of those in the isolated group would show comparatively little added development.

If the median of the groups is used as the dividing line for low- and high-ranking brain weights, the isolation-enrichment differences would be most apparent between the low-ranking brain weights from each group. This is so because although all animals in the enriched group, both low-ranking and high-ranking in terms of the genetic brain weight component, would show an upward shift as a result of their experience, the low-ranking animals in the isolated group would be more likely to be those which had received little or no benefit from the limited stimulation they had received. Those isolated animals that would have ranked low on the basis of the genetic component alone but had shown some additional environment-dependent development would be likely to move above the median displacing somewhat higher-ranking animals that had exhibited little response to the limited stimulation.

Two predictions can be made on the basis of this model. (i) The amount of intragroup variance among the isolated animals will exceed that among the enriched. Such a prediction results from the presence of a genetically determined developmental ceiling against which the enriched animals will be pushed, combined with the increased probability that enrichment will lift all animals from the base line. This comparison was performed by computing the coefficient of variation for each of the enriched and isolated groups. In eight of the nine studies the variance of each isolated group exceeded that of its enriched counterpart, and in the remaining study (group 80) they were equal.

The second prediction is based on the assumption that the two groups fall on opposite sides of the midpoint of the asymptotic curve. (ii) Since there exists both an effective ceiling to development and a base line from which such development commences, the variance distribution on either side of the median should be opposite for the two environmental groups. Within the enriched groups, the variance between the brain weights lying below the group median should exceed the variance of those above; the opposite should be the case for the isolated groups. This prediction was tested by using a Fisher exact probability test ($P < .05$).

We conclude that the data support the hypothesis that sensory stimuli act by inducing environment-dependent neural development in accordance with the model (Fig. 2). The nature of this stimulation has been the subject of a previous paper (*4*) wherein we argued that the basic mechanism is that of nonspecific activation of the cortex during arousal. In this regard, the rank-ordering effect decreased after 60 days of differential rearing. This may be explained if progressive habituation of the sensitization derived from arousal-inducing aspects of the environment occurs in the enriched animals, while spontaneous recovery of habituated sensitization occurs in the isolates as a result of the absence of such arousal-inducing (sensitizing) stimuli (*7*). The net effect of these alterations in the arousal threshold would be that the arousal potential of both types of living environment would become more similar to their respective inhabitants with increasing duration of exposure.

There remains, however, an additional consideration. While, in accordance with our prediction of sensitization during isolation, there was an inverse relationship between the magnitude of the rank-ordering effect and the duration of isolation, an overall enrichment-isolation difference was present at both 90 and 120 days (4.5 percent, $P < .01$, and 6.8 percent, $P < .001$, respectively). In groups 90 and 120, the forebrains of the enriched animals were consistently heavier than those of the isolates for all brain-weight ranks. Other reports have also indicated that, under some conditions, enrichment-isolation differences in cortex weight have been observed beyond 80 days of differential rearing (*6*, *8*). Thus, it appears likely that sensitization of the isolated animals does not fully compensate for the reduced stimulus levels, and that development stabilizes at a suboptimal level.

Finally, we suggest that environmental enrichment has been greatly overemphasized as the causative agent for enrichment-isolation brain changes. The degree of sensory deprivation suffered by the isolated animals is likely to be the critical factor, since the stringency of this condition determines the proportion of subjects retarded in their environment-dependent neural development.

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Disruption of Sex Pheromone Communication in a Nematode

Abstract. *Males of* Nippostrongylus brasiliensis, *an intestinal parasite of rodents, were maintained in an environment permeated with pheromone produced by females of the species. After the males were removed from that environment, their subsequent ability to orient to a gradient of the pheromone emanating from living females was greatly reduced for periods up to 2 hours. This phenomenon might serve as the basis for a new, selective antihelminthic technique in which the premating communication between males and females is disrupted.*

The first report concerning the use of chemical signals in premating communication between the sexes of a nematode was published in 1964 (*1*). Since that time, it has rapidly become apparent that sex pheromones are the primary communicative signals that bring together the sexes of many plant-parasitic, free-living, and animal-parasitic nematode species. We propose that a necessity for distance communication by pheromones prior to mating may constitute a "weak link" in the life cycle of certain pestiferous species, wherein man might manipulate the chemical signals to his advantage and to the disadvantage of the nematodes.

Entomologists have recognized for some time that pheromone communication is an essential component of the premating behavior of a number of pest insect species and are now advancing rapidly toward environmentally safe insect-control strategies based on manipulation

of the pests' communication systems (*2*). One of the most promising of such strategies being developed by entomologists, called "disruption of pheromone communication" involves the permeation of the pests' environment with a synthetic pheromone that is identical to the pheromone released by the living insects prior to mating (*3*). With the synthetic pheromone odor being everywhere in the environment, the sex that normally responds to the pheromone is rendered incapable of locating the pheromone-releasing sex. Considerable experimental evidence indicates that sensory adaptation and habituation of the insects to the omnipresent pheromone are major factors causing this communication disruption (*2, 4*). We now report that the disruption concept might be useful in pest-control systems directed against certain nematode species, including nematodes that are parasites of mammals. We have maintained males of *Nippostrongylus brasiliensis*, an intestinal parasite of rodents, in an environment permeated with female pheromone and have found that the males' subsequent ability to orient to a gradient of pheromone emanating from living females was greatly reduced for periods of at least 2 hours.

