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14. The samples of livers for electron microscopic studies were fixed in cold 1 percent OsO_4 in *s*-collidine buffer, pH 7.4, and embedded in methacrylate. The sections were stained by the Karnofsky lead method A. More intensive staining of the crystalloid is obtained by Lawn's $KMnO_4$ method.
15 Investigation was supported by PHS grants CA-05310 and CA-03544 and by an Advanced Clinical Fellowship to Z.H. from the American Cancer Society. Azaserine was supplied by Cancer Chemotherapy National Service Center. Hepatomas were supplied by Dr. M. Rechcigl.

20 July 1964

Catnip: Its Raison d'Être

Abstract. Catnip (nepetalactone) is closely related chemically to certain cyclopentanoid monoterpenes recently isolated from insects, and it shares with some of these terpenes an ability to repel insects. It is suggested that the adaptive function of catnip is to protect the plants that produce it against phytophagous insects.

Catnip (nepetalactone, Fig. 1, IV), a cyclopentanoid monoterpene produced by certain plants of the mint family, has always been the source of much interest because of its peculiar ability to excite cats and their feline relatives. The true adaptive significance of this substance, like that of so many others extracted for one purpose or another from plants, has remained a mystery. Surely, a mint plant derives no benefit from an ability to stimulate cats!

In recent years, certain compounds chemically allied to catnip have been isolated from insects (Fig. 1). Two of these (I, II) stem from ants (*1*); the other (III) is known from both ants (dolichodial) (*2*) and a walkingstick (anisomorphal) (*3*). There can be no doubt, at least as regards iridomyrmecin and anisomorphal, that these substances have a defensive role. Iridomyrmecin has considerable insecticidal activity (*4*), and anisomorphal, which is ejected by the walkingstick as a spray against predators, has been shown to be strongly repellent to ants, beetles, spiders, birds, and even ourselves (*5*). Also of interest is the fact that two cyclopentanoid monoterpenes (V, VI) other than nepetalactone, but both with catnip-like activity, have recently been identified from a Japanese plant; one of these (V) is identical to iridomyrmecin (see *6*).

The preceding data strongly suggest that catnip may itself be a defensive substance, perhaps protecting the plant against phytophagous insects. This possibility was investigated by a series of simple experiments (*7*).

One of these consisted in observing the response of a variety of insects to the vapors emanating from the tip of a fine capillary tube filled with pure liquid nepetalactone, and pointed to their bodies from a few millimeters away. The insects tested (Table 1) were a mixed assortment that had come to rest at night on an illuminated surface. The majority (part *A*) showed a distinct avoidance response, which varied somewhat with the particular species. The caddis-flies flew away. The alleculid beetles fell to the ground (as do many beetles when disturbed). The remainder simply turned away from the capillary and walked off. As they escaped, complete control could be exercised over the course of their locomotion by maintaining the capillary at close range, pointed at them from various directions; their every move was in distinct avoidance of the vapors. Capillaries filled with water had no comparable effect.

Only relatively few species remained undisturbed by catnip (part *B*). In the pentatomids and reduviids, this indifference might be taken to reflect an insensitivity to catnip, since these same insects showed strong avoidance of two other compounds that were tested (methacrylic acid, *p*-benzoquinone) (*8*). The chironomid, as well as the pyralid and arctiid moths, remained undisturbed even by these substances, indicating perhaps that under the conditions of the experiment (that is, while the insects are illuminated and at rest) they are generally unresponsive to noxious vapors.

Insects that responded to the vapors of nepetalactone also reacted characteristically to direct contact with the liquid. When mouthparts or antennae were touched with the wet tip of the capillary, they fled instantly and quickly, pausing occasionally to cleanse antennae with their front legs, or to wipe mouthparts against the substrate.

Additional tests were made with ants foraging along trails. The results were similar for the two species tested (*Solenopsis germinata, Monomorium pharaonis*). When a droplet of nepetalactone was placed on the trail, the ants stopped abruptly, building up in numbers around the droplet, but with not a single one venturing to within about 5 centimeters from it. When a circle was drawn around a group of ants with a glass rod dipped in nepetalactone, the ants similarly refused to cross the "line" and remained temporarily trapped within the circle. An ant (*S. germinata*) that was carrying a small live curculionid beetle, dropped its prey instantly when a droplet of nepetalactone was applied to the beetle, and began intensive cleansing activities. Other workers that made casual contact with this ant were themselves induced to cleanse.

