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Race Specificity and Methods of Study

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I. Introduction

As pointed out in the previous chapter, within most species of the cereal rust fungi, there are a number of *formae speciales.* These *formae speciales* in turn are composed of many biotypes that differ in several characteristics but primarily in their virulence on host cultivars. A biotype is defined as a population of individuals of the same genotype; thus, theoretically, the progeny of an aeciospore or urediospore would constitute a pathogen biotype. However, current technology permits only the identification of the pathogen phenotype expressed by a limited number of genes. Thus the biotype remains a useful concept but often has been incorrectly applied in practice to different cultures of the same race because they had a similar avirulence/virulence phenotype.

The avirulence/virulence pattern of a culture is determined by inoculating a selected group of host plants of differing genotypes for rust resistance. A group of biotypes with a similar avirulence/virulence pattern on a selected group of host plants is considered a physiologic race. The race thus is a taxon below the *forma specialis* level, which is distinguished by physiologic differences (pathogenic differences in host–pathogen interactions) rather than morphologic differences. The physiologic differences are shown as differing avirulence/virulence patterns when the differential host series is independently inoculated with different cultures. Thus a race could be a single biotype but is more likely to be a group of similar biotypes that can be distinguished from other phenotypes with a reasonable amount of effort and certainty by differences in their virulence patterns on a selected differential series. The avirulence/virulence phenotype is determined from the disease infection types. Therefore, even though two cultures of a single race result in the production of the same phenotypes (avirulence/virulence pattern), they may not be the same biotype (genotype) even for pathogenicity, as the infection type may be due to either a homozygous or heterozygous pathogen genotype for virulence.

A. DIFFERENTIAL SERIES

A selected group of host lines has been designated a differential host series or set for many cereal rusts. Thus the 12 host cultivars chosen for the race differential series for wheat stem rust (Stakman *et al.*, 1962) have become known as the international, standard, or sometimes as the Stakman differentials. Other differential sets were used by other workers, which necessitates the designation of the differential set used. In some cases, additional differential hosts were included with the international differential series, and they often became known as supplemental differentials. Unfortunately, sometimes this resulted in physiologic races so described, to be considered as subraces (based on less important differences) rather than as subdivisions of a standard race (that were as important as the original divisions) (Stakman *et al.*, 1962) as intended.

B. INFECTION TYPES

The use of the infection type as a measurement of disease had been developed and was used by Stakman and co-workers at Minnesota by 1919 (Hoerner, 1919). The characterizations of the infection types have been described in slightly different ways during the past 65 years. However, those developed by Stakman and co-workers for the wheat stem rust system have been adapted to most of the cereal rusts. A major exception is stripe rust, *P. striiformis*, which results in a systemic infection. The modification of the original system currently in use at the Cereal Rust Laboratory is shown in Table I. Two variations of the mesothetic reaction class (X infection type) have been recognized since 1919. The Y infection type was added for wheat leaf rust by Johnston (1963) and the Z infection type for common corn rust by Van Dyke and Hooker (1969). These infection types have also been observed with *Puccinia graminis* Pers. f. sp. *tritici* on wheat and barley.

Other systems of classifying infection types have been developed but offer little advantage for use in most race specificity studies. Under carefully controlled environmental conditions and inoculum densities, the system of Browder and Young (1975), which considers the size of the sporulating area and lesion independently on a 0 to 9 basis, should be considered. This system is very advantageous in genetic studies, but its precision often is superfluous for race surveys with host lines possessing a "single gene" for disease resistance. Generally the distinction

Table I

Description of Infection Types Used in Physiologic Specialization Studies of the Cereal Rusts at the Cereal Rust Laboratory[a]

Host response (class)[b]	Disease: Infection type[c]		Disease: Symptoms
Immune (Res)	0	low	No uredia or other macroscopic sign of infection
Nearly immune (Res)	;	low	No uredia, but hypersensitive necrotic or chlorotic flecks of varying size present
Very resistant (Res)	1	low	Small uredia surrounded by a necrosis
Moderately resistant (Res)	2	low	Small to medium uredia often surrounded by chlorosis or necrosis; green island may be surrounded by chlorotic or necrotic border
Heterogeneous (Mes)	X	low	Random distribution of variable-sized uredia on single leaf with a pure culture
Heterogeneous (Mes)	Y	low	Ordered distribution of variable-sized uredia, with larger uredia at leaf tip
Heterogeneous (Mes)	Z	low	Ordered distribution of variable-sized uredia, with larger uredia at leaf base
Moderately susceptible (Sus)	3	low	Medium-sized uredia that may be associated with chlorosis or rarely necrosis
Susceptible (Sus)	4	high	Large uredia without chlorosis or necrosis

[a] After Roelfs and McVey (1979); Stakman *et al.* (1962).

[b] Res, Resistant; Mes, mesothetic; Sus, susceptible.

[c] The infection types are often refined by modifying characters as follows: =, uredia at the lower size limit for the infection type; −, uredia somewhat smaller than normal for the infection type; +, uredia somewhat larger than normal for the infection type; ++, uredia at the upper size limit for the infection type; C, more chlorosis than normal for the infection type; and N, more necrosis than normal for the infection type. Discrete infection types on a single leaf when infected with a single biotype are separated by a comma (e.g., 4,; or 2=,2+ or 1,3C). A range of variation between infection types is recorded by indicating the range, with the most prevalent infection type listed first (e.g., 23 or ;1C or 31N).

needed in race surveys is between the high-infection types (e.g., between pathogen genotype *P_* and host *hh* and pathogen type *pp* and hosts *HH* and *hh*) and the low-infection types (e.g., between pathogen genotype *P_* and host *HH*). This distinction is adequate for a host–pathogen system that follows a gene-for-gene relationship. Although some aspects for gene-for-gene theory in relation to race identification are discussed in Section IV, the full impact of gene-for-gene theory on cereal rust studies is beyond the scope of this chapter. However, the gene-for-gene theory proposed by Flor has resulted in a better understanding of the significance of infection types (see Loegering, Chapter 6, this volume). A major change has been that if the high-infection type

is 4 (e.g., pathogen genotype *pp* and host *hh*), then any lower infection type indicates a level of resistance.

II. Why Study Race Specificity?

This question is answered differently depending on the interest of the respondent. Thus race surveys may vary in operation depending on their established goals. The race concept has been important in enabling the development of useful resistance to the small-grain cereal rusts in North America and Australia. Apparently, the race concept is the most useful in asexually reproducing pathogens that are obligate parasites or function only as parasites. The usefulness of a race concept decreases as frequency of virulence changes increases as a result of sexual or parasexual recombinations, or mutation. Thus experience has shown the race concept is most useful in *P. graminis* and the least so for *P. striiformis* among the wheat rusts.

