homogenate (1 part wet-weight CAF$_1$ liver homogenized in twice its weight of 0.15M KCl); 1 ml of 0.067M phosphate buffer at pH 6.8; 1 ml of 0.01M glutathione; and the volume of ALA and analog added as indicated in Tables 1 and 2. After evacuation of air, the mixture was activated at 37°C for 1 hour. In the experiments whose results are given in Table 1 and the lower half of Table 2 (tubes 5 to 8) the ALA was added 10 minutes after the analog. In the experiments in the upper half of Table 2 (tubes 1 to 4) the ALA and analog were added simultaneously. The incubation was continued for 1 hour and the amount of porphobilinogen was determined by use of Ehrlich's reagent after precipitation of protein as described by Gibson, Neuberger, and Scott (*1*). Pooled homogenates were used in the study of each compound. The percentage inhibition was calculated from the formula

$$100\ [1-\ (P_I/P_S)]$$

where P_I is the amount of porphobilinogen formed in 1 hour in the presence of inhibitor and substrate and P_S is the amount of porphobilinogen formed in 1 hour in the presence of substrate alone.

In Table 1 are presented the inhibitory effects of a group of compounds on ALA dehydrase. All the compounds produce inhibition of varying degrees at high concentrations, but only δ-oximinolevulinic acid is inhibitory at low concentrations. Two different concentrations of ALA (using 1.32 ml of $7.5 \times 10^{-3}M$ ALA and 0.33 ml of $5 \times 10^{-2}M$ ALA) were studied with the same ratios of δ-oximinolevulinic acid to ALA in both cases. The percentage inhibition is constant for a given ratio of inhibitor to analog at the different ALA concentrations. In these experiments the analog was added 10 minutes before ALA.

In the upper half of Table 2 (tubes 1 to 4) are presented the inhibitory effects of δ-oximinolevulinic acid when it is added simultaneously with ALA in the same concentrations and ratios as in Table 1. The inhibition is the same as it is when the analog is added 10 minutes before ALA. In the lower half of Table 2 (tubes 5 to 8) are the data obtained by using a constant concentration of δ-oximinolevulinic acid ($7.5 \times 10^{-4}M$) with varying concentrations of ALA added 10 minutes after the analog. It is seen from both tables that increasing concentrations of ALA overcome the inhibition of δ-oximinolevulinic acid, thus demonstrating the competitive nature of the inhibition. Also it is seen in both tables that at various concentrations of ALA and δ-oximinolevulinic acid the inhibition is relatively constant for a given ratio of analog to substrate.

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A Genetic Constitution Frustrating the Sexual Drive in Drosophila paulistorum

Abstract. Hybrids obtained in the laboratory between two subspecies of *Drosophila paulistorum* possess a genetic constitution which is discordant enough so that the hybrid females repel the courtship of all males, and will mate with none. The hybrid males will court and will be rejected by almost all females, including their own hybrid siblings.

It has been shown (*1*) that the species *Drosophila paulistorum* actually represents a cluster of six subspecies, and that reproductive isolation of various sorts is being evolved between these incipient species, now in *statu nascendi*. Crosses between three (Centro-American, Amazonian, Andean-South Brazilian) of the six subspecies result in the production of fertile female and sterile male hybrids. The cause of the male sterility has been investigated (*2*) and found to depend upon the genotype of the mother involved. Any female which carries any mixture of the chromosomes of different subspecies deposits eggs giving rise to sterile male zygotes and to fertile female ones. (Intersubspecific insemination is accomplished more quickly here, and hybrid females can be tested for fertility by etherizing the females and immediately placing them with mature, unetherized males. The males will then approach and will often mount the females while they are still partly anesthetized. Subsequently, the females always produce offspring.) The male sterility is independent of the genotype of the male parents and the genotype of the sons themselves.

