

main coronary artery and three from the distal midventricular portion of the artery). Two 5- μ m sections were cut from each block and stained with either hematoxylin and eosin or Verhoeff-Van Gieson stains. For each block, the percentage of the apparent lumen occupied by atherosclerotic lesion (stenosis) was recorded (Table 1). Consistent with the presence of atherosclerosis in all of the arteries except the thoracic aorta was the more extensive stenosis in the coronary artery of the vasectomized monkeys compared with the sham-vasectomized controls. From the gross appearance of the hearts, it seemed that the coronary artery atherosclerosis was more distally extended among the vasectomized animals. For this reason, both proximal and distal segments of the left anterior descending artery were examined. Both gross and microscopic observations showed a greater difference in the distal segment. The mean stenosis of the proximal segment of the left anterior descending coronary artery of the vasectomized animals was 69.8 ± 4.3 compared with 42.1 ± 16.2 for the sham-vasectomized group. This difference was not statistically significant ($P < .20$). However, the mean stenosis of the distal segment of the same artery in the vasectomized animals was 67.0 ± 4.9 , whereas in the sham-vasectomized group it was only 25.6 ± 11.4 , and this difference was statistically significant ($P < .02$).

In our experiments, the monkeys were fed a diet containing about twice as much cholesterol as that consumed by the average North American and the plasma cholesterol concentrations in these monkeys were generally around 500 to 600 mg/dl, in contrast to the usual concentrations of 220 to 250 mg/dl for North American adult human males. Although the findings are statistically significant, the data are based on two groups of only five animals each.

We suggest that the basis of the observed effects after vasectomy may be immunologic injury to the vascular endothelium, resulting in a rapid progression of the lesions. The data collected under the conditions of our study showed that vasectomy was associated with a marked exacerbation of diet-induced atherosclerosis in *M. fascicularis*.

NANCY J. ALEXANDER
*Reproductive Physiology,
Oregon Regional Primate Research
Center, Beaverton 97005*

THOMAS B. CLARKSON
*Arteriosclerosis Research Center,
Bowman Gray School of Medicine,
Wake Forest University,
Winston-Salem, North Carolina 27103*

