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AVERMECTIN B_1a: EFFECTS ON THE OVARIES OF RED IMPORTED FIRE ANT QUEENS (HYMENOPTERA: FORMICIDAE)

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Abstract. Avermectin B_1a causes irreversible cell and tissue damage to the ovaries of queens of the red imported fire ant, *Solenopsis invicta*. The damage is characterized by hypertrophy of the squamous epithelium that sheaths the ovarioles and pycnosis of the nurse cell nuclei, and results in complete sterility or reduction in the numbers and size of eggs laid.

Chemicals for use in baits for control of ants can be of 2 types. One type is characterized by delayed toxicological effects, and most or all members of the colony are affected. The 2nd type in some way affects colony reproduction, acting directly on the fertility of the queen or on larval development. The latter type is effective because if the queen is killed or her reproductive ability is lost, the colony will die (Tschinkel & Howard 1980). The feasibility of control through action on the queen was demonstrated by Edwards (1975) with *Monomorium pharaonis* (L.) when he showed that queens of colonies treated with the juvenile hormone analogue methoprene [isopropyl (*E,E*)-11-methoxy-3,7,11-trimethyl-2,4-dodecadienoate] were irreversibly sterilized. Dissection of the queens revealed that their ovaries were reduced in size and lacked both oocytes and trophocytes (nurse cells).

Recently, Lofgren & Williams (1981) described a new type of chemical that inhibits reproduction in queens of the red imported fire ant (RIFA) at extremely low dosages. This compound, avermectin B_1a, is a macrocyclic lactone glycoside that was isolated from the soil microorganism *Streptomyces avermitilis* (Putter et al. 1981). The compound, fed to laboratory colonies at concentrations of 0.0025 to 1.0%, either killed the queen or induced irreversible sterility. Lower concentrations caused a decline in brood production; however, a return to normalcy was observed in 16 weeks. Histological examination of the ovaries of some of these queens showed severe tissue and cell abnormalities. This paper presents a description of these effects.

MATERIALS AND METHODS

Five queens from colonies treated 22 weeks previously with avermectin B_1a and that were expressing permanent sterility effects were killed; ovaries were removed, fixed in Bouin's solution and embedded in paraffin (mp: 57 °C). The tissue was sectioned at 5 μm and stained with Harris's hemotoxylin and eosin. Three normal colony queens

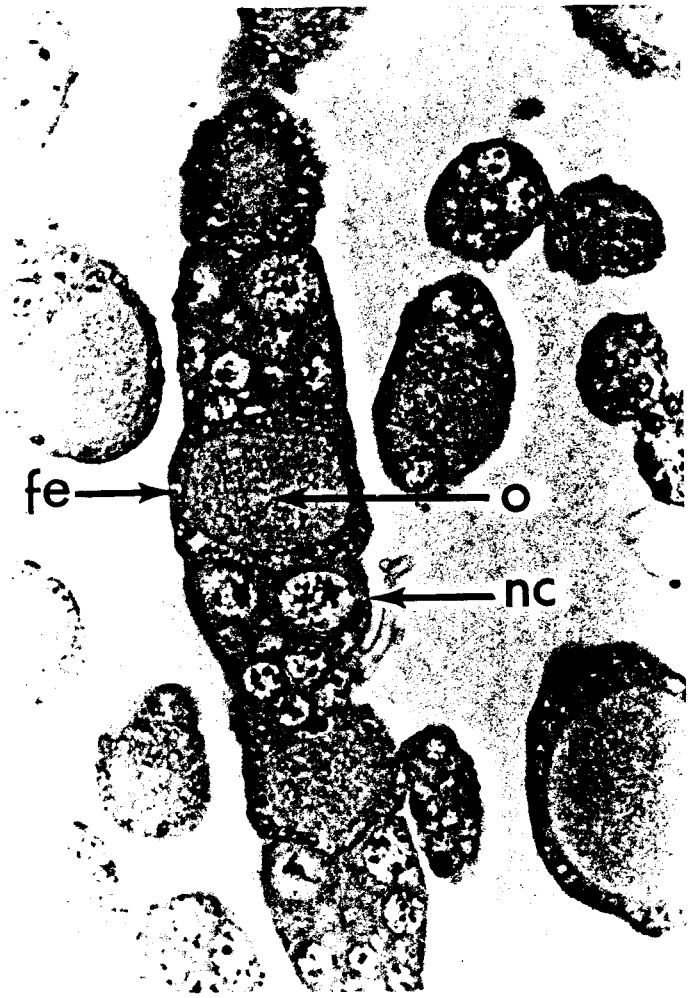

FIG. 1. Longitudinal section of meroistic fire ant ovariole showing oocyte (o) surrounded by follicular epithelium (fe) and preceded by area of nurse cells (nc). 385×.

