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26 March 1965

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Desert Locusts: Sexual Maturation Delayed by Feeding on Senescent Vegetation

Abstract. *A diet of senescent* Brassica *spp. delayed sexual maturation in the desert locust. The senescent leaves were shown to be short of gibberellins, and a dietary supplement of gibberellin A_3 (1 microgram per locust per day) restored the rate of maturation to that found in animals feeding on green leaves. An external application of eugenol had a similar effect. The sexual immaturity of desert locusts during the dry season may result from the senescent condition of their desert food plants.*

The time desert locusts (*Schistocerca gregaria* Forskål) take to mature varies considerably. Under favorable conditions both in the laboratory and in the field no more than 3 or 4 weeks may elapse between fledging (imaginal moult) and egg laying. At the other extreme, no eggs may be laid for as long as 9 months after fledging, while delays of 2 to 5 months are quite common. During this period the adults retain their immature color and the ovaries show no signs of vitellogenesis.

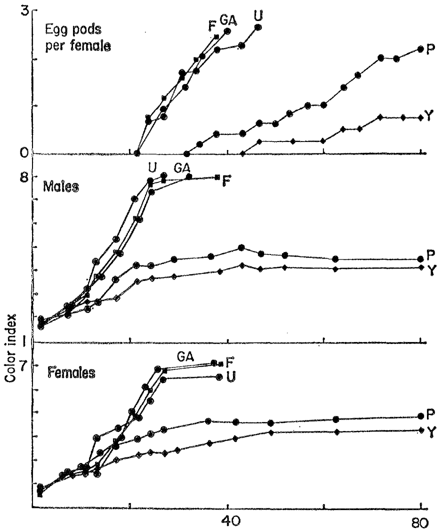

Fig. 1. Egg laying and color change in groups of desert locusts fed on *Brassica* spp. The color index changes from 1 at fledging to 7 or 8 at sexual maturity (*1*). *F*, fed on fresh green leaves; *Y*, fed on yellow, senescent leaves; *GA*, fed as *Y*, but diet supplemented with 1 μg of gibberellin A_3 per day from fledging; *P*, fed as *Y*, but diet supplemented with casein freely available to the locusts; *U*, fed as *Y*, but treated externally with 1 μl of eugenol on the 7th day after fledging.

Our recent studies (*1*) have emphasized the importance of plant monoterpenoids, such as eugenol, in triggering the onset of reproduction, and the importance of a plant hormone, the modified diterpene gibberellin, in hastening ecdysis (*2*). We have therefore investigated what part the physiological state of vegetation might play in controlling the time of maturation. Desert locust swarms frequently spend a large part of their lives in arid zones where the bush is green only for brief periods. During the dry season, when they do not breed, the locusts feed on sere and yellow vegetation. Only after the rains do they have fresh green leaves to eat. We have fed groups of desert locusts, starting about a week before the imaginal moult, on fresh green leaves of *Brassica* spp. (savoy cabbage, kale, and Brussels sprouts) and on senescent leaves of the same species (Fig. 1). Locusts fed the senescent vegetation changed color more slowly and started laying eggs several weeks later than those fed green vegetation.

Evidence is accumulating that aging of leaves is associated with a declining level of one or more of the natural plant hormones, auxins, gibberellins, or kinins (*3, 4*). Both yellowing and senescence of attached leaves can be retarded by supplying the appropriate hormones, and the hormonal requirements for this retardation appear to be species specific (*4, 5*). This has led to the proposal that senescence of leaves is regulated by their endogenous hormone levels (*4*). Senescence in all species of *Brassica* so far investigated can be retarded by gibberellin A_3. Hence the old leaves may well contain a relatively low level of gibberellin. A measure of the deficiency of gibberellins in some species of *Brassica* is shown in Fig. 2. It is based on the retention of chlorophyll in excised leaf discs after 2 or 3 days' incubation in darkness on filter papers moistened with either distilled water or gibberellin A_3 solution (25 or 50 μg/liter). Chlorophyll was extracted from the discs in hot 80-percent ethanol, and the optical density of the extract was measured at 665 mμ, the absorption maximum for chlorophyll *a*. Results are expressed as percentage retention of chlorophyll compared with the initial values. Because exogenous application of gibberellin A_3 delays yellowing, it seems likely that senescent *Brassica* leaves are naturally deficient in gibberellins.

When the diet of yellow leaves was supplemented with casein [an adequate protein source for locusts reared on a synthetic diet (*6*)] egg laying started earlier than it did in locusts feeding on yellow leaves alone, but the color change was no more than marginally

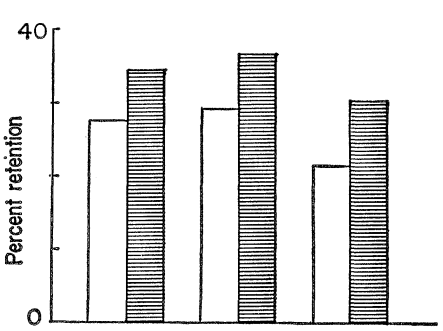

Fig. 2. Retardation of leaf senescence by gibberellin A_3 in three species of *Brassica*, measured by percentage of retention of chlorophyll in treated (cross-hatched blocks) and control (white blocks) leaf discs. Left, kale; center, Brussels sprouts; right, savoy cabbage.