A. DETECTION OF NEW VIRULENCES

Originally, most race surveys were designed to detect new virulent pathogen phenotypes before they increased to economically important levels (Simons and Michel, 1959). Cultures that resulted in a susceptible host response when previously cultures had produced a mesothetic or resistant host response were considered to be potentially threatening. Such cultures were therefore used to test commercial cultivars and breeding lines. The detection of new virulent phenotypes remains a major goal of most race surveys. However, by using host differential lines with a "single gene" for resistance, it is now possible to detect changes in virulence on a gene for resistance and know if that results in a virulence combination that is capable of overcoming the combinations of resistance in commercially grown cultivars or advanced breeding material. Most new combinations of virulence that we have detected are avirulent on the commercial cultivars and have little economic potential for causing crop losses.

B. SOURCE OF CULTURES

Race surveys historically have been the source of cultures used in testing host lines in genetic and plant breeding studies. The advances made in culture storage techniques (Rowell, Chapter 10, this volume)

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B. SOURCE OF CULTURES

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greatly improved the precision of these studies by making it possible to use the same culture over a period of years without the risk of loss or contamination. This reduced the need to obtain large numbers of cultures for this purpose annually. The race survey, however, remains the source of nearly all new combinations of pathogen virulence. With the development of "single-gene" differential host lines, it is now possible to search for particular pathogen avirulence/virulence phenotypes. These cultures are extremely useful in host genotype postulation. A postulation of the host genotype for specific resistance is possible by infecting the host with a selected group of cultures of known virulence phenotype (McVey and Roelfs, 1975). The most efficient method is to use cultures that are identical except for the pathogenicity of a single host gene pair. These differences exist in a very low frequency in asexually reproducing populations. Cultures that differ in pathogenicity only on a single host gene pair are common in a sexually reproducing population; however, it becomes necessary to use a very large number of differential hosts to avoid missing other differences in pathogenicity on other host genes for resistance that were previously unknown. These undetected host genes could result in incorrect host gene postulation.

C. DISTRIBUTION OF PATHOGEN VIRULENCES

Race surveys provide data for mapping the distribution and frequency of races, thereby providing the necessary information for selection of sources of host resistance, or in establishing a host gene deployment system. This information in the past has not always been as effectively used as was possible, but renewed interest may enhance its value. That certain races of wheat leaf rust were principally present in the same area, year after year, was reported by Chester (1946). Such patterns still exist, and generally the reasons are unclear (Roelfs, 1974). Host resistance, pathogen adaptability, environment differences, and geographic isolation may all be factors affecting the pathogen distribution patterns observed. The leaf rust resistance provided by the resistance gene (*Lr9*) was adequate in Indiana for 10 years before 1982, whereas the same cultivars were seriously rusted in the states bordering the Gulf of Mexico.

D. EPIDEMIOLOGY STUDIES

The race surveys have an important role in determining the source of disease inoculum. The rusts are obligate parasites and have no mac-

sistance exerts selection pressure on the pathogen, which can result in a shift in the virulence in the pathogen population. This has been documented in the case of oat stem rust (Stewart and Roberts, 1970). For wheat stem rust in the United States, pathogen distribution appears to be controlled more by isolation, or pathogen adaptability and aggressiveness, than by virulence. Another example of the effect of isolation is given in detail by Luig in Vol. II of this treatise.

E. PATHOGEN STABILITY

In early years (1918–1925) the races of wheat stem rust changed rapidly from year to year (Roelfs and Groth, 1980). However, currently there is, for wheat stem rust at least, an underlying stability. The degree of stability is apparent only by studying race survey data over a period of years. Green (1975) explained that most of the variation in race 15 of wheat stem rust in Canada was by single gene changes. Roelfs and Groth (1980) broadened this approach to show that in the Great Plains of the United States, clusters of races (closely related genotypes) existed that differed only by a few gene pairs for virulence/avirulence (0–2 genes) and that between these clusters there were more gene pair differences (4–10), when pathogenicity to 16 host resistance genes was studied. They suggested that the variation within the cluster was similar, but that the differences between clusters increased if the number of host resistance genes was increased to all the designated ones (approximately 40) for wheat stem rust resistance. The within-cluster differences in combinations of virulences were interpreted as evidence for single mutations from an existing genotype within the cluster. The distances between clusters were taken to indicate a lack of sexual or parasexual recombination between members of the different clusters. Examination of a sexually reproducing population showed neither clusters of genotypes nor spaces between groups of genotypes. The combinations of virulence observed closely fitted a Poisson distribution based on random gene association.

Because the wheat stem rust population in Canada and the United States is currently relatively stable, host cultivars with combinations of resistance genes that match the pathogen virulence midway between clusters should have a long period of usefulness. However, little is known about these clusters of virulence. Virulence and avirulence for a few *Sr* genes are found in almost every cluster (i.e., *Sr8*), whereas virulence or avirulence for other *Sr* genes occur only in a single cluster (i.e., *Sr9e, Sr15, Sr30*). The latter combinations of resistance may be of more value in breeding for resistance. Some clusters, such as the race

roscopic differences among cultures in the field of a given species. So the epidemiologist studying naturally occurring populations is limited to following the disease occurrence. The only suitable marker available for distinguishing between individuals of a given *forma specialis* or species is virulence. Thus in determining spore movement, the marker used is race (see Roelfs' epidemiology chapter in Vol. II).

Although sources of inoculum cannot be determined by race frequency data alone, with adequate sample size some possible sources often can be eliminated (Rowell and Roelfs, 1971) by a comparison of the races present. A new race in an area indicates (1) an input of exogenous inoculum (Luig, 1977), (2) a mutation for virulence or avirulence in an existing race (Stakman *et al.*, 1930), (3) sexual or parasexual recombinations (Newton *et al.*, 1930), or (4) the detection of a race previously below the detection threshold. These sources of variation can often be distinguished.

With exogenous inocolum the "new" race should be identical with one in the source area. A mutation for avirulence/virulence should result in a race identical in all but one characteristic with a race previously present. Sexual or parasexual recombination results in a race that varies from existing races by possessing combinations of characters of the putative parents and would likely differ from races in adjacent source areas. However, in the United States differences in virulence are of limited use for wheat stem rust, as a single race makes up 50% of the population, and one oat stem rust race currently makes up nearly 95% of the isolates annually. The use of races to distinguish between sources of inoculum generally provides negative information; that is, the inoculum could not have come from an area, but currently virulence is the only marker adequately studied to use. In recent years, a group of wheat stem rust races of pathogen genotypes that probably are from a series of single mutations (clusters) from a source culture have been studied. The race 15 "cluster" has not been found in southern Texas or Mexico (Roelfs *et al.*, 1978). However, it has been the predominant cluster in the central and northern Great Plains. Thus we have postulated that a source of overwintering inoculum exists outside of Mexico. Over several years these types of data indicate that the principal source of inoculum for the race 15 cluster is northern Texas and southern Oklahoma in most years. The inoculum of the race 29-32, 113, 11, and 151-32 clusters probably originates farther south in Texas and Mexico. The distribution of pathogen virulence can also be compared with the distribution of host genes for resistance. The effects of the alternate host(s) and wild grasses that can be hosts must be considered; however, little is known about their resistance. Host re-

15 or 113 clusters, have many members that differ by a single gene, whereas other clusters seem to be composed of only a couple of phenotypes (i.e., race 56). Clusters like race 15 and 56 have had a history of being important and resulted in epidemics in the United States and Canada (Stakman and Harrar, 1957), whereas others like race 11, race 32–151, and race 113 never have, although they have occurred during the same years and often apparently have had the necessary genes for virulence. Different genotypes of the race 15 cluster have predominated for the past 20 years in North America, and several have been able to incite at least local epidemics. Possibly certain clusters have accumulated combinations of genes for aggressiveness that remain in the asexual reproducing population, even though some changes have occurred in virulence patterns. This accumulation of genes for aggressiveness probably has been the result of many mutations and selection for over 30 years. If adaptability is a multigenically inherited character, such genes might accumulate and remain in an asexually reproducing cluster. A mutation for virulence at a locus formerly avirulent in the race 15 cluster (this cluster is well adapted) would likely result in an adapted new race, whereas the same mutation for virulence in another cluster that never had indicted an epidemic might be expected to be similar in adaptation to other races in the cluster.