Figure 2 illustrates yet another experiment, done with the ant *Pogonomyrmex badius,* an especially aggressive species. Two freshly killed cockroaches, one with a droplet of nepetalactone on its abdomen, the other an untreated control, were placed near the

Table 1. Species of insects exposed to vapors of nepetalactone. Families are numbered.

A. Repelled by catnip	

1. Fulgoridae
    Acanalonia sp.
	8. Staphylinidae
	    Two unidentified small species

2. Cercopidae
    Monecophora bicincta
	9. Scarabaeidae
	    Ataenius sp.

3. Formicidae
    Camponotus floridanus (winged queen)
	10. Chrysomelidae
	    Disonycha conjugata

4. Leptoceridae
    Leptocella sp.
    Oecetis inconspicua
	11. Tenebrionidae
	    Leichenum canaliculatum

5. Dytiscidae
    Coptotomus interrogatus
	12. Alleculidae
	    Hymenorus sp.

6. Lampyridae
    Photinus sp.
	13. Curculionidae
	    Derelomus sp.
	    Conotrachelus sp.
	    An unidentified small species

7. Cicindelidae *Cicindela trifasciata*	
B. Indifferent to catnip	

14. Pentatomidae
    Thyanta sp.
    Euschistus sp.
	16. Pyralidae
	    Paraponyx allionealis

15. Reduviidae
    Oncocephalus geniculatus
    Pnirontis sp.
	17. Arctiidae
	    Afrida ytadodes
	18. Chironomidae
	    Chironomus sp.

nest entrance of the ant colony. Within minutes the control was overwhelmed by a swarm of ants, while the treated specimen remained virtually ignored.

Catnip is evidently an insect repellent of considerable effectiveness, and its potential for serving in this capacity in the plants that make it appears demonstrable. Matatabilactone and actinidine (Fig. 1, V, VI) might similarly serve in defense. The precise way in which these terpenes exert their repellency in the living plant, and the extent to which they have successfully contributed to the restriction of phytophagy, will obviously need to be determined by experiments of an entirely different sort. Insects are extraordinarily diverse, and there is nothing surprising in the finding that some species are apparently insensitive to catnip. Limited effectiveness is the rule rather than the exception among so-called insect repellents. An exhaustive search might even have uncovered insects that are actually attracted to catnip, and one wonders how the particular species would respond that routinely feed on the plants that produce this or related terpenes.

There is nothing new in the idea that so-called "secondary plant substances," of which catnip is but one example, may function for protection against insects or other herbivores, or both. An excellent paper has recently appeared on the subject (9). Although many of these secondary substances have no counterparts in insects or other animals, the case of the present terpenes is by no means the only one known of defensive compounds that have arisen both in insects and plants. Citronellal, perhaps the most familiar of insect repellents of plant origin, has recently been identified as a product of the mandibular glands of an ant (10). Cyanogenesis, generally believed to be restricted to plants, occurs also in certain moths and millipedes (11). trans-2-Hexenal, a compound produced as a defensive spray by several insects (12), appears to be widespread in plants (13). Additional examples of such parallel evolution could be cited, and there are undoubtedly many cases yet to be discovered. There is always the possibility that secondary plant substances might be of potential practical use as insect repellents. In view of the proven activity of nepetalactone, iridomyrmecin, and anisomorphal, one would think that the cyclopentanoid monoterpenes might themselves be investigated profitably with this end in mind.

Also in need of explanation is the

interesting although entirely different matter of the response of cats to these terpenes. Whether the Felidae actually produce such terpenes themselves, employing them perhaps as pheromones for the regulation of some of their behavior, is a possibility worth exploring.