SCIENCE, VOL. 201, 11 AUGUST 1978

References and Notes

1. H. V. Lamberson, Jr., and K. E. Fritz, *Arch. Pathol.* **98**, 9 (1974).
2. C. R. Minick, G. E. Murphy, W. G. Campbell, Jr., *J. Exp. Med.* **124**, 635 (1966).
3. A. N. Howard, J. Patelski, D. E. Bowyer, G. A. Greshan, *Atherosclerosis* **14**, 17 (1971).
4. H. M. Sharma and J. C. Geer, *Am. J. Pathol.* **88**, 255 (1977).
5. R. Ansbacher, K. Keung-Yeung, J. C. Wurster, *Fertil. Steril.* **23**, 640 (1972).
6. N. J. Alexander, B. J. Wilson, G. D. Patterson, *ibid.* **25**, 149 (1974).
7. K. S. K. Tung, *Clin. Exp. Immunol.* **20**, 93 (1975).
8. T. Samuel, A. H. J. Kolk, P. Rumke, J. M. J. Van Lis, *ibid.* **21**, 65 (1975).
9. P. H. Rumke and M. Titus, *J. Reprod. Fertil.* **21**, 69 (1970).
10. P. E. Bigazzi, L. L. Kosuda, L. L. Harnick, R. C. Brown, N. R. Rose, *Clin. Immunol. Immunopathol.* **5**, 182 (1976).
11. N. J. Alexander and K. S. K. Tung, *Anat. Rec.* **188**, 339 (1977).
12. P. E. Bigazzi, L. L. Kosuda, L. L. Harnick, *Science* **197**, 1282 (1977).
13. N. J. Alexander, *Fertil. Steril.* **28**, 562 (1977).
14. L. Linnet and T. Hjort, *Clin. Exp. Immunol.* **30**, 413 (1978).
15. P. E. Bigazzi, L. L. Kosuda, K. C. Hsu, G. A. Andres, *J. Exp. Med.* **143**, 382 (1976).
16. M. L. Armstrong, in *Primates in Medicine*, J. P. Strong, Ed. (Karger, Basel, 1976), vol. 9, p. 16.
17. T. B. Clarkson and N. J. Alexander, in *Vasectomy: Immunologic and Pathophysiologic Effects*, I. H. Lepow and R. Crozier, Eds. (Academic Press, New York, in press).
18. The monkeys were obtained from Primate Imports, Inc., Port Washington, N.Y.
19. Each 100 g of diet contained 9.0 g of casein, 8.0 g of lactalbumin, 33.5 g of wheat flour, 10.0 g of dextrin, 3.6 g of sucrose, 23.5 g of butter, 6.26 g of Alphacel, 0.7 g of applesauce, 3.8 g of Hegsted salts mixture, 2.0 g of complete vitamin fortification mixture, and 0.145 g of cholesterol.
20. R. L. Rush, L. Leon, J. Turrell, in *Advances in Automated Analysis-Technicon International Congress, 1970*, E. C. Barton et al., Eds. (Thurman Associates, Miami, 1971), p. 503.
21. All plasma cholesterol and triglyceride determinations were performed at the Lipid Analytic Laboratory of the Bowman Gray School of Medicine, a participant in the surveillance phase of the Lipid Standardization Program of the Center for Disease Control, Atlanta, Ga.
22. T. B. Clarkson, T. E. Hamm, B. C. Bullock, N. D. M. Lehner, in *Primates in Medicine*, J. P. Strong, Ed. (Karger, Basel, 1976), vol. 9, p. 66.
23. S. Kibrick, D. L. Belding, B. Merrill, *Fertil. Steril.* **3**, 430 (1952).
24. S. Isojima, T. S. Li, Y. Ashtaka, *Am. J. Obstet. Gynecol.* **101**, 677 (1968).
25. The fluorescein-conjugated antibodies to monkey IgM and IgG were purchased from Cappel, Cochranville, Pa. We thank Dr. K. S. K. Tung, Albuquerque, N.M., for the fluorescein-conjugated antibodies to human IgA and C3. The specificity of all conjugates was checked by immunoelectrophoresis. Blocking experiments demonstrated the specificity of the response.
26. N. J. Alexander and K. S. K. Tung, in *Vasectomy: Immunologic and Pathophysiologic Effects*, I. H. Lepow and R. Crozier, Eds. (Academic Press, New York, in press).
27. P. E. Bigazzi et al., in *ibid.*
28. J. Folch, M. Lees, G. H. S. Stanley, *J. Biol. Chem.* **226**, 497 (1957).
29. The following measurements were determined for each artery: total surface area, surface area containing fatty streaks, fatty plaques, fibrous plaques, and diffuse intimal thickening. The presence and extent of ulcerated plaques, hemorrhage into plaques, and thrombosis were recorded.
30. We thank J. L. Palotay and his staff for necropsy assistance, W. D. Wagner and his staff for angiochemical determinations, M. G. Bond and his staff for assistance with pathologic evaluations, and L. Wood, J. Hren, and B. Mixon for technical assistance. Publication No. 971 of the Oregon Regional Primate Research Center. Supported by NIH grants HL-14164, HD-5969, and RR-00163.

21 March 1978; revised 8 May 1978

Proctodeal Feeding by Termitophilous Staphylinidae Associated with *Reticulitermes virginicus* (Banks)

Abstract. *Trichopsenius depressus* Le Conte, *Xenistusa hexagonalis* Seevers, and *Philoterme howardi* Kistner and *Gut solicit and receive proctodeal and stomodeal fluids from their host, as well as engage in allogrooming with them. No other trophic behaviors were observed, suggesting that the beetles are completely integrated into the termite's trophic system.*

Behavioral and biochemical studies of trophic relationships between termitophiles and their hosts are scarce. A few authors (1) have reported that integrated termitophiles (2) solicit and receive saliva or regurgitated crop contents (stomodeal food) (or both) from their host termites. But no one has reported the solicitation and receiving by a termitophile of the liquid hindgut contents of its host (proctodeal food). I report here the first observation of such proctodeal feeding by a termitophile, and also describe two other trophic relationships between the subterranean termite *Reticulitermes virginicus* (Banks) and its three termitophiles, *Trichopsenius depressus* Le Conte, *Xenistusa hexagonalis* Seevers, and *Philoterme howardi* Kistner and Gut.