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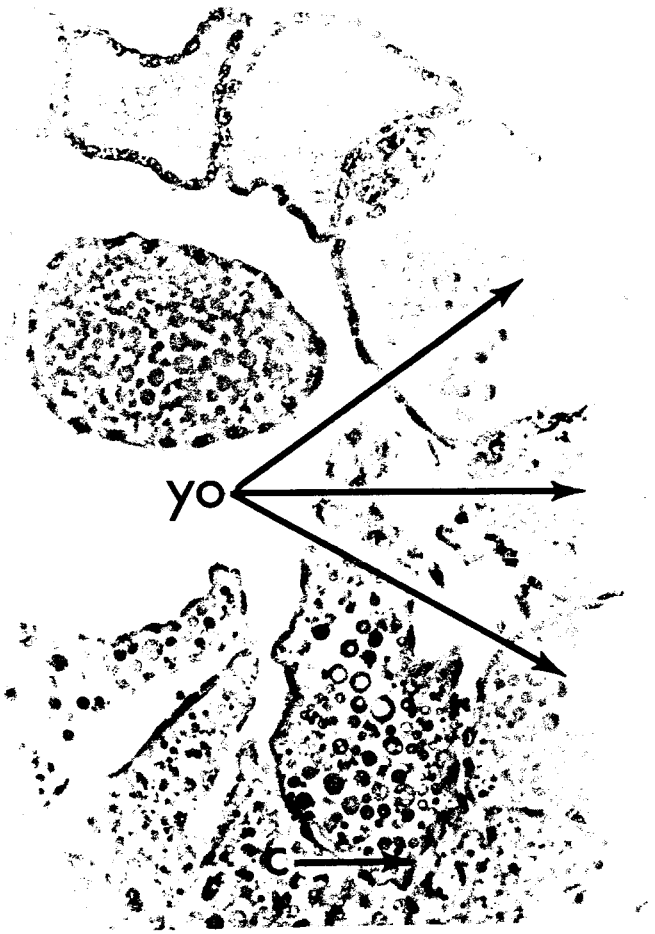

FIG. 2. Longitudinal section of distal end of ovariole showing calyx (c) and 3 oocytes containing yolk (yo). 190×.

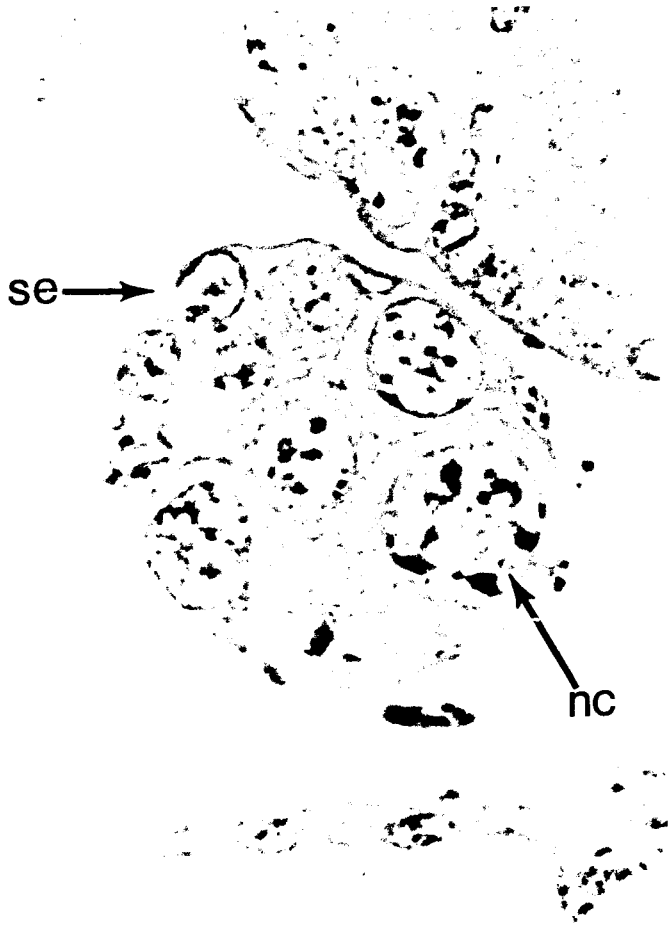

FIG. 3. Cross section of ovariole showing normal chromatin distribution in nurse cells (nc) and the thin sheath of squamous epithelium (se). 500×.