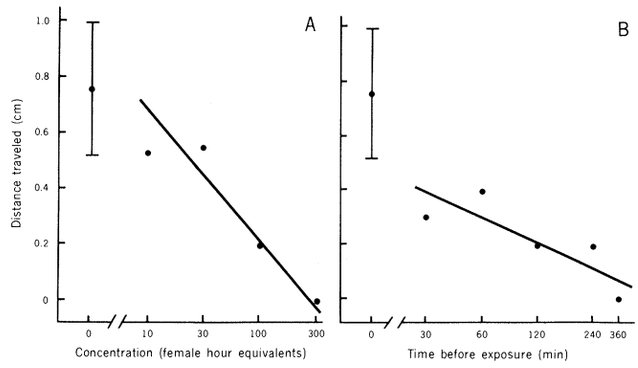

Fig. 1. Mean distance traveled toward a 35-female pheromone source by males of *Nippostrongylus brasiliensis* after they had been exposed for various intervals in Tyrode's solution containing various concentrations of the pheromone. Pheromone concentrations during prior exposure are stated as female hours per milliliter of solution. For example, a concentration of 100 was prepared by allowing 100 females to condition 2 ml for 2 hours. Vertical lines indicate the magnitude of the standard error for the mean movement of control (unexposed) males toward the female pheromone source. Each point represents the mean for 40 separate assays. (A) Males exposed for 4 hours to various concentrations of pheromone. A significant ($P \leq .05$) regression of distance traveled by the males (Y) as a function of the log of pheromone concentration of the prior exposure (X), was obtained, with the regression equation being $Y = -0.39X + 1.00$. (B) Males that had been exposed for various time intervals to a concentration of 100 female hours per milliliter. A significant regression of distance traveled by the males (Y) as a function of the log of prior exposure time in minutes (X) was obtained, with the regression equation being $Y = -0.29X + 0.82$.

Nippostrongylus brasiliensis was maintained by subcutaneous inoculation of white mice (*5*). Young adult nematodes were collected for experimentation at 5 days after infection, because pheromone production by females and pheromone responsiveness by males are maximal at that age (*6*). Standard female pheromone solutions were obtained on each day of experimentation by maintaining known numbers of females in 2 ml of Tyrode's solution (*7*) for 2 hours, after which the females were removed. Groups of 50 males were then placed in the solution for varying intervals of exposure, after which they were removed and placed singly in bioassay chambers to determine their responsiveness to pheromone gradients emanating from living females.

In each bioassay chamber (*8*), 35 females were confined, starting 6 hours prior to the assay, by a filter paper barrier at one end of a rectangular trough filled with 5 ml of Tyrode's solution. A single male nematode was placed in the center of the remainder of the chamber and his distance of travel either toward or away from the female pheromone source was recorded after an additional 2 hours.

Responses of males that had been exposed for 4 hours to various female pheromone concentrations, ranging from 10 to 300 female-hour equivalents, indicate varying degrees of inhibition of subsequent male responsiveness, with essentially no males responding to the pheromone gradient when they had been exposed to the highest concentration (Fig. 1A) (*9*). In a related experiment, with males that had been exposed to a single concentration (100 female-hour equivalents) for varying time periods ranging from 30 to 360 minutes, a similar inhibition of male responsiveness occurred when they were subsequently exposed to the female pheromone gradient (Fig. 1B). Finally, males were allowed various time intervals of "recovery" in clean Tyrode's solution between their exposure to 100 female-hour equivalents for 4 hours and their exposure to the pheromone gradient. Even with a 2-hour recovery period, the males only traveled a mean distance of 0.20 cm (± 0.12, standard error) toward the pheromone source, as compared with 0.75 cm (± 0.24, S.E.) for nematodes that had not received any exposure prior to treatment.

The results of these experiments clearly show that prior exposure in vitro of male *N. brasiliensis* to female pheromone inhibits their subsequent ability to orient to pheromone gradients and move toward females for periods up to 2 hours after the prior exposure. Considering that prior exposure to the nematode pheromones causes such a strong inhibition of male responsiveness, it seems reasonable that a continuous exposure would create at least as strong an inhibition while at the same time creating "confusion" in the communication system by destroying the coherent gradients or other characteristics of the pheromone message that normally guides males to individual pheromone-releasing females. Therefore, we propose that the permeation of the normal environment of certain nematode species with their synthetic pheromones, perhaps through the application or ingestion of slow-release pheromone formulations, might offer considerable potential as a selective anthelminthic technique.

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9. Males were rinsed in 10 ml of Tyrode's solution prior to their exposure to female pheromone gradients.
10. We thank L. B. Bjostad and C. C. Hanlon for their assistance with this research.

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