This histological study revealed some of the minute details of the vector-virus-host relationship of the sweetpotato yellow dwarf virosis and demonstrated the precision of the surgical operation performed by this insect in its normal feeding operations. Other unreported evidence supports the hypothesis, which definitely applies to all whiteflies and aphids, that whenever a stylet is less than 3 μ in diameter penetration will be intercellular, because its physical size makes it too limber for direct penetration.

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5 December 1960

Chemical Nature of Antheridogen-A, a Specific Inducer of the Male Sex Organ in Certain Fern Species

Abstract. Antheridogen-A has been shown to be a complex carboxylic acid. The carboxyl function is necessary for inductor activity since this activity disappears on esterification and reappears after hydrolysis of the ester.

Recently a substance was isolated from culture filtrates of the bracken fern *Pteridium aquilinum* which induces the formation of antheridia in gametophytes of the sensitive fern *Onoclea sensibilis* at a concentration of less than 1 part in 10,000,000,000 (10^{-4} μg/ml) (*1*). The present report describes what is known of the chemical nature of this antheridium-inducing factor, for which the trivial name antheridogen-A is proposed.

Since the material is present at a concentration of the order of 1 μg or less per liter of culture filtrate, it has not yet been possible to produce enough material to analyze directly. The chemical constitution of antheridogen-A was studied indirectly by observing the effects of chemical manipulations on the purified concentrate prepared as previously described (*1*). Antheridogen-A dialyzes readily through cellophane membranes, so it appears to have a low molecular weight. It is relatively stable in acid solution but is readily inactivated at pH above 7. It is also readily inactivated by oxidizing agents. During the course of isolation the compound behaved as a weak acid.

Antheridogen-A was found to have a distribution coefficient close to unity when partitioned between normal butanol and 5 percent ammonium acetate buffer of pH 6.65 (*1*). When partitioned between ethyl acetate, isoamyl acetate, tertiary amyl alcohol, or peroxide-free diethyl ether and McIlvaine buffers at various pH's, antheridogen-A distributed in a pattern which suggested it to have a $p\mathrm{K}_a$ of about 5.0.

The compound appears to be free of phosphorus. When an amount of material equivalent to 10 μg was tested by the method of Hanes and Isherwood (*2*) no color was produced. All phosphate esters tested at this concentration gave strong positive tests. Ninhydrin tests on filter paper chromatograms of the material were consistently negative.

Treatment of a dry ethereal solution of antheridogen-A with excess diazomethane completely inactivated the material. When the methyl ester was refluxed with 5*N* hydrochloric acid for 3 hours, biological activity was restored.

Examination of the infrared absorption spectrum of antheridogen-A preparations revealed a well-defined maximum at 1700 cm^{-1} which is consistent with a carboxyl functional group and perhaps suggests that the molecule contains an unsaturated carbon-carbon bond in the vicinity of the carboxyl group (*3*). The ease with which the inductor activity is lost by oxidation also suggests unsaturation. On the other hand, when the compound was treated with bromine in carbon tetrachloride solution for 4 hours, no loss of activity occurred.

The preceding indirect evidence indicates that antheridogen-A is a complex carboxylic acid. Furthermore, the carboxyl function is necessary for biological activity since this activity disappears on esterification and reappears after hydrolysis of the ester. A large number of naturally occurring carboxylic acids were tested for antheridium-inducing activity on *O. sensibilis.* None were found to have activity. It is of interest to note that certain long-chain aliphatic fatty acids produce a threefold increase in the potency of antheridogen-A preparations. This relationship may have some bearing on the mode of action of antheridogen-A and will be further investigated (*4*).

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30 September 1960

Maintenance of Avoidance Behavior under Temporally Defined Contingencies

Abstract. Operant behavior of rats was maintained at moderate frequencies by a temporally defined shock avoidance schedule. Progressive reductions in one of the temporal parameters of this schedule—the portion during which behavior may have consequences—yield an orderly response rate function which first rises to a maximum and then declines gradually to extinction.