Another use of the historical approach to race studies is the study of shifts in virulence frequencies. In the race 15 cluster in the 1950s a high proportion of the population was virulent on *Sr17;* however, by 1974 this had decreased to a few percent (Roelfs and McVey, 1975), and by 1980, it had increased again to 31% (Roelfs *et al.*, 1982). No reason is apparent for this shift in virulence. In other clusters during this period, virulence on *Sr17* always or never occurred.

F. DISSEMINATION OF INFORMATION

Because cultures of a *forma specialis* of the cereal rusts differ in their capacity to cause disease on cultivars of a host species, the portion of the pathogen population under discussion must be specified. If only a few host genotypes exist it is easy to describe the pathogen phenotypes as virulent or avirulent on a particular host. This can also be done by specifying the virulence/avirulence phenotype of the culture. However, when 10 or more gene pairs are involved, a long and difficult symbol is required for identification. In most of the cereal rust fungi there are relatively large numbers of pathogen phenotypes and host genotypes, making some system of coding groups of similar pathogen phenotypes necessary. These groups of phenotypes are races and are designated by

numbers, letters, or a combination of both. After the codes are used for several years they become well known and very useful; however, with the advent of new pathogen phenotypes and cultivars the system nomenclature has to be expanded. These problems and their possible solutions are discussed in Section IV.

III. History of Race Specificity

In the second decade of this century, E. C. Stakman at Minnesota started his studies in an effort to control wheat stem rust. The early research at Minnesota involved the testing of Marshall Ward's theory of bridging hosts. This involved studying changes in pathogenicity of cultures of stem rust that were serially passed from a susceptible to a moderately resistant to a highly resistant host. The bridging-host theory held that through this process virulence would be gained. In 1916, during the course of this study, differences were found in the ability of two cultures of *Puccinia graminis* f. sp. *tritici* to attack two cultivars of wheat (Stakman and Piemeisel, 1917). The two variants were initially designated as strains, then as biological forms, and finally as races. Races subsequently were identified by differences in infection types produced on a set of host cultivars differing in resistance. This was followed by a search for races in many plant pathogens. The earliest reports of races in the cereal rusts are shown in Table II. The 1920s became the decade of the race (Stakman, 1929; Stakman *et al.*, 1935).

Most previous taxonomic work had been based on morphological differences; thus it was natural to seek morphological differences between races. Levine (1928) made many measurements on spore width and length; and although differences existed, ranges overlapped between races. Levine concluded that although there was some morphological basis for distinguishing races, they were most adequately identified by their parasitic behavior. Morphological differences certainly would now be unrealistic to use with 343 races described on the international differentials. Hartley and Williams (1971) reported differences in infection structures formed by different races on an artificial medium, but this was not confirmed in our studies (A. Roelfs and L. Martell, unpublished). Burdon *et al.* (1982) have reported different isozymes present in sporelings of different races. Through these differences, evidence was gained to support the proposed evolution of wheat stem rust in Australia (see the chapter by Luig in Vol. II of this treatise). These studies may have far-reaching effects in future studies

Table II

Initial Studies of Physiologic Specialization in the Cereal Rust Diseases Caused by *Puccinia* spp.

Host	Pathogen (*Puccinia*)	Number of		Date published	Author(s)
		Host differentials	Pathogen races		
Triticum spp.	*graminis* f. sp. *tritici*	—	—	1917	Stakman and Piemeisel
Triticum spp.	*graminis* f. sp. *tritici*	12	—	1922	Stakman and Levine
Avena spp.	*coronata* f. sp. *avenae*	2	4	1919	Hoerner
Avena spp.	*graminis* f. sp. *avenae*	3	5	1923	Stakman *et al.*
Triticum spp.	*recondita* f. sp. *tritici*	7	12	1926	Mains and Jackson[a]
Secalis	*recondita* f. sp. *secalis*	1	2	1926	Mains[b]
Hordeum	*hordei*	2	2	1926	Mains
Zea	*sorghi*	3	4	1926	Mains[c]
Triticum	*striiformis*	6	4	1930	Allison and Isenbeck
Secalis	*graminis* f. sp. *secalis*	5	3	1932	Cotter and Levine[b]

[a]Earlier abstracts by Mains and Jackson (1921, 1923).
[b]Earlier abstract by Levine ane Stakman (1923), with three differential hosts.
[c]Also an abstract by Stakman and Christensen (1926).

of evolution and diversity of the cereal rusts. The isozymes represent markers that are inherited but relatively unaffected by the selective influences of host resistances. Many questions concerning origin, genetic interchange, and diversity may be answered in the future. However, at this time, isozyme markers must be considered a new technique with many possible uses. A direct association may not necessarily exist between isozymes and virulence.

Infection types are not always a perfect measurement of the host or pathogen genotype (Luig and Rajaram, 1972). Infection types are affected by temperature, light, host nutrition, humidity, infection density, and plant age. Chester (1946) reviewed much of the literature on this subject. Some of the differences in infection types were due to very large variations in experimental methods. In the case of wheat stem rust this variation can be greatly reduced by standardizing experimental conditions. Some host–pathogen interactions are very sensitive to temperature and light, but temperatures of 18° to 22°C and a 12-hour day length with 10,000 lux of fluorescent light were generally adequate even for the most sensitive interactions with wheat stem rust. It has also been noted that the host genetic background affects some *Sr* genes, and some backgrounds result in more stable and recognizable infection types than others (Roelfs and McVey, 1979).

The development of the gene-for-gene theory by Flor was gradually refined by many workers, resulting in the relationship between the host, disease, and pathogen (see Loegering, Chapter 6, Fig. 4, this volume). For the cereal rusts, the definitive phenotypes are usually low-infection types; the nondefinitive are usually high-infection types. As indicated in the figure in Chapter 6, the infection type is a property of the interaction between host and parasite (of the aegricorpus), and a low-infection type (a definitive phenotype) can be used to determine both the host resistance and pathogen virulence phenotypes. The low-infection type occurs only when the pathogen is avirulent (*PP* or *Pp*) with respect to the corresponding host gene pair (*HH* or *Hh*). With rye, the self-sterility of the host has hampered the use of homozygous host lines.