Terpenes are intriguing compounds, widely distributed, and have functions that are yet to be explained in many species. In plants and insects, similar terpenes appear to have evolved in fulfillment of similar defensive needs. It is certainly conceivable that the same com-
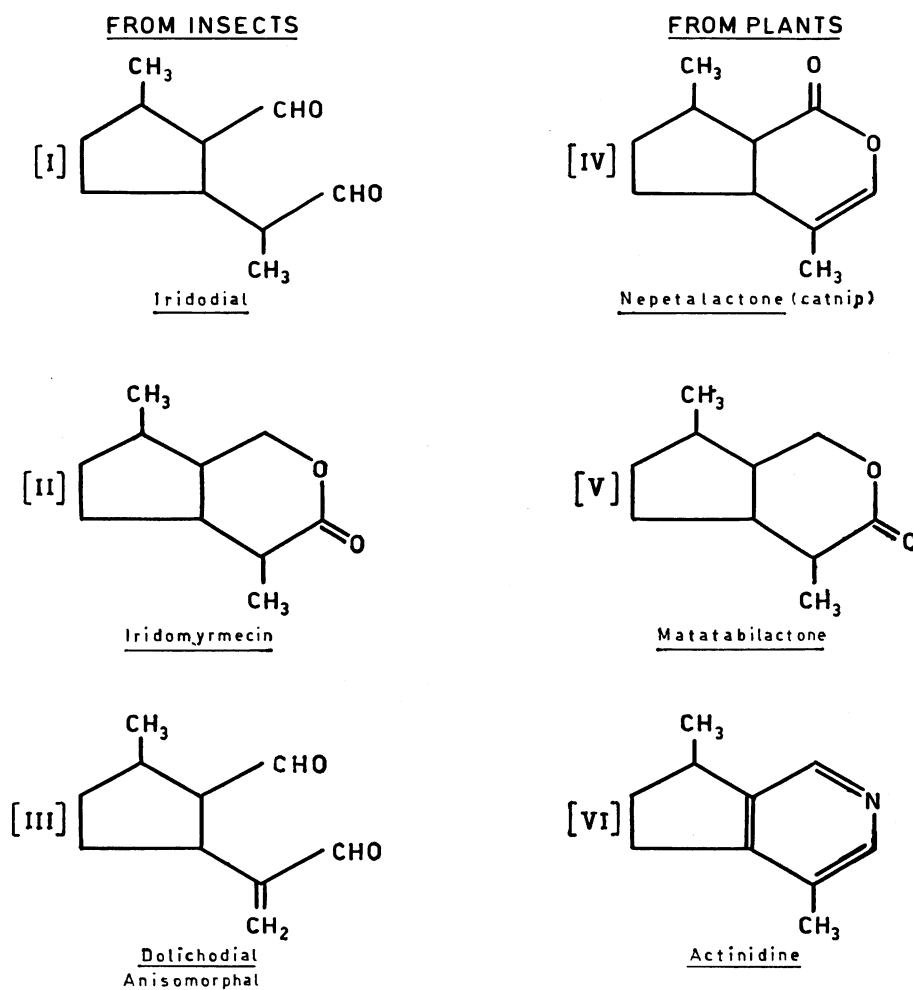

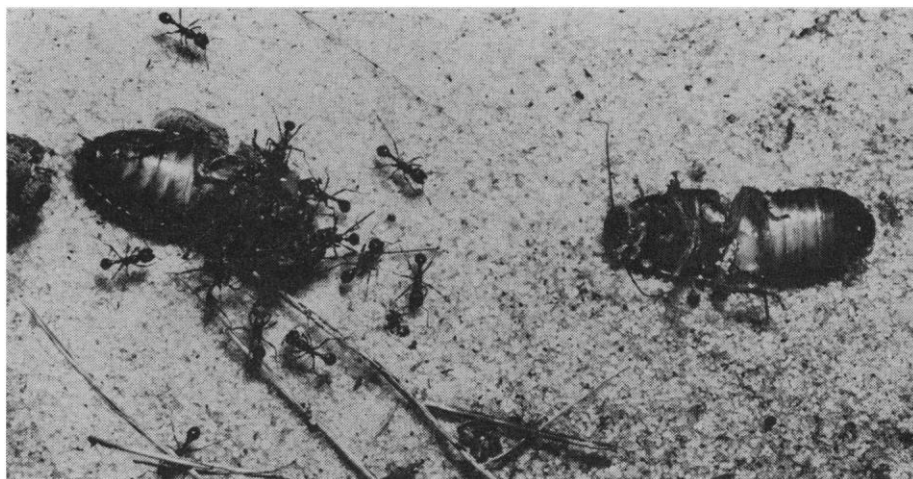Fig. 1. Closely similar cyclopentanoid monoterpenes from insects and plants.