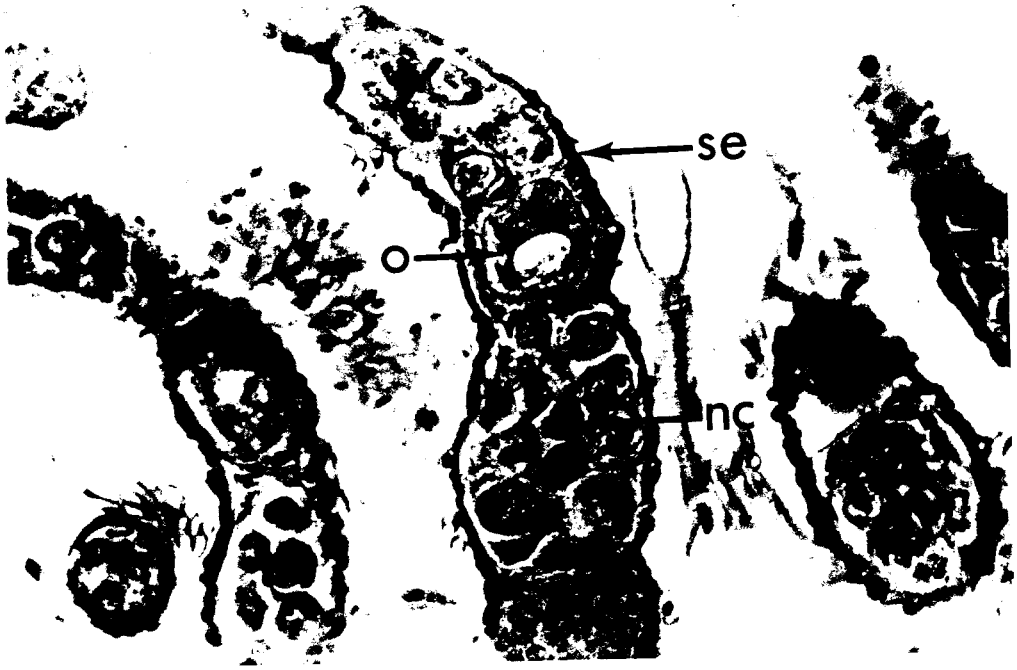

FIG. 4. Longitudinal section of ovariole of avermectin-treated queen showing hypertrophied squamous epithelium (se), an oocyte (o) and a group of nurse cells (nc). 400×.

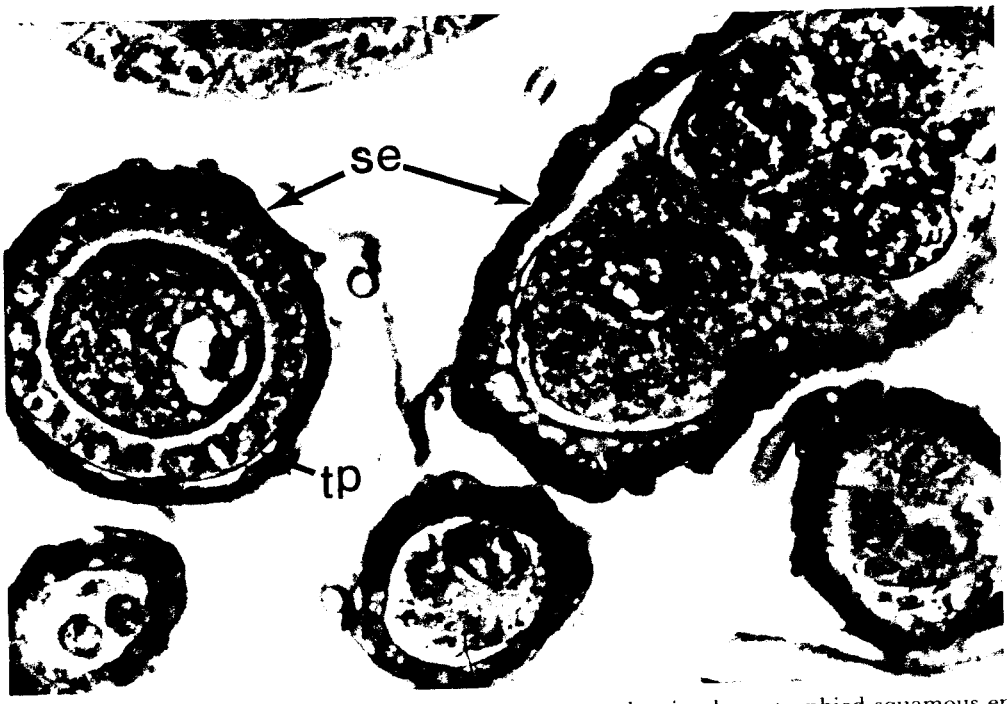

FIG. 5. Cross section of ovarioles of avermectin-treated queen showing hypertrophied squamous epithelium (se) and the tunica propria (tp). 500×.