The gene-for-gene relationship of Fig. 4 in Chapter 6 of this volume can be represented in the more familiar square, Table IIIA. For the purposes of discussion, the low- and high-infection types were indicated by their numerical value. Although these infection types are not actual data, they represent general experience. The possible host–pathogen combinations are represented in Table IIIA in a gene-for-gene system involving the interaction of one host and one pathogen gene. These types of data are typical in genetic studies when crossing is done

Table III

Theoretical Scheme Showing Infection Types Resulting from Gene-for-Gene Host–Pathogen Relationships[a,b]

A

	Pathogen		
Host	*PP*	*Pp*	*pp*
HH	0	1	4
Hh	;	1+	4
hh	4	4	4

B

	Pathogen		
Host	*PP*	*Pp*	*pp*
HH	0	1	4
hh	4	4	4

C

	Pathogen			
Host	*PPQQ*	*PPqq*	*ppQQ*	*ppqq*
HHTT	0	0	2	4
HHtt	0	0	4	4
hhTT	2	4	2	4
hhtt	4	4	4	4

D

	Pathogen								
Host	*PPQQ*	*PPQq*	*PPqq*	*PpQQ*	*PpQq*	*Ppqq*	*ppQQ*	*ppQq*	*ppqq*
HHTT	0	0	0	1	1	1	2	2+	4
HHtt	0	0	0	1	1	1	4	4	4
hhTT	2	2+	4	2	2+	4	2	2+	4
hhtt	4	4	4	4	4	4	4	4	4

[a]Low-infection types are based on limited data; some variation may also occur in the high-infection types.

[b]Resistance [*H* and *T*] and avirulence [*P* and *Q*] are shown as dominant characters, which most frequently is the case. Part **A** of table shows the interaction between single host–gene pairs. **B,** as in **A,** except heterozygous gene pair in host is omitted. **C,** interaction between two host–pathogen gene pairs, heterozygous gene pairs omitted, demonstrating effect of epistasis. **D,** as in **C,** except heterozygous gene pairs in the pathogen are included, demonstrating effects of incomplete dominance.

with both the pathogen and host. In the small number of cases studied, incomplete dominance exists with both heterozygous host and pathogen genotypes (Loegering, Chapter 6, this volume). In studying pathogen races, only homozygous host genotypes are usually used, reducing the combinations as shown in Table IIIB. The race is often based on differences between the high- and low-infection types, and no distinction is normally made between the two low-, and four high-infection types.

Most commercial host cultivars possess several genes for rust resistance, and Table IIIC is a theoretical representation of a two-gene system of a gene-for-gene relationship, with the five possible heterozygous host and five heterozygous pathogen genotypes omitted. In this example, infection type 0 results when the *HHPP* host–pathogen genotype is expressed, and infection type 2 results when the *TTQQ* genotype is expressed. In the presence of both *HHPP* and *TTQQ*, the lower of the two infection types (infection type 0), is expressed. The gene pair that results in the lowest infection type is generally expressed in cereal rusts, although exceptions may occur (Loegering, Chapter 6, this volume). The dominance of a gene pair over a nonallelic gene pair is termed *epistasis*.

The combined effect of incomplete dominance and epistasis results in a wide range of low-infection types, especially as in the case illustrated in Table IIID when the two corresponding host–pathogen gene pairs result in infection types that are considerably different. This range of low-infection types frequently is seen with a differential host possessing two resistance genes that are both ineffective against a portion of the pathogen population. Marquis, one of the original standard differential cultivars for wheat stem rust, has five genes for resistance to stem rust (Roelfs and McVey, 1979). Assuming Marquis was homozygous for resistance at the five loci and all possible pathogen genotypes exist, then 242 different host–pathogen gene pairs could result in a low-infection type. Such complex host resistance would result in almost continuous variation from the lowest to the highest low-infection type, making it impossible to classify them accurately in the trichotomous key Stakman used for the standard differentials. Distinction of the same cultures for avirulence/virulence (*P_/pp*) on the five "single-gene" host lines for these *Sr* genes is not a major problem.

A race classification based on adult rather than seedling host response has been proposed several times. Although such a classification was not done on a large scale, it may have some value in field studies and with those resistances expressed only in adult plants (see chapter

by Zadoks in Vol. II of this treatise). It would seem to be limited by many of the same factors limiting seedling evaluation. Additionally, if adult plant evaluation were used in the field—where it would be most useful—temperature, race mixtures, and inoculum density would be difficult to control. Currently with wheat stem rust, only *Sr2* is a single-gene adult plant resistance that cannot be adequately detected in seedling plants. For wheat leaf rust, host genes *Lr12*, *Lr13*, *Lr22a*, and *Lr22b* condition adult plant resistances, as do *Pg11* and *Pg12* for oat stem rust. Currently, information is incomplete on the effect of plant growth stage, plant age, inoculum density (Roelfs *et al.*, 1972), temperature, light, and perhaps host nutrition on the response of adult plants to the rust. The host response with *Sr2* is not only in size of lesion (infection type) but also the location of lesions and number of lesions (Sunderwirth and Roelfs, 1980). The latent period (period between inoculation and sporulation) for *P. graminis* f. sp. *tritici* and *P. hordei* is about 7 days for seedlings maintained at 18°C, but it is twice as long for plants inoculated after heading for both wheat stem rust (Sunderwirth and Roelfs, 1980) and barley leaf rust (Andres, 1982). Perhaps this increased length of latent period will be another factor to evaluate in adult plant responses. The disadvantages of using adult plants in the glasshouse as differentials as opposed to seedlings are the need for more space (at least 10 times), a longer time (at least 5 times), more inoculum, and the resulting problem in monitoring and maintaining plants free of other diseases and insects for a longer time.

Bjorkmann (1960) and several others have proposed using detached leaves for identification of races of the cereal rusts. This technique allows a great reduction in space required for growing infected differential series. Disadvantages have been some variation in infection types resulting from the detached leaf culture, and the unique facilities required.

The development of methodology and techniques to handle large numbers of both host and pathogen genotypes has greatly improved race specificity studies. These advances include use of special equipment for planting and inoculating (Browder, 1971), and use of cyclone separators, long-term spore storage, and oil as a spore carrier (reviewed in detail by Rowell, Chapter 10, this volume). These techniques at the Cereal Rust Laboratory permit two people to do all the activities associated with 20 new collections, making 60 single uredia isolates, and taking notes on 60 differential series daily. Thus it is possible to determine the races in a collection within 40 days after it arrives at the laboratory.

IV. Race Nomenclature

Initially, most of the cereal rusts were grouped into races based on an internationally used set of differential hosts. Because of local differences in pathogen virulence and host resistances, investigators gradually adopated local sets of differential hosts that better reflected their needs. Although this change was important in making local progress, it gradually reduced the possibility of international communication. In an effort to improve the international understanding of the diversity and evolution of pathogen phenotypes, the members of the First International Congress of Plant Pathology established a worldwide survey of pathogen virulence. This survey again demonstrated the advantage of international communications. The current use of hosts with "single-gene" resistance could allow international exchange of data and make possible meaningful comparisons of pathogen populations. In fact, exchanges among Australian, Canadian, and United States scientists have already facilitated our understanding of *P. graminis*. This exchange of data has occurred despite the use of different types of race nomenclature and some different differential hosts. These successes certainly would increase if even a basic set could be evaluated worldwide annually.