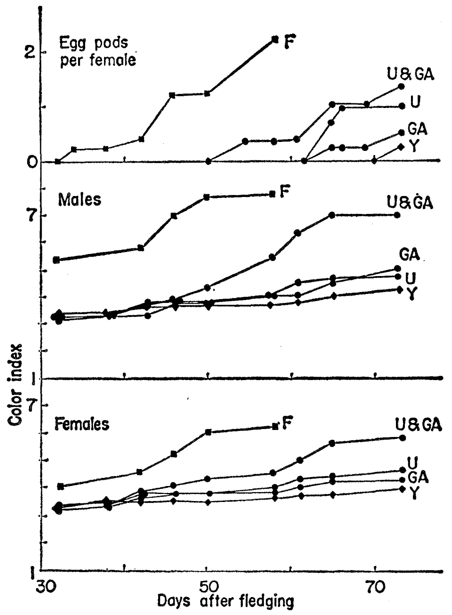

Fig. 3. Egg laying and color change in groups of desert locusts fed for 38 days after fledging on yellow, senescent leaves of *Brassica* spp. and then treated as described in Fig. 1. Records for locusts from the same batch but fed on green leaves are also shown (*F*).

affected. Locusts treated with a dietary supplement of gibberellin A_3 (about 1 μg per locust per day), administered with a diet of yellow leaves from the time of fledging, matured at the same rate as those fed on green vegetation; but when gibberellin A_3 was given to locusts fed on green leaves, it had no noticeable effect, or even retarded maturation. Locusts fed on yellow vegetation and receiving an external dose of 1 μl of eugenol (applied as a microdrop on the ventral surface of the thorax), 7 days after fledging, also showed the color change of approaching maturity as early as those fed on green leaves, and they started laying eggs on the same day. Unlike gibberellin A_3, eugenol hastens both the color change and the time of oviposition in animals fed a green diet (*1*). In other words, gibberellin A_3 only hastens maturity in animals fed the senescent vegetation presumed to be low in natural gibberellin, while eugenol is able to trigger the onset of reproductive capacity whether the locusts are feeding on green or on senescent vegetation.

In further experiments we have fed animals for 5 weeks after fledging on senescent *Brassica* and then either supplemented the diet with gibberellin A_3 (about 1 μg per locust per day) or applied a single external dose of eugenol, or the two in combination. After about 2 weeks the treated animals began to change color, and a week later oviposition started (Fig. 3). Under these conditions the two substances seem to be synergistic.

Marshall (*7*), writing of the breeding seasons of vertebrates in general and of birds in particular, states: "If animals that populated the equatorial regions, or special areas (e.g. arid regions) often far away from the equator, did not, in fact, adopt diverse regulatory and 'timing' devices they could not have survived. . . . They abandoned their traditional response to photostimulation (those that already possessed such a response) and came to obey more appropriate stimuli that would ensure that their young would be produced at the period most propitious for their survival." What is true of vertebrates is true of organisms generally in this context and, in particular, is true of locusts.

We would suggest that a diet low in gibberellin and essential oils, such as the desert locusts have in the dry season when they are feeding on old and withered vegetation, delays the attainment of sexual maturity, and delays the color changes which accompany it. At the onset of the rains, bud-burst in the aromatic desert shrubs provides a trigger, in the form of vegetation rich in gibberellin and eugenol and other monoterpenoids, which suffices to initiate sexual maturation and subsequent breeding (*1*). In this way breeding of the desert locust is geared to the rains.

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29 April 1965

Roots as Organs of Assimilation of Sulfate

Abstract. *Roots of the field pea (*Pisum arvense *L.) can reduce a small proportion of the sulfate that they absorb from the external medium. Some of this reduced sulphur is transported to the shoot as methionine and, to a lesser extent, as cysteine and glutathione.*

Remarkably little is known about the metabolism of sulfate in the higher plant. In certain species free sulfate is reported to be the only form of sulfur transported from roots (*1*). In others, organic compounds of sulfur are regular constituents of bleeding sap or fluids extracted from the xylem (*2, 3*), and this has been claimed as evidence that roots may act as important sites for metabolism of sulfate (*2*). However, in the absence of proof of synthesis in the roots, it is not clear whether the appearance of organic sulfur in the xylem really reflects sulfate assimilation, or whether it signifies a breakdown of root protein or even a circulation of substances originally synthesized in the shoot system.

There is general agreement that in all plants photosynthesizing leaves function as the main centers for reduction of sulfate (*4*). Nevertheless, it is also evident that nonphotosynthetic tissues may incorporate sulfate-sulfur into organic compounds. The activity of roots in this respect is proved beyond doubt in cultures of excised roots where, with the exception of traces of the sulfur-containing B vitamins, all sulfur is normally supplied in the form of sulfate. In excised roots, incorporation of sulfate-sulfur into sulfur-containing amino acids has been recorded by using radioactive sulfate (*5*). The experiment described here provides evidence that roots of the field pea (*Pisum arvense* L.) contain an active system for reducing incoming sulfate and that certain of the products of assimilation become available for transport to the shoot.

The roots of sulfur-sufficient, nodulated plants were supplied with radioactive sulfate, and at specified intervals afterward the shoots of 20 plants were removed and bleeding sap collected from the root stumps (*6*). The distribution of S^{35} in the sap and in the water-soluble and protein fractions of the shoot and root was examined for each harvest of plants (*7*).

Free sulfate accounted for more than 90 percent of the radioactivity of all