Portions of three colonies of *R. virginicus* and their associated termitophiles

were collected in early April 1976 from fallen logs in the De Soto National Forest approximately 30 km north of Gulfport, Mississippi. Colony 1 yielded 29 *P. howardi*, 9 *X. hexagonalis*, and 30 *T. depressus*. Colony 2 yielded 4 *P. howardi*, 2 *T. depressus*, and 1 *X. hexagonalis*. Colony 3 yielded 18 *P. howardi*, no *X. hexagonalis*, and 6 *T. depressus*. The beetles from each source colony were placed with a representative mixture of about 200 of their host termites in petri dishes (14 cm) containing 13.5 g of moistened synthetic termite diet (3). Twenty-four hours were allowed for gallery establishment before the beginning of behavioral observations. A portion of a fourth colony of *R. virginicus* from the same area was collected during April 1977 and was used as a source of termitophiles for the dissection experiments. Behavioral observations (4) were made

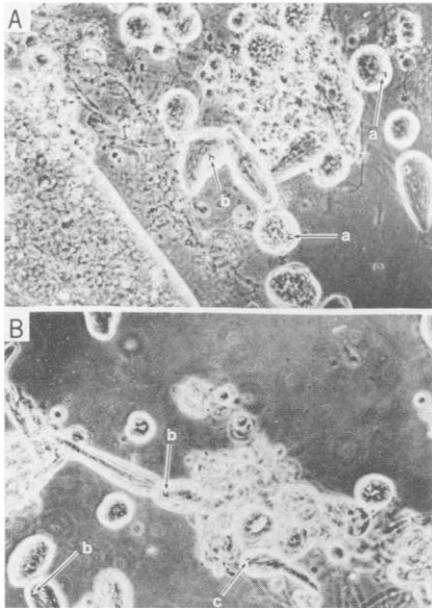


Fig. 1. Protozoan fragments isolated from hindguts of (A) *Xenistusa hexagonalis* Seevers and (B) *Philoterme howardi* Kistner and Gut. a, *Spirotrichonympha* sp.; b, *Trichonympha agilis* Leidy; c, *Pyrsonympha* sp.

with either a 10× hand lens or a dissecting microscope with minimal illumination. Hindguts of both beetles and termites were dissected in insect Ringers solution and examined for protozoa by phase-contrast microscopy.

Twenty-five hours of observation extending over a 6-week interval revealed that the three beetle species both solicited and received proctodeal and stomodeal fluids from their hosts, and that they engaged in frequent allogrooming exchanges with them. All three species used the same behavioral sequence to solicit proctodeal fluids from the termites. A beetle would first approach a termite from the rear and then vigorously tap the lateral margins of the termite's abdominal tip with its antennae and then with its mouthparts. This tapping usually resulted in the termite's secreting a drop of fluid, which was then quickly removed in whole or part by the beetle's mouth parts. On one occasion, *X. hexagonalis* was observed to grasp and hold onto a termite's abdominal tip with its mandibles until the fluid was secreted (5). The complete sequence normally required less than 30 seconds and was observed 10 times for *T. depressus*, 12 times for *X. hexagonalis*, and 4 times for *P. howardi*. It closely mimicked behavior of *R. virginicus* larvae soliciting proctodeal fluids from their nest mates.