A. HISTORY

The debate on the type of nomenclature to use for identifying groups of rust biotypes has been continuous since Waterhouse (1929) found that the cultivar Thew (now known to possess *Lr20*) would subdivide the international races of wheat leaf rust. The original systems were all closed systems; that is, there was no provision for adding additional differential hosts to the series. The problem of variation with international races was solved in many ways. In wheat stem rust, supplemental differential cultivars were chosen but without international acceptance. In crown rust, new sets of differential host cultivars were chosen (Fleischmann and Baker, 1971; Simons and Murphy, 1955). Wheat leaf rust races were reduced in number by Basile (1957) by establishment of the unified numeration (UN) scheme, which eliminated the three differentials that were the most sensitive to changes in environmental conditions. This system was later modified further in North America by the use of a system of supplemental differentials (Loegering *et al.*, 1959, 1961; Young and Browder, 1965). There has been a need to add differential hosts as new resistances are found and used or as the pathogen population gains virulence on previously "universally" resistant

host genotypes. Previously differential hosts that become universally susceptible or nearly so to the evolving pathogen population may be advantageously omitted. Thus is is probably unrealistic to assume that race surveys can use an international differential set for more than 10 years. However, continual changing of differential sets also leads to confusion and restricts communication as well as historical points of reference. In oat stem rust, three differential sets have been used since 1970 in the United States (a modification of Stakman *et al.*, 1923; Stewart and Roberts, 1970; Martens *et al.*, 1979).

B. RACE NOMENCLATURE SYSTEMS

No agreement exists on ways to name races or not to name them; however, in general they can be classified into a small number of similar systems. Some workers prefer only a listing of selected virulent and avirulent combinations (Browder *et al.*, 1980). However, as more genes are included the listing becomes longer and longer, or else much information is omitted that may be valuable in understanding pathogen populations. Race nomenclature is merely a means to simplify communication of information. Long designations are expensive to publish and difficult to communicate accurately, and they may be understood only by those who use them regularly. Some of the systems of race nomenclature in current use are shown in Table IV. Most of the current systems are based on differential series with host lines having a single known gene for disease resistance. The comparisons are based on a low- and high-infection type per host–pathogen gene pair. For comparison purposes, the system used by Stakman and Levine (1922) is included, and because this system had 12 differential hosts, all the values were calculated on that basis. Most of the early systems (Table II) had two classes of low and one class of high-infection type, and thus were trichotomous. Infection types from 0 (immune) to 2 were classified as resistance host response, and the X infection type was classified as a mesothetic host response. Infection types 3 and 4 indicated that the host had a susceptible response. The use of a trichotomous key with 12 host differentials would result in 531,411 instead of the 4096 races obtained with a dichotomous or high–low system.

C. OPEN-ENDED SYSTEMS

The original systems for race identification were all closed; that is, no provision was made for the inclusion of new differential hosts. An

Table IV

Comparison of Race Classification Systems Assuming a Low- and a High-Infection Type for Each of 12 Host–Pathogen Pairs Using Hosts with "Single-Gene" Resistance (Total of 4096 Pathogen Races)

Differential system[a]	Type of system	Type of key	Race assignment	Unique nomenclature combinations	Mean no. of characters/code[b]
Stakman	Closed	Trichotomous	Chronologic	531,411[c]	5.8
Dichotomous	Closed	Dichotomous	Chronologic	4096	3.7
Virulence formula	Open	None	None	4096	27.0[d]
Coded virulence formula	Semiopen[e]	Virulence list	Chronologic	4096	3.7
Modified potato–*Phytophora infestans*	Open	Host list	Preassigned	4096	12.2[f]
Binomial and decanary	Open	Mathematical	Preassigned	4096	3.7
Octal notation	Open	Mathematical	Preassigned	4096	3.8
Coded sets	Semiopen[g]	Dichotomous	Preassigned	16[h]	3.0

[a]References: Stakman *et al.* (1962), Loegering *et al.* (1959), Samborski (1968), Green (1965), Watson and Luig (1961), Fleischmann and Baker (1971), Habgood (1970), Gilmour (1973), and Roelfs and McVey (1972).

[b]Assuming all races are found, the number of digits or letters that describes the pathogen phenotype.

[c]Based on 3 host reaction classes for 12 hosts (3^{12}).

[d]A character code per pathogen gene separated by a space, comma, or slash.

[e]Addition of differential host requires new code or retesting of type cultures.

[f]Susceptible hosts are listed separated by a space.

[g]Sets of differential hosts can be added; less than a set require a partial virulence formula.

[h]Based on 3 sets of 4 host differentials, 16 combinations per set; thus 16^3, or 4096 phenotypes, are described.

ideal system of race nomenclature would consist of a short code, easily obtained, and open-ended so additional differential hosts could be added without greatly changing the nomenclature. In order to obtain a usable system, compromises normally are made. Because the coding in the modified potato–*Phytophthora infestans* system only indicates the susceptible hosts, it is impossible to determine what differential hosts were evaluated (Table V). Thus the code is the same when a host is resistant or not evaluated. For example, in Table V, culture 96, races 1, 2, 3, 4, 8, and 9 indicate that hosts 1, 2, 3, 4, 8, and 9 were susceptible, and that if tested, hosts 5, 6, and 7 were resistant. Further, it is impossible without a list of differential hosts to know if hosts 10 through 12 were tested, were resistant, or were some combination of the two (Table V). In articles about physiologic race surveys a host list is usually provided that eliminates the confusion; however, in articles lacking a list there is no way to tell which differentials were resistant or not tested. Some systems have solved this problem by indicating a year with the race or by designating the differential host set by a code, making it possible to find the pathogen phenotype. Of the open-ended systems examined, the binomial (Fleischmann and Baker, 1971), decanary (Habgood, 1970), and octal notation (Gilmour, 1973) seem to offer the most advantages; however, all may be too complicated for those who work with the system only occasionally. Addition of a new host differential that is susceptible can change the race designation considerably; that is, for the decanary and binomial systems, races 0 and 1 (host R) become 2 and 3 (hosts S,R), and likewise races 3 and 4 (hosts S,S) change to races 7 and 8 (hosts S,S,S), respectively. In the octal notation race 0 becomes 2 and race 3 becomes 7; however, adding an additional differential that is susceptible retains the last digit thus, hosts S,S,S,S is race 17 and hosts S,S,S,S,S, is race 37.