were also sacrificed and fixed to serve as controls. The spermathecae of all queens were removed, placed in Ringer's solution and examined for sperm motility.

RESULTS

The ovarioles of RIFA queens are of the meroistic type (Hermann & Blum 1965), with each ovariole essentially an egg tube with 4 distinguishable regions as follows: (1) a terminal filament or suspensory ligament that attaches the ovariole to the body wall; (2) the germarium, which contains oogonia that differentiate into primary oocytes and nurse cells (trophocytes), and somatic mesodermal cells (cuboidal epithelium) at its base that form the prefollicular tissue; (3) the vitellarium, which con-

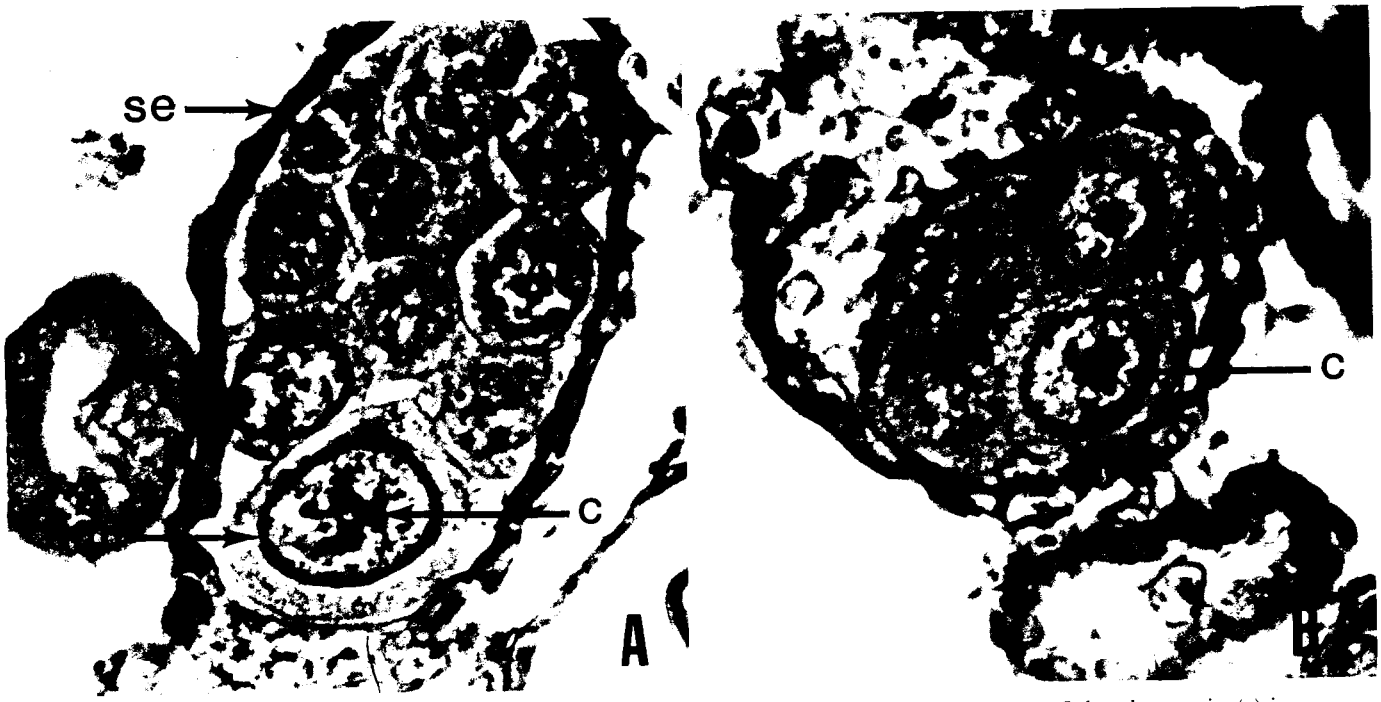

FIG. 6A, B. Cross section of ovariole of avermectin-treated queens having clumping of the chromatin (c) in a nurse cell with a thickened nuclear membrane (nm). 500×.