Fig. 2. Two freshly killed cockroaches exposed to attack by ants (*Pogonomyrmex badius*). The one on the right has been treated with a droplet of nepetalactone and is being ignored.

pounds might be present in yet a third group of organisms (perhaps in cats), but for entirely different adaptive purposes.

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30 October 1964 ∎

Saturation Deficit of the Mesophyll Evaporating Surfaces in a Desert Halophyte

Abstract. *The tensions developed in the internal evaporating leaf surfaces were estimated for a desert halophyte,* Reaumuria hirtella, *growing in its natural habitat. The method was based on the assumption that at zero transpiration the vapor pressure of the inner parts of the leaf was in equilibrium with that of the atmosphere, provided that stomatal resistance was constant during the measuring period. This could be ensured, since the measuring system controlled both the concentration of carbon dioxide and the atmospheric humidity, while measuring photosynthesis and transpiration simultaneously. Tension values of 180, 240, and 320 bars were recorded for three different* Reaumuria hirtella *plants.*

In general discussions of transpiration, the evaporating surfaces of the leaf cell walls are usually considered to be saturated. During transpiration, saturation deficits may develop (*1*), and evidence for appreciable deficits has been put forward (*2–4*). In the course of measuring transpiration, T, and photosynthesis in a number of desert plants, we developed a method of estimating the magnitude of this deficit. A halophyte, *Reaumuria hirtella*, (J. et Sp.) (Tamaricaceae), growing in natural desert conditions, was chosen for detailed studies. Measured values supported our conclusion that large saturation deficits can develop.

The principle of our method was to estimate the equilibrium vapor pressure of the external atmosphere at which the net flux of water vapor between the plant shoot and the sur-

rounding air was zero (see 5). Photosynthesis and transpiration were measured simultaneously throughout the day on intact plants in a natural hillslope community at the Desert Research Farm of the Botany Department at Avdat, in the Negev desert, Israel. Climatological data for this site are given in (*6*). The measuring system, operating from a mobile laboratory, has been described (*7*). This "null-point compensating system" operates on the principle of measuring rates of compensation for the changes in atmospheric composition (CO_2 and H_2O) produced in a transparent chamber by the activity of the enclosed shoot, while maintaining this composition constant.

Water vapor and CO_2 were maintained at the desired concentrations by continuous monitoring with an electric

hygrometer and a recording infrared gas analyzer. Compensation for changes in the concentration of CO_2 was made by addition of CO_2-enriched air, while changes in humidity were counteracted by controlling the rate of drying of the circulating air. Leaf temperature was measured to $\pm 0.3°C$, by inserting a thermocouple into the leaf, along its axis. Two leaves were thus measured in each plant and the mean was taken to represent that of the entire leaf population on the shoot. This appears reasonable, since the plants were small and open and were subjected to constant turbulence. Air temperature was measured by shaded thermocouples in the air outlet from the chamber.

Under constant turbulent conditions and uniform stomatal resistance, transpiration is a linear function of the gradient of water-vapor pressure between the evaporating surface and the atmosphere. If the evaporating surface is water-saturated and leaf and air temperature are equal, $T = 0$ when the vapor pressure gradient is zero, and the plotted curve of T against gradient passes through the origin. With an unsaturated evaporating surface, $T = 0$ at an atmospheric water vapor pressure less than the saturation value. For technical reasons, the equilibrium vapor pressure at $T = 0$ could not be attained in the field. This value was found by regression analysis.

In order that leaf and air temperatures would be more nearly equal, the radiation load on the plant was reduced to half by shading it with wire mesh. Transpiration values were recorded for three successive 10-minute intervals at each level of atmospheric humidity. Values used in the subsequent analysis were recorded between 10 a.m. and 3 p.m., during which period the light intensity at the plant was 5.5 ± 0.5 lumen/cm² and air temperature varied only by $\pm 2°C$. The concentration of CO_2 was held constant at 320 ± 5 parts per million. Under these conditions, uniform rates of photosynthesis were taken as an indication of a constant stomatal resistance. In earlier studies with this and other species, we found quite a close relationship between change in stomatal resistance and changes in CO_2 uptake, at constant, saturating light intensities. Any transpiration values recorded during marked fluctuations in rates of photosynthesis were not included.

Linear regressions of T on atmo-