- **Italy**
  - Paoli Crino
  - Erik de Groot
  - Nadia Ficcadent
  - Gianni Gatto
  - Jean M. Poulos
  - Vittorio Mario Stravato
- **Japan**
  - Hiroshi Ezura
  - Hisashi Funakushi
  - Toshitsugu Hagihara
  - Tetsuo Hirabayashi
  - Takayuki Ikegami
  - Kimio Ito
  - Minru Kanda
  - Kenji Kato
  - Sugiyama Keita
  - Yoshihiro Konno
  - Yasuhisa Kuginuki
  - Seiji Matsurra
  - Tatsuya Mochizuki
  - Takeo Saito
  - Hisako Yamanaka
- **Kenya**
  - Edward E. Carey
  - Torbjorn Kerje
- **Korea, Rep. of**
  - Chang-Soon Ahn
  - Sang Joo Han
  - Soo Nyeon Kwack
  - Cheol-Sang Kwon
  - Young-Seok Kwon

- Young-Hyun Om
- **Mexico**
  - Sergio Garza Ortega
  - Roberto Compean Melendez
- **Namibia**
  - Gillian Maggs
- **Peru**
  - Miguel Holle
- **Philippines**
  - Manuel van Eijk
- **Poland**
  - Katarzyna Niemirowicz-Szczytt
- **Spain**
  - Pilar Corella
  - Laia Fito
  - M. Luisa Gomez-Guillamon
  - Peter Kraakman
  - Fernando Nuez
  - Gloria Palomares
  - Jose Luis Peiro Abril
  - Luis A. Roig
- **Sudan**
  - Ali Elamin El Jack
  - El Tahir Ibrahim Mohamed
  - Yousif Fadlalla Mohamed
  - Sadig Khdir Omara
- **Sweden**
  - Louis Carl Lehmann
- **Taiwan, R.O.C.**
  - Fure-Chyi Chen
  - Wendy Y. Wu
- **Thailand**
  - Usa Duangsong
  - Suphot Iamsangsri
  - Jolanda Kouters
  - Sompong Milerue
- **The Netherlands**
  - P.A. Boorsma
  - Monique Bosma
  - A.C. de Ruiter
  - K. Hertogh
  - Ad Klapwijk
  - Jeroen Nannes
  - Gerhard T.M. Reuling
  - Henk C. van Kooten
- **Turkey**
  - Gulat Caglar
  - J.P. Mazereeuw
- **Yugoslavia**
  - Janos Berenji
  - Zoran Susic

Cucurbit Genetics Cooperative Report 23:151-153 (article 51) 2000

Covenant and By-Laws of the Cucurbit Genetics Cooperative

Article I. Organization and Purposes

The Cucurbit Genetics Cooperative is an informal, unincorporated scientific society (hereinafter designated "CGC") organized without capital stock and intended not for business or profit but for the advancement of science and education in the field of genetics of cucurbits (Family: Cucurbitaceae). Its purposes include the following: to serve as a clearing house for scientists of the world interested in the genetics and breeding of cucurbits, to serve as a medium of exchange for information and materials of mutual interest, to assist in the publication of studies in the aforementioned field, and to accept and administer funds for the purposes indicated.

Article II. Membership and Dues

The membership of the CGC shall consist solely of active members; an active member is defined as any person who is actively interested in genetics and breeding of cucurbits and who pays biennial dues. Memberships are arranged by correspondence with the Chairman of the Coordination Committee.

The amount of biennial dues shall be proposed by the Coordinating Committee and fixed, subject to approval at the Annual Meeting of the CGC. The amount of biennial dues shall remain constant until such time that the Coordinating Committee estimates that a change is necessary in order to compensate for a fund balance deemed excessive or inadequate to meet costs of the CGC.

Members who fail to pay their current biennial dues within the first six months of the biennium are dropped from active membership. Such members may be reinstated upon payment of the respective dues.

Article III. Committees

1. The Coordinating committee shall govern policies and activities of the CGC. It shall consist of six members elected in order to represent areas of interest and importance in the field. The Coordinating Committee shall select its Chairman, who shall serve as spokesman of the CGC, as well as its Secretary and Treasurer.

2. The Gene List Committee, consisting of five members, shall be responsible for formulating rules regulating the naming and symbolizing of genes, chromosomal alterations, or other hereditary modifications of the cucurbits. It shall record all newly reported mutations and periodically report lists of them in the Report of the CGC. It shall keep a record of all information pertaining to cucurbit linkages and periodically issue revised linkage maps in the Report of the CGC. Each committee member shall be responsible for genes and linkages of one of the following groups: cucumber, *Cucurbita* spp., muskmelon, watermelon, and other genera and species.