FIG. 7. Longitudinal section of avermectin-treated queen showing an ovariole occluded with connective tissue (ct). 480×.

stitutes the major part of the ovariole and consists of a series of developing oocytes (ca. 16 with 4 to 5 maturing oocytes with yolk) enclosed in follicular epithelium, with a packet of nurse cells located at the anterior end (Fig. 1, 2); and (4) the calyx area or union of all the ovarioles (Fig. 2). The entire ovariole is sheathed with simple squamous epithelium (Fig. 3) separated from the developing oocytes by a thin, structureless tunica propria.

Examination of the avermectin-treated queens with the aid of a dissecting microscope showed the ovaries to be translucent to milky white and reduced in size, with only a few small eggs.

Microscopic examination of the sectioned ovaries showed gross morphological changes. Immediately apparent was the hypertrophy of the squamous epithelium entirely covering the ovarioles (Fig. 4). In some cross sections the squamous epithelium/tunica propria tissues were 4× the thickness of the normal tissue (Fig. 5). The prefollicular and follicular epithelium appeared normal in size; however, it stained very heavily in contrast to the normal tissue.

Although the nurse cells appeared normal in size, the nuclei were extremely distorted, showed irregular distribution of the chromatin (pycnosis) and a thickening of the nuclear membrane (Fig. 6A, B). The lumen of some of the ovarioles appeared to be infiltrated with connective tissue. In fact, the tissue was so thick in places that the lumen was occluded (Fig. 7). Finally, the number of developing oocytes per ovariole was reduced to 2 or 3 and these were very small. Only 1 small maturing oocyte with yolk was found in some ovarioles.

Motility of sperm in the spermathecae of the queens was normal in the controls but appeared to be reduced by 40–60% in those queens exposed to avermectin B_1a.

DISCUSSION

According to Wigglesworth (1973), egg production and maturation normally begins with the differentiation of oogonia in the germarium into oocytes and nurse cells. As the oocyte begins its journey toward the oviduct, it is surrounded by simple cuboidal epithelial cells, which eventually form an egg follicle around the oocyte 1 cell layer thick. The nurse cells, which accompany the developing egg, obtain nourishment from the blood and pass it on to the oocyte. Ultimately, the nurse cells break down and are absorbed into the egg. In the late stages of egg development, the eggs are nourished by secretions from the follicular epithelium.

The development and activity of the aforementioned reproductive system is generally considered to be under the control of hormones secreted by the neurosecretory cells of the pars intercerebralis (Chapman 1969, Wigglesworth 1973, Gilbert & Sking 1973). Secretions of the neurosecretory cells are responsible for vitellogenesis, having a direct effect on protein synthesis, including yolk protein, and an indirect effect via the corpora allata (Engelmann 1970).

Experiments with chemosterilants and insecticides have shown that the function of the neurosecretory cells can be interfered with; however, the mechanism by which this occurs is unknown. For instance, Taneja et al. (1979) showed that apholate suspended the release of a neurosecretory sub-

stance from the pars intercerebralis for 3 days in the red cotton bug *Dysdercus koenigii* (Fabricius). Riviere (1977) found that the insecticide propoxur prevented vitellogenesis in the roach *Blatella germanica* (L.), and an endocrine lesion was postulated to explain the effect.

Although previous studies with avermectin B_1a showed that the chemical affected muscle membrane resistance in lobsters (Fritz et al. 1979), the gross pathology of the RIFA queen's reproductive system lends support to an endocrine lesion hypothesis. The extreme hypertrophy of the squamous epithelium suggests that the products of metabolism are being used to produce larger epithelial cells, or more of them, rather than for the production of eggs. The abnormal clumping of the chromatin (pycnosis) in the nurse cells would in all probability interfere with the nourishing of the eggs by the nurse cells. A similar clumping effect has been reported by Morgan & LaBrecque (1962) for house flies treated with apholate. Comparison of slides of apholate-treated house fly nurse cells with those of RIFA ant queens treated with avermectin B_1a revealed a striking similarity. Finally, the lack of eggs containing yolk, the very small number of eggs produced and the very small size of the eggs point to an effect on the neurosecretory cells.

Although it may be some time before the mode of action of avermectin B_1a is elucidated, its ability to disrupt the reproductive system and cause irreversible sterility in queens may make it a useful compound for control of the red imported fire ant.

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