The hindguts of several beetles of each species from colony 4 were examined for the presence of the characteristic pro-

tozoa found in the hindguts of their host (6). Figure 1 depicts at least three such protozoan species (Fig. 1, a to c) in various stages of swelling and disintegration, dissected out of *X. hexagonalis* and *P. howardi* (7). Such fragments were found in two out of six specimens examined for each species. Four specimens of *T. depressus* were examined, but no recognizable protozoa were detected. In none of the preparations were any of the protozoa found to be alive. Failure to find protozoa in most of the beetles examined is possibly a reflection of the length of time since their last proctodeal feeding. The three major species of protozoa found in the hindgut of the termites from colony 4, have been identified as *Spirotrichonympha* sp. (Fig. 2, a), *Trichonympha agilis* Leidy (Fig. 2, b), and *Pyrsonympha* sp. (Fig. 2, c). All were alive when the photograph was taken. Identifying the species of termite protozoa is always difficult at best, and the swelling and disintegration present in these specimens dissected from beetles precludes their accurate identification. That they are members of the same genera as those found in *R. virginicus* seems certain, and since these genera are unknown from Coleoptera, they must have been procured from the termites.

Successful procurement of stomodeal fluids from their host termites was also observed for these beetles, albeit for a lesser number of times than for the proctodeal exchanges (four times for *T. depressus*, five times for *X. hexagonalis*, and eight times for *P. howardi*). Whether these frequencies are a reflection of the "ad lib" sampling procedure (4), or a true reflection of differential feeding behavior, is not known. The behavioral sequence followed was essentially the same for all three species and mimics closely that used by the termites. A beetle would approach the termite from the front and tap the termite's labium, frons, and mandibles vigorously with its antennae and mouthparts. Then the termite usually regurgitated a drop of fluid between its mandibles (8) which was then quickly, wholly or in part, removed by the beetle's mouthparts. The complete sequence often lasted no more than 15 seconds.

The third form of possible trophic interaction observed between the beetles and *R. virginicus* was allogrooming (9). This behavior among social insects partly functions to remove solid debris from appendages not easily reached by the groomed individual. It can also function, however, as a mechanism for transmitting surface-borne chemicals among

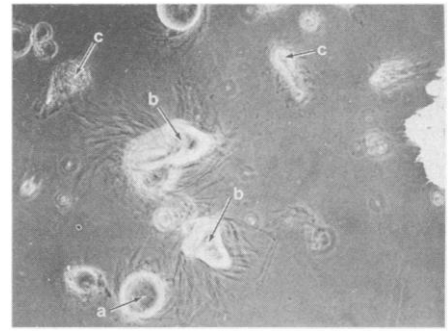


Fig. 2. Protozoa isolated from hindgut of *Reticulitermes virginicus* (Banks). a, *Spirotrichonympha* sp.; b, *Trichonympha agilis* Leidy; c, *Pyrsonympha* sp.

colony members. One such class of surface-borne chemicals is cuticular lipids, which, aside from their possible roles as caste or colony recognition cues (10), might also function as potential energy sources. It is thus conceivable that the three termitophiles may be able to meet at least some of their dietary lipid requirement by ingestion of the cuticular lipids removed from the legs of their hosts during allogrooming. The behavioral sequence went like this. The beetle approached a termite from its side. Then with its mandibles the beetle grasped one of the termite's legs, usually at the tibia, occasionally at the femur. The termite then ceased locomotion, while the beetle, starting from the initial point of attachment down toward the tarsi, proceeded to groom the termite's leg for up to a minute. When the beetle finished grooming and released the leg, the termite then resumed locomotion. Such allogrooming was observed 21 times for *T. depressus*, 13 times for *X. hexagonalis*, and 4 times for *P. howardi*. On at least two occasions, as many as three beetles were observed grooming one termite simultaneously (one beetle per leg). Despite the extended period of behavioral observation, no other feeding behavior (such as coprophagy or predation on eggs or larvae of *R. virginicus*) by the three beetles was noted.

If the beetles' sole source of food is that proffered by their hosts, then possibly their trophic regime approximates that of a first or second instar termite larva (their biomass equivalent). The young larvae of *Reticulitermes lucifugus* Rossi are known to be fed predominantly saliva and proctodeal food with little, if any, regurgitated stomodeal food (11). The proportion of each food type fed to the larvae is not known, nor has the biochemical makeup of the two fluids been ascertained. From data on other termites, however, it is possible to suggest

that the saliva functions to supply a carbon source of energy-rich lipids (12) and that the proctodeal fluids constitute, at least for the beetles, a major source of nitrogen via the protoplasm of ingested symbiotes (13). Since there is no evidence of the beetles' proffering any of their digestive fluids to the termites, their trophic relationship with the termites seems to be unidirectional.