The mode of action of a reproductive isolating mechanism such as this seems to be unprecedented in genetic literature, but the same species-complex has evolved still another extraordinary isolating device: intersubspecific hybrids have been obtained by crossing Amazonian males with Andean-South Brazilian females. Most crosses between these two subspecies fail because of the powerful sexual isolation barrier. However, after repeated and lengthy attempts, viable male and female hybrids were obtained. It should be emphasized that these were normal males and normal females as far as the external and internal anatomy were concerned. Yet the genic endowments contributed by the parents of these hybrids are so discordant, that the hybrids are virtually unable to perform, successfully. the mating rituals that are normal in this species.

A study of the behavior of living flies under a microscope in special observation chambers showed that the hybrid females (25 have been observed and dissected so far) will not accept any males which court them, regardless of how vigorous or persistent the courtship is. They have been observed to reject consistently the males of both parental subspecies, as well as their own hybrid males. They accomplish this by assuming the posture of rejection of the courtship which is characteristic of *D. paulistorum*: the female lowers her head and elevates the tip of her abdomen so that the vaginal orifice is inaccessible to an approaching male. Only twice have Andean-South Brazilian males been seen to rush in so quickly that they succeeded in mounting the hybrid females; however, in one case, it took the female 2 minutes to repel the male by shaking violently from side to side, and, in the second instance, it took only 1 minute and 47 seconds. [Copulation normally takes an average of 17 minutes and 12 seconds in this species (*3*).] Furthermore, dissection of the female reproductive tracts involved, in physiological saline, showed that no sperm was transferred to the females in these two instances.

The hybrid males (19 have been observed and dissected so far) are of less interest in this respect, because they are completely sterile. Even so, they are rarely successful in courting females, and they have been placed and observed with mature females of both parental subspecies, as well as with their own hybrid females. These males have been observed in a total of only seven copulae, whereas a normal *D. paulistorum* male will begin courting again immediately after dismounting one female, and may inseminate several females per day.

It is here suggested that the disharmonies in the sexual behavior of the hybrid females may serve as a very efficient isolating mechanism between the incipient species. It is unfortunately not known whether the Amazonian and the Andean-South Brazilian subspecies occur anywhere sympatrically, but such sympatric occurrence has been recorded for other pairs of *D. paulistorum* subspecies (*1*). If such sympatric occurrence does exist, the possibility of hybridization cannot be excluded. However, the hybrid females, though potentially fertile, in the sense that their ovaries may be full of normal and mature eggs, would probably never mate. This would make the appearance of backcross progenies impossible.

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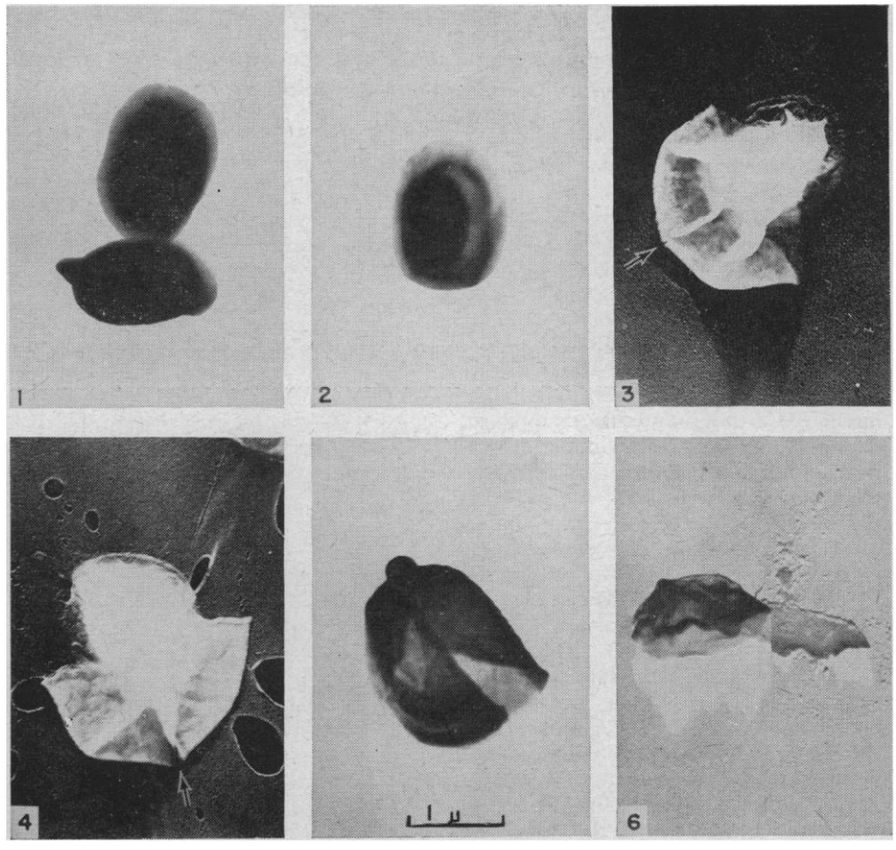