D. RACE KEYS

Most of the systems, except for the virulence formula and modified potato–*Phytophthora infestans* system, require some sort of key or mathematical device for assigning race codes. The modified potato–*Phytophthora infestans* system, however, requires a listing of the hosts (see also wheat and rye stem rusts chapter by Roelfs in Vol. II). The easiest and quickest systems for assigning races are the simple mathematical and short dichotomous keys. Ideally, the key should be simple enough for the daily user to learn and for others to use within a few minutes. The race codes can be assigned in many ways, but a

Table V

Examples of Race Designations Using the Various Systems of Race Nomenclature[a]

Culture	Response on differential hosts											
	1	2	3	4	5	6	7	8	9	10	11	12
1	S	S	S	S	S	S	S	S	S	S	R	R
40	S	S	S	S	S	S	S	S	S	S	S	R
63	S	S	R	S	S	S	S	S	S	S	R	R
75	S	S	R	S	X	X	X	S	S	S	R	R
96	S	S	S	S	R	R	R	S	S	R	R	R

Type of system	Race designation for culture				
	1	40	63	75	96
Stakman	11	15	17	29	56
Dichotomous	11	15	17	17	56
Virulence formula	1,2,3,4,5,6,7, 8,9,10/11,12	1,2,3,4,5,6,7, 8,9,10,11/12	1,2,4,5,6,7,8,9, 10/3,11,12	1,2,4,8,9,10/ 3,5,6,7,11,12	1,2,3,4,8,9/ 5,6,7,10,11,12
Coded virulence formula	C12	C-9	C-1	C-1	C-17
Modified potato–*Phytophthora infestans*	1,2,3,4,5, 6,7,8,9,10	1,2,3,4,5,6, 7,8,9,10,11	1,2,4,5,6, 7,8,9,10	1,2,4,8,9,10	1,2,3,4,8,9
Bionomial	4093	4095	3581	3357	3865
Decanary	4092	4094	3580	3356	3864
Octal notation	7774	7776	6774	6434	7430
Coded sets	TTQ	TTS	RTQ	RCQ	TCL

[a]Hypothetical data were chosen to demonstrate differences between systems.

preassigned number (the race designation is determined before the race is actually found) eliminates a delay in communication until a new key is issued, a major fault of chronological keys. With 12 differential hosts and a dichotomous system, the number of possible unique combinations becomes large—4096 (2^{12}, a number of alternatives to the power of the number of differential hosts).

A coding system shorter than a virulence formula can be developed by placing the sets of differentials in subsets (e.g., set of 4) and then using a repeating system for additional sets (Roelfs *et al.*, 1982). For each set of 4 differential hosts, there are 16 unique combinations (2^4) of high- and low-infection types. These 16 combinations were arranged in a dichotomous system from all low- to all high-infection types. Each combination was assigned an English letter code in alphabetic order using consonants only (B through T). Thus the combination of 4 low-infection types on the first subset of differentials is coded B, and of 4 low-infection types on the second set of differentials is also coded B. Thus the 4096 unique pathogen phenotypes in a 12-differential system can be divided into 3 sets of 4 differentials each and be described by means of 16 codes repeated three times. The biggest disadvantage of the system is that it is only semi-open-ended. Its utility is the shortness of the race notation—which in the studied systems varied from 3.0 in the coded sets to 24.0 with a virulence/avirulence formula (Tables IV and V)—and its ease in coding and decoding (see also Roelfs' chapter on wheat and rye stem rusts in Vol. II).

V. Source of Collections

The usefulness of a race survey depends on the source of samples. Ideally, the collections should be made on a stratified random basis, but currently this is not done. Thus an effort has been made to increase the number of samples to compensate for some nonrandomness in sampling. In many cases quality of sampling could replace quantity of collections if quality collections could be defined and obtained at a cost within the economic limit. Currently, the number of samples is usually correlated with the ability to detect races occurring at a low frequency. The lower the frequency at which a hazardous culture is initially detected, generally the longer the time available for finding new sources of resistance and developing them into cultivars, or for initiating other control strategies. With organisms that have as high a reproduction rate as the cereal rusts, no number of samples that can be

handled, even with current technology, will result in all pathogen phenotypes being detected. Thus, even in years when over 2600 isolates from 900 collections from the United States were studied, some physiological races were still detected only once (Roelfs and McVey, 1975). Other races were detected only once in several years, so either the same mutation has recurred in otherwise the same pathogen phenotype, or, more likely, the race has existed undetected.

A. SAMPLES FROM COMMERCIAL FIELDS AND WILD HOSTS AT PEAK DEVELOPMENT

The most important and most nearly randomly obtained cultures studied are those made in commercial fields at the height of rust disease development. These collections would be improved if they were collected on a purely random basis. Random sampling was attempted in the Dakotas and in northeastern Montana one year with wheat stem rust, but such a survey was very expensive, and no rust was found even though using traditional methods rust was found in trace amounts. The expense involved locating the random points and getting to them, as most sites were not near roads. Furthermore, experienced personnel in field surveys gain a sense of where to look for rust, as its location in the field and on a plant varies with the environmental conditions. In the year the random survey was done, stem rust was limited to field edges in low-lying areas of late-planted fields. The field edges were not considered in the random survey, and the majority of the fields were upland, thus reducing the likelihood of finding the pathogen in the random survey. Therefore, the Cereal Rust Laboratory has continued with the traditional survey. In the United States, this survey involves approximately six trips over 25,000 km (15,000 miles), with stops at the first small-grain field after each 32 km (20 miles) on the car odometer. The routes are prechosen through the major cereal-producing areas where rust has historically been a problem. To these collections are added collections made by other cooperators, which may or may not be taken on a systematic basis, but the collections are from commercial fields or wild hosts. These samples provide the basis of our data on pathogen phenotype frequency and distribution. Because of host resistance, however, such data indicate only the extent of the disease spread but give no clue to viable pathogen spore dispersal. Because most urediospores are generally produced in the last 10 days of the epidemic, races occurring primarily early in the season, races originating outside the area arriving late in the season, mutants occurring on a

previously susceptible cultivar, and nonagressive phenotypes can be missed totally in these samples.

B. SAMPLES FROM COMMERCIAL FIELDS AND WILD HOSTS EARLY IN THE SEASON

Collections are also made early in the season from the first uredia observed. The early collections from the overwintering area and from the early exogenous inoculum in the northern areas are used in epidemiologic studies on disease movement. The early-season surveys are conducted in the same manner, and because of the limited number of total collections for the purpose of studying pathogen distribution and frequency, data from these collections are included with the other data from commercial fields.

Generally, the pathogen phenotype found initially in an area is the most common race at host maturity. This is because the short time between initial infection and host senescence is sufficient to permit only a few pathogen generations. Exceptions normally occur when a previously resistant host cultivar, which is planted on a significant part of the area, is highly susceptible to a race that appears late in the season.

C. SAMPLES FROM NURSERIES AND PLOTS

Samples of rust are also taken from uninoculated nurseries and trap plots. Although this inoculum is part of the natural rust population, the selection pressure created by the combinations of host resistance, or lack of resistance that exists in such nurseries often affects the frequencies of the races found. In the case of oat stem rust in the United States, race NA-27 predominates in the production fields, and race NA-16 is seldom found except on wild oats (*Avena fatua* L.) and on nursery lines without *Pg2* and *Pg4* (Roelfs *et al.*, 1980). These two host genes nearly eliminate NA-16 from commercial fields.