3. Other committees may be selected by the Coordinating Committee as the need or fulfilling other functions arises.

Article IV. Election and Appointment of Committees

1. The Chairman will serve an indefinite term while other members of the Coordinating Committee shall be elected for ten-year terms, replacement of a single retiring member taking place every other year. Election of a new member shall take place as follows: A Nominating Committee of three members shall be appointed by the

Coordinating Committee. The aforesaid Nominating Committee shall nominate candidates for an anticipated opening on the Coordinating Committee, the number of nominees being at their discretion. The nominations shall be announced and election held by open ballot at the Annual Meeting of the CGC. The nominee receiving the highest number of votes shall be declared elected. The newly elected member shall take office immediately.

In the event of death or retirement of a member of the Coordinating Committee before the expiration of his/her term, he/she shall be replaced by an appointee of the Coordinating Committee.

Members of other committees shall be appointed by the Coordinating Committee.

Article V. Publications

1. One of the primary functions of the CGC shall be to issue an Annual Report each year. The Annual Report shall contain sections in which research results and information concerning the exchange of stocks can be published. It shall also contain the annual financial statement. Revised membership lists and other useful information shall be issued periodically. The Editor shall be appointed by the Coordinating Committee and shall retain office for as many years as the Coordinating Committee deems appropriate.

2. Payment of biennial dues shall entitle each member to a copy of the Annual Report, newsletters, and any other duplicated information intended for distribution to the membership. The aforementioned publications shall not be sent to members who are in arrears in the payment of dues. Back numbers of the Annual Report, available indefinitely, shall be sold to active members at a rate determined by the Coordinating Committee.

Article VI. Meetings

An Annual Meeting shall be held at such a time and place as determined by the Coordinating Committee. Members shall be notified of time and place of meetings by notices in the Annual Report or by notices mailed not less than one month prior to the meeting. A financial report and information on enrollment of members shall be presented at the Annual Meeting. Other business of the Annual Meeting may include topics may include topics of agenda selected by the Coordinating Committee or any items that members may wish to present.

Article VII. Fiscal Year

The fiscal year of the CGC shall end on December 31.

Article VIII. Amendments

These By-Laws may be amended by simple majority of members voting by mail ballot, provided a copy of the proposed amendments has been mailed to all the active members of the CGC at least one month previous to the balloting deadline.

Article IX. General Prohibitions

Notwithstanding any provision of the By-Laws or any other document that might be susceptible to a contrary interpretation:

1. The CGC shall be organized and operated exclusively for scientific and educational purposes.
2. No part of the net earnings of the CGC shall or may under any circumstances inure to the benefit of any individual.
3. No part of the activities of the CGC shall consist of carrying on propaganda or otherwise attempting to influence legislation of any political unit.
4. The CGC shall not participate in, or intervene in (including the publishing or distribution of statements),

any political campaign on behalf of a candidate for public office.

5. The CGC shall not be organized or operated for profit.
6. The CGC shall not:

(a) lend any part of its income or corpus without the receipt of adequate security and a reasonable rate of interest to;
(b) pay any compensation in excess of a reasonable allowance for salaries or other compensation for personal services rendered to;
(c) make any part of its services available on a preferential basis to;
(d) make any purchase of securities or any other property, for more than adequate consideration in money's worth; or
(e) sell any securities or other property for less than adequate consideration in money or money's worth; or
(f) engage in any other transactions which result in substantial diversion of income or corpus to any officer, member of the Coordinating Committee, or substantial contributor to the CGC.

The prohibitions contained in this subsection (6) do not mean to imply that the CGC may make such loans, payments, sales, or purchases to anyone else, unless authority be given or implied by other provisions of the By-Laws.

Article X. Distribution on Dissolution

Upon dissolution of the CGC, the Coordinating Committee shall distribute the assets and accrued income to one or more scientific organizations as determined by the Committee, but which organization or organizations shall meet the limitations prescribed in sections 1-6 of Article IX.

Cucurbit Genetics Cooperative Report 23:154-154 (article 52) 2000

Cucurbit Genetics Cooperative

Financial Statement

31 December 1999

Balance (31 December 1998)		**$3,679.78**
Receipts		
Dues and CGC back issue orders	$2,488.00	
Interest on savings	$63.84	
Total receipts		**$2,551.84**
Expenditures		
CGC Report No. 22 (1999)		
Printing	$1,929.99	
Mailing	$613.72	
Call for papers (Report No. 23)	$165.46	
Member/Subscriber Renewal Notices	$71.80	
Miscellaneous (envelopes, postage, etc.)	$42.00	
Bank fees and adjustment charges	$11.00	
Total Expenditures		**$2,833.27**
Balance (31 December 1999)		**$3,398.35**