Figs. 1–6. Electron micrographs of *Bacillus megaterium*, taken with an R.C.A. electron microscope (EMU-2A) at an original magnification of 7400. Figures 3 and 4 are positive images; the others are negative. Fig. 1. Resting spores. Fig. 2. Spores incubated for 10 minutes—germination. Figs. 3 and 4. Spores incubated for 50 minutes—emergence of new cell and some elongation. The arrows indicate the split in the spore coat. Fig. 5. Empty spore case of an emerged cell, 60 minutes. Fig. 6. Empty spore case of a resting spore, obtained by grinding with glass beads. (× 12,500)

Spore Germination and Emergence of Bacillus megaterium

Abstract. Bacillus megaterium spores have a characteristic polar knob and equatorial ridge, or groove. During germination, the spore case appears to split along this ridge, and thus allows the new cell to emerge. Mechanically ground spores also split along this ridge, one part of the spore case being hinged to the other, the ridge being evident along a free edge. The equatorial ridge appears to be an area of susceptibility to mechanical pressures and, perhaps, in normal germination, to enzymic action as well.

We are primarily interested in the initial stages (*1*) of spore germination, when the spore loses its heat resistance, becomes stainable, and begins to consume oxygen. The study of later stages such as emergence may, however, help to illuminate early changes in spore germination. Electron micrographs of resting and germinating spores of *Bacillus megaterium* were prepared. The spores were germinated at 30°C in a medium buffered with 0.05*M* phosphate at *p*H 6.9 and containing 0.5 percent peptone, 0.02 percent yeast extract, and 0.025*M* glucose. Respiration in this medium has been described by Mandels *et al.* (*2*). The resting spores are opaque to electrons (Fig. 1). After 10 minutes in the germination medium, the spores become somewhat swollen, and dense material moves toward the periphery (Fig. 2). The spore case splits after 50 minutes of incubation, and a new cell protrudes from the case and begins to elongate (Figs. 3, 4).

The spore very often has what appears to be a polar knob and a ridge or groove circumscribing the major equator of the spore case. During the transition from spore to vegetative cell the spore case is often freed of the emerging cell (not adherent as in Figs. 3 and 4). The polar knob and equatorial ridge or groove are particularly pronounced in the discarded spore case shown in Fig. 5. We conceive of the equatorial ridge, or groove, as a line of weakness in the spore case more susceptible than the rest of the case both to physical and to enzymic attack. It is along this ridge, weakened perhaps by enzymic action (*3*) or by mechanical pressure of the swelling spore, that the case splits and allows the new cell to emerge. The cracks indicated by arrows in Figs. 3 and 4 support this idea. Furthermore, spores ground with glass beads show the same sort of split, one part of this physically damaged coat being hinged to the other with the ridgelike appearance evident along a free edge (Fig. 6) (*4*).

Note added in proof. Since we submitted this report, P. C. Fitz-James and I. E. Young have published electron micrographs which also show an equatorial ridge in the outer coat of *B. megaterium* spores (*5*).

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