In nurseries, unique combinations of host resistance may exist on which a virulent but generally nonaggressive race may increase without competition from its avirulent aggressive competitors and thus may be detected. The real value of trap plots is when unique host genotypes are used that can detect virulent pathogen phenotypes, perhaps before they become a major part of the pathogen population. However, with asexual reproduction most pathogen variation is due to mutation; thus new races would occur at random. Because most spores

are produced in commercial fields, most mutations would occur there, but they could multiply selectively on appropriate host genotypes in nurseries. For the detection of unique pathogen phenotypes and the estimations of viable inoculum movement, we have maintained 50–75 trap plots in the eastern two-thirds of the United States for the past 10 years. Data from these plots are included in the annual race survey reports but are kept separately from collections made in commercial fields and on wild hosts. Otherwise, cultures with many virulence or avirulence genes, and cultures with little aggressiveness could be overemphasized. This is especially true in those years when the number of collections is low because of a sparsity of rust, as in 1980 when, of the 54 collections of wheat stem rust that were made, 45 (83%) of them were from nurseries (Roelfs *et al.*, 1982).

D. SAMPLES FROM THE ALTERNATE HOST

Samples from the alternate host can also be very important in areas where it is a significant source of inoculum. A problem arises because of the high number of pathogen phenotypes produced (Roelfs and Groth, 1980). Roberts *et al.* (1966) used a screening nursery of cereal hosts of varying genotypes adjacent to a barberry hedge to aid in selecting pathogen phenotypes of a potential threat to crop production. Caution must be used, as most cultures from sexual recombination commonly have avirulences for previously undetected host resistances. Some of these cultures are unusual in being avirulent on *SrLC*, *Sr16*, *SrMcN*, while being virulent on *Sr6*, *Sr10*, *Sr17*, etc. Thus the differential host can be resistant because of a previously undetected resistance gene. Because teliospores and basidospores travel only short distances, unique local populations may occur near the alternate host. This means that more samples are required per unit of land area. In isolated areas where the alternate host is not an important source of inoculum for cereal crops, rust collections can still serve as an important source of new pathogen phenotypes that are useful in genetic studies of the host.

E. SAMPLES FROM INOCULATED NURSERIES

Data from collections made in inoculated nurseries should not be included in reports on pathogen frequency or distribution. Our procedure has normally been to exclude frequency data of any collections made within 2 to 10 km of a known inoculated nursery depending on

its size. The collections, however, provide valuable information on frequency of races in nurseries and thus aid in decisions on host resistances. Some information is also obtained about survival of races in composite inoculum after several generations of uredial increase.

VI. Importance of Type Cultures

With the advent of vacuum drying, liquid nitrogen, and ultra-low refrigeration storage of urediospores (Rowell, Chapter 10, this volume), cultures can be maintained indefinitely with relative ease. Thus unique pathogen cultures can be preserved and used as a type for physiologic races. In early physiologic specialization studies, most comparisons were between data on a previously studied race and the currently available isolates of that race. It was impossible to determine whether the differences or similarities in infection types were due to differences in environmental conditions, host resistance, or pathogen virulence. The latter, of course, is the only reasonable basis for race designations. It is now possible with better storage methods to compare cultures directly, to provide the much-needed historical continuity. These stored cultures are also useful in verifying that differential host lines have remained genetically identical for resistance to that pathogen. We have had several experiences in which the least effective of two or more genes for stem rust resistance was lost from the population over a period of years despite normal procedures for maintaining seed purity. Stored cultures assure that the same culture can be used throughout a breeding, genetic, or other type of study.

VII. Single Uredium Isolates

The understanding of race distribution and the ease in classification of infection types can be improved by the use of single uredium isolates. Bulk collections of urediospores used to inoculate differential hosts often result in a mixture or range of infection types on a single differential host. This can mask some low-infection types completely, such as the infection type 0 obtained with *Sr5* or *Sr36* (*Tt1*) in wheat stem rust (Roelfs and McVey, 1979). This can also result in the missing of an X infection type or misclassifying a mixture of infection types as an X infection type. Single uredium isolates eliminate this problem.

Stakman *et al.* (1962) outlined the procedure of making separations when bulk collections were used to inoculate differential series. Although processing a bulk collection is initially faster than making single uredium isolates first, the separations generally required subsequently when using bulk collections often lengthen that process considerably. Separations are also difficult to handle in a determination of race frequency. Separations consist of isolating one or more single uredia from about 1000 that occur on a differential set. The progeny of each uredium is then inoculated to a separate differential series.

However, the frequency at which it occurred in the field population is then impossible to estimate. Contamination may also occur in race survey cultures because of the large volumes of materials handled. These contaminants are often isolated when making such separations. The initial use of single uredium isolates reduces the contamination problem and solves the problem of estimating frequencies. The number of isolates needed per collection may vary depending on the variation in the pathogen population, although three or four seem adequate (Roelfs and Johnston, 1966). Single uredia are easily obtained by inoculating maleic hydrazide-treated host plants (see Rowell, Chapter 10, this volume). Six to ten plants are planted in clumps at the four corners of a 7 × 7 cm pot. When uredia erupt, four isolated uredia on separate plants are saved, and remaining diseased tissue is clipped out. The plants are reincubated to germinate loose urediospores and then placed in isolation cages for 24 to 48 hr, after which cyclone separators are used to collect rust separately from three single uredia. Each uredium furnishes adequate inoculum to inoculate as many as six to eight seedlings each of up to 24 cultivars in the case of stem rust. For leaf rust of wheat and barley and crown rust of oats, the single uredium isolates usually are increased on a susceptible host before inoculating differential hosts, which lengthens the time for identification by 2 weeks.

VIII. Selection of Differential Hosts

The differential hosts chosen determine the usefulness of race data. The original differential hosts were all those discovered that resulted in a different host response pattern when infected with the limited number of cultures available. Using every differential host known is no longer desirable for most of the cereal rust pathogens, as thousands of resistant host lines are known. We have comprehensively studied 45 different "single-gene" host resistances (Roelfs and McVey, 1979) and

now are investigating 12 more for wheat stem rust. Thus some selection of differential hosts must be made even when "single genes" are being used. In selecting differential cultivars, investigators give varying weight to different factors. Six factors that merit important consideration are discussed in the following paragraphs.

A. HISTORICAL BASE

A historical base is important. Not all races are detected annually; therefore, several years of data are needed to build a working base. In the case of wheat stem rust, most of the original host differentials had a multiple-gene basis for their resistance. However, an examination of the historical data for the United States showed that *Sr5*, *Sr7b*, *Sr9d*, *Sr9e*, and *Sr21* were the resistances on which separation of races had been done throughout the surveys. These resistance genes were those for which the differences between low- and high-infection types were clearly distinguishable and that were stable over a range of environmental conditions. Thus with the current asexual population of *P. graminis* in the Great Plains, it is possible to predict the international race by knowing the response of *Sr5*, *Sr7b*, *Sr9d*, and *Sr9e*. The response of *Sr21* is unnecessary, as a low-infection type is obtained only when *Sr9d* is also low. This combination of genes does not work with a sexually reproducing population or one in which other genes in the standard differential host series were important in determining race differences as they exist outside the Western Hemisphere.