A system for classifying schedules of reinforcement for operant behavior in terms of temporally defined parameters has recently been proposed by Schoenfeld, Cumming, and Hearst (*1*). This system, which provides a common dimensional framework for the specification of reinforcement schedules generally, defines two basic variables: t^D and t^Δ, time periods during which, respectively, reinforcement may be given and reinforcement is never given.

Conventionally, t^D and t^Δ are held constant and are alternated, and only the first response in t^D is reinforced. Some of the parameters of that system have been experimentally explored in a number of recent studies (*2*) in which positive reinforcement procedures were employed, but to date their effects have not been observed in avoidance conditioning contexts, where the occurrence of a given response prevents the presentation of an aversive stimulus. In the procedure adopted in the experiment reported here, an avoidance schedule lacking a warning stimulus (*3*) was cast in t^D, t^Δ terms, and the effects of systematically varying one of the temporally defined parameters were studied.

Four adult male hooded rats, all without prior experimental history, were exposed for 30 minutes daily to an avoidance conditioning schedule. The sound-resistant chamber in which all the animals worked was equipped with a lever and a grid floor through which electric shock (0.3 ma) could be delivered to the rat's feet. Depression of the lever activated counters and a cumulative recorder. Relays and timers established a temporally defined avoidance schedule composed of alternating t^Δ and t^D time periods, one following the other in such a way that a complete cycle $t^{\Delta} + t^{D} = T = k = 30$ seconds. In t^{D}, the first lever press made by the rat enabled it to avoid shock; in t^{Δ}, lever presses were without effect. Therefore, at the beginning of each $t^{\Delta} + t^{D}$ cycle, shock occurred unless a response had been made in the immediately preceding t^{D} period. Lever presses occurring during shock terminated the shock, but except for this escape contingency, responses in t^{Δ} portions of the 30-second cycle had no effect on the schedule.

The animals were first exposed to 22 consecutive sessions at $t^{\Delta} + t^{D} = 30$ seconds, $t^{\Delta} = 0$ to allow their response rates to stabilize; t^{D} was then progressively reduced over the course of the experiment from 30 seconds to 15, 7.5, 3.75, 1.8, and 0.4 seconds. The animals were given 13 sessions of exposure at each of these t^{D} values except 0.4 second. At $t^{D} = 0.4$ second, extinction of the avoidance response appeared to be complete by the eighth session, and the experiment was therefore terminated at that time.

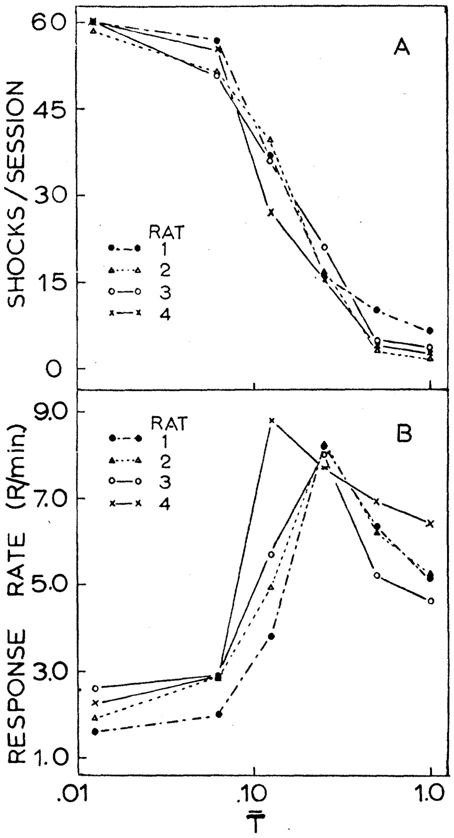

Fig. 1. (*A*) Number of shocks received per session; (*B*) lever-press response rates for four rats as a function of the temporal schedule parameter $\bar{T}$. $\bar{T}$ is defined as $t^{D}/(t^{D} + t^{\Delta})$ and represents here the relative portion of a 30-second shock-shock interval in the period during which the first response resulted in avoidance of the next shock due.