Much remains unknown regarding the trophic interactions between these beetles and their hosts. A quantitative measurement of the relative frequency of each type of feeding behavior as well as a biochemical characterization of the stomodeal, proctodeal, and surface lipid reservoirs will be necessary before a detailed understanding of the trophic interactions can be obtained.

RALPH W. HOWARD

Southern Forest Experiment Station,
Forest Service, U.S. Department of
Agriculture, Gulfport, Mississippi 39503

References and Notes

1. A. E. Emerson, *Ann. Entomol. Soc. Am.* **28**, 369 (1935); D. H. Kistner, *ibid.* **66**, 197 (1973); *Sociobiology* **1**, 78 (1975); *Cimbebasia Ser. A* **4**, 99 (1975); _____, and H. R. Jacobson, *Sociobiology* **2**, 1 (1976); J. M. Pasteels, *Arch. Biol.* **79**, 381 (1968).
2. A record of the convoluted history of various classification schemes for the relationships between termitophiles and their hosts has been summarized (10). I find the classification scheme developed by Kistner [D. H. Kistner, in *Biology of Social Insects*, H. Herman, Ed. (Academic Press, New York, in press), vol. 1, chap. 8] to be most useful. Thus, "an integrated termitophile is one which from its behavior and its host's behavior can be clearly seen to be incorporated into the social life of the colony. In contrast, a nonintegrated termitophile is one which is not incorporated into the social life of the colony but rather which has adapted to the nest as an ecological niche. If it is not known whether a species is integrated or not, judgment is suspended until the requisite information is available."
3. J. K. Mauldin and N. M. Rich, *Ann. Entomol. Soc. Am.* **68**, 454 (1975).
4. J. Altmann, *Behaviour* **49**, 227 (1974).
5. A similar behavior has been observed [E. A. McMahan, *Ann. Entomol. Soc. Am.* **56**, 74 (1963)] between a pair of *Cryptotermes brevis* engaged in proctodeal exchange.
6. B. M. Honigberg, in *Biology of Termites*, K. Krishna and F. M. Weesner, Eds. (Academic Press, New York, 1970), vol. 2, pp. 1-36.
7. The taxonomic assistance of Dr. J. K. Mauldin, U.S. Department of Agriculture, Forest Service, Gulfport, Miss., and Dr. B. M. Honigberg, Department of Zoology, University of Massachusetts, in identification of these protozoa is acknowledged.
8. This fluid was consistently perceived as clear under my experimental conditions, suggesting that it was probably saliva rather than regurgitated crop contents. This conclusion would support the observations of McMahan, working with *Cryptotermes brevis* (5). She noted that regurgitated food is not fed to the dependent castes, and may, in fact, be exchanged rather infrequently between the other members of the colony.
9. Allogrooming is grooming of another individual, in contrast to self-grooming.
10. E. O. Wilson, *The Insect Societies* (Harvard Univ. Press, Cambridge, Mass., 1971).
11. H. Buchli, *Physiol. Comp. Oecol.* **2**, 145 (1950).
12. S. H. W. Cmelik, *J. Insect Physiol.* **17**, 1349 (1971); *Insect Biochem.* **2**, 361 (1972).
13. Ch. Noirot and C. Noirot-Timothee, *Biology of Termites*, K. Krishna and F. M. Weesner, Eds. (Academic Press, New York, 1969), vol. 1, pp. 49-88.