B. RESISTANCE USED IN COMMERCIAL CULTIVARS

Genes that are used or that are under consideration for use in commercial cultivars often are valuable differential hosts. In some areas of the world, race-specific resistances may exist in other native, cultivated, or escaped hosts and may also be important to consider. Differential hosts used should have a differential reaction; that is, they should not be susceptible or resistant to all cultures evaluated from the population.

C. STABILITY OF THE DISEASE INFECTION TYPE

Ideally, hosts chosen as differentials must not be severely affected by the range of temperatures, light, and inoculum density likely to occur

during the race survey. Even though *Sr6* is sensitive to small changes in temperature, we have successfully used this host gene. It is stable enough to use in the greenhouse when the temperature fluctuates with a daily minimum of less than 21° or 22°C (Roelfs and McVey, 1979). Further, when differential hosts are examined daily with similar pathogen genotypes, an investigator becomes accustomed to the gradual changes in infection types caused by changes in temperature and light (e.g., a change with *Sr6* from a 0; to 3$^+$C over a period of a month. Additionally, with "single-gene" host lines, the high-infection type on the background line without *Sr6* is consistently 44+. Some other lines (i.e., *Sr7a* and *Sr15*) are so sensitive to environmental conditions that they are of little use in a race survey (Roelfs and McVey, 1979).

D. USEFULNESS OF INFORMATION

The host differentials must also provide information useful to the purpose of the survey. In the United States, for example, *Sr12* occurs in many of the host cultivars grown in the spring wheat area, but it provides resistance only against one race that is already distinguished by the infection types produced on other more stable host lines. This example is similar to the one of *Sr21* given in Section VIII,A; however, *Sr21* does not occur in the commercial cultivars grown in the United States. This does not necessarily indicate that these host genes or the virulences on them are linked; it merely reflects that in the asexual population we are sampling, all cultures avirulent on *Sr9d* are also avirulent on *Sr12* and *Sr21*. This association should remain until a mutation for virulence occurs at one of the loci or until sexual or parasexual recombination occurs.

E. "SINGLE-GENE" DIFFERENTIALS

Although it is impossible to be sure a host line has a single gene for rust resistance, it is possible to establish lines that have only one gene that is effective against the pathogen population available for study. These "single-gene" lines have many advantages as differential hosts, the chief one being that the pathogen phenotype is clearly measured. With multiple-gene differential hosts, some interactions are masked because of epistasis, and specific low-infection types may be difficult to distinguish because of the large number resulting from the many gene-for-gene interactions as described earlier (Table IIID).

A "single-gene" line has a smaller range of low-infection types, and

thus environmental effects are of less importance. Often when a multigene differential is evaluated, epistasis and similar low-infection types produced by different host genes prevent a complete determination of the effect of each individual host gene. Thus a low-infection type on the tested multigene differential may not help in predicting the host response of other untested hosts. The use of "single-gene" differential host lines often enables one to predict the infection type of host lines with a known genotype. For example, the cultivar Selkirk has *Sr6*, *Sr7b*, *Sr9d*, *Sr17*, and *Sr23* and could be susceptible to a given culture, whereas Bowie with *Sr6* and *Sr8* was resistant, and vice versa. However, if the "single-gene" differentials *Sr6* and *Sr8* are susceptible, then Bowie inoculated with the same culture will also be susceptible; if a culture inoculated to *Sr6* results in a fleck, then Bowie will result in a fleck; if *Sr8* results in a type 2 and *Sr6* a 4, then Bowie will be a fleck. A note of caution, however; other genes in the host can modify the infection type expression and in rare cases can completely change it (Dyck and Samborski, 1982; Kerber and Green, 1980).

F. SEED AVAILABILITY

When a host line with a new resistance is found, insufficient seed is generally available to use it in the race survey. However, a line with limited seed can be used as a supplemental differential for a part of the pathogen population, by being tested only against selected cultures. Some of the currently available "single-gene" host lines are very difficult to grow in some areas of the world because of a lack of adaptation, and thus inadequate seed stocks continue to be a major problem.

IX. "Universal" Resistance Series

Most race surveys in time identify a group of cultivars, lines, or "single-gene" lines that are resistant to all the cultures evaluated (Loegering *et al.*, 1959, 1961). In a few cases, these lines are treated as differential hosts, although they obviously are not. They are more effectively evaluated in a special "universally" resistant series. This series is primarily used for detecting new virulence or combinations of virulence. It is not needed to evaluate frequency or distribution in the main survey. Thus the use of bulk collections instead of single uredium isolates is more efficient. The bulk used is composed of a

portion of the urediospores from each collection received from a particular area or cultivar over a period of days. The infection density on the susceptible check (low, medium, or high) is noted, and the low-infection types for each line are compared against the expected range of low-infection types. A high-infection type on a resistant line normally indicates a new gene or combination of genes for pathogen virulence. A higher than expected low-infection type, if stable, usually indicates a virulence for the previously expressed resistance gene, but avirulence on another host gene previously not expressed as a result of epistasis. The cultures producing a higher low-infection type and/or high-infection type are isolated and (re)evaluated on the differential and "universal" resistance series. Off-type plants (those without the desired resistance genes) can be a problem in this series; however, if the high-er(er) infection types are on a single plant it is almost always an off-type plant. This kind of series is a powerful tool for detecting new virulences although probably not as effective as planting large trap nurseries of resistance genotypes in scattered locations.

X. Prospects

A long-range emphasis needs to be placed on methodology of field sampling. Current sampling methods have many biases of unknown effect. In comparison to the large numbers of pathogen individuals, even the largest survey samples only a minute part of the population. Major gains in sample quality are not expected in the immediate future. The possible and probable sources of inoculum for most major cereal-producing areas should be delineated. This would allow early sampling of the population and an early forecast of the pathogen phenotype perhaps before the crop is planted in some areas.

Additional markers (other than virulence markers) should be available for use in studying pathogen populations in the future. These additional markers may permit more conclusive studies of changes in the pathogen population. Studies on mutation rates of individual pathogen virulence genes from asexually reproducing populations will permit a better basis for selecting resistance sources for use in commercial cultivars.

"Single-gene" lines will be developed for many more resistances. These resistance genes will be placed in genetic backgrounds that result in more stability in the resulting infection types with a wide range of cultures and environments. Studies of interorganismal genetics will

clarify the differences among infection types formed by all the combinations of host–pathogen gene pairs. Interactions between pairs of host and pairs of pathogen genes will be studied. These category IV interactions (see Loegering, Chapter 6, this volume) may open a whole new field of understanding of host–pathogen relations and result in major changes in the way host–pathogen interactions are viewed in race surveys. New nomenclature systems may be required to express these interactions.

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