Figure 1, *A* and *B*, illustrates for each of the four rats the number of shocks received per session and the rate of lever pressing as a function of progressive reduction in t^{D}. The data are averages for the final 3 days on each schedule and have been plotted against the variable $\bar{T} = t^{D}/(t^{D} + t^{\Delta})$, the relative proportion of the complete cycle for the t^{D} period.

The response rate functions are similar for each animal; rats 1, 2, and 3 exhibit a maximum of between 8.0 and 9.0 responses per minute at $t^{D} = 7.5$ seconds ($\bar{T} = 0.25$), and rat 4 a maximum at $t^{D} = 3.75$ seconds ($\bar{T} = 0.12$). The rates then decline over the remaining t^{D} values to the asymptotic shock escape rate of 2.0 responses per minute at the smallest t^{D}. The rate function is not related in a simple way to the number of shocks received, for, as Fig. 1*A* shows, this latter variable bears a constant inverse relationship to $\bar{T}$ throughout the range.

Cumulative records for one rat, obtained from the final session at each t^{D} value (except $t^{D} = 0.4$ second, the value at which there was no avoidance behavior), are shown in Fig. 2 (due to a recording failure a bit each session is omitted at $\bar{T} = 0.12$ and 0.06). The records reveal a roughness in over-all character and sections of negative acceleration terminated by the delivery of the aversive stimulus. Visual inspection of these records shows that little systematic change in the qualitative aspects of the rate occurs with changes in t^{D}.

These findings may be compared with recent data reported by Hearst (*4*) in which similar changes in the length of a t^{D} period within a constant cycle length of 30 seconds were made under conditions of positive food reinforcement. The rather similar rate functions that result in the two cases indicate that the effect of $\bar{T}$ reductions on response rate is a general one. The first decreases in $\bar{T}$ lead to a compensatory rate increase, but when t^{D} is decreased below the minimum interresponse time, responding cannot be sustained.

The procedure described here might be expected to lead to the formation of a temporal discrimination at $\bar{T}$ values at which a fairly large period of zero probability for reinforcement occurs and the average rate continues to be well sustained. The resolution of the cumulative records of Fig. 2, however, is not of sufficient detail to establish such a discrimination unequivocally; hence, a definitive answer must await a more refined within-cycle interresponse analysis of the behavior (*5*).

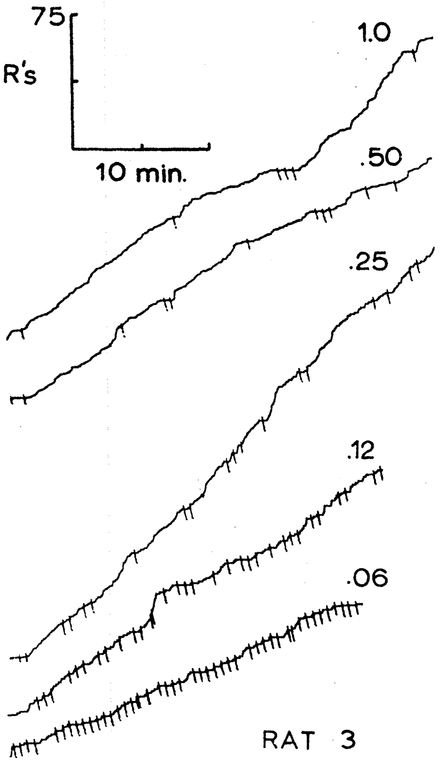

Fig. 2. Cumulative response records of lever pressing for one rat at each of six values of $\bar{T}$. Shocks are indicated by short oblique strokes, and each complete segment represents data from the final session at each $\bar{T}$ value.

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30 September 1960