23 January 1978; revised 17 April 1978

Disulfiram Enhances Pharmacological Activity of Barbitol and Impairs Its Urinary Elimination

Abstract. *Disulfiram or diethyldithiocarbamate significantly enhanced the sleeping time induced by barbitol in rats. At identical time intervals after rats were injected with barbitol the concentration of barbitol in the blood or brain of animals that had previously received disulfiram was significantly higher than the concentrations in the corresponding tissues of control animals. Urinary excretion of barbitol was significantly reduced in disulfiram-treated animals.*

Disulfiram has been in use in avoidance therapy in certain cases of alcoholism for many years. Although disulfiram by itself is generally regarded as a relatively safe drug, several cases of exaggerated responses to some drugs, and sometimes toxic manifestations, were re-

ported in patients receiving disulfiram. For example, enhanced reduction of prothrombin level and evidence of bleeding were reported when an anticoagulant, warfarin, was administered to patients taking disulfiram (1). Typical symptoms of phenytoin overdosage were observed in patients receiving this drug and disulfiram (2). Psychosis and confusion were reported in patients taking both disulfiram and metronidazole (3). It has been suggested that disulfiram impairs the biotransformation of these drugs and thus enhances their pharmacological activity (4). Rats treated with disulfiram (200 mg/kg, intraperitoneally) 2 hours before the administration of hexobarbital slept three times as long as control rats (5). This effect was presumed to be due to a potentiation of the hexobarbital action in the central nervous system, because the amount of hexobarbital metabolized by the whole body in 30 minutes was not significantly altered by prior administration of disulfiram.

We have observed that disulfiram (400 mg/kg, injected intraperitoneally) enhances the pharmacological activity and toxicity of barbitol in the rat (6), although barbitol undergoes virtually no metabolic transformation in this species and is eliminated almost entirely in the urine (7). This suggests that enhancement by disulfiram of the pharmacological activity of certain drugs could involve some aspects other than the impairment of enzymes involved in the biotransformation of drugs.

Here we report that treatment of rats with disulfiram is associated with changes in the distribution of barbitol in blood, brain, liver, and kidneys, and that urinary excretion of barbitol is significantly reduced in disulfiram-treated rats.

Male Sprague-Dawley rats (180 to 200 g) were used in all experiments. They were housed in temperature-regulated quarters (23° to 25°C) on a 12-hour light-dark cycle (lights on from 0700 to 1900) and given free access to food and water. The animals were divided into groups of six rats each. The rats in some of the groups received an intraperitoneal injection of 1 percent carboxymethyl-

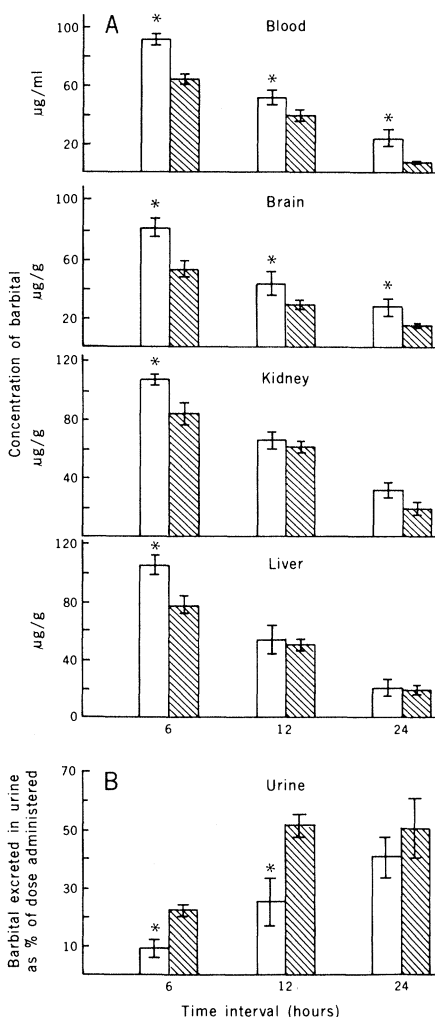


Fig. 1. The concentrations of barbitol in blood, brain, kidneys, and liver (A), and the amounts of barbitol excreted in urine (B) at 6, 12, and 24 hours after an intraperitoneal injection of barbitol (100 mg/kg). The data in (B) are expressed as percentages of the dose administered. Unshaded bars, disulfiram-treated rats; shaded bars, CMC-treated rats. Each value represents the mean $\pm$ standard error of the mean obtained from a different group of six rats each. Asterisks indicate results statistically different from control ($P < .05$) by two-tailed Student's t -test.