If we now assume that Th²³² in deepsea sediments is a constituent of the nonauthigenic minerals, the dates obtained by the use of the ratio are subjected to rather large errors, since the rate of sedimentation may change with time as Broecker, Turekian, and Heezen (8) have shown. The postglacial rate of noncarbonate sedimentation in the equatorial Atlantic is only one-third or less of that during the last glacial age. A smaller change can be expected in the Pacific. This change would alter the Th²³⁰/Th²³² ratio by the same factor when the rate of Th²³⁰ precipitation from seawater is assumed to have remained constant. It can easily be shown that the error in the age determination becomes

$$t_1 - t = \frac{1}{\lambda_{io} - \lambda_{th}} \ln p = 1.1 \times 10^5 \ln p \text{ years}$$

where t_1 is the age obtained under the assumption that the rate of sedimentation has not changed, t is the real age, and p is the ratio of the rates of sedimentation at the deeper level to the recent rate of sedimentation. If the rate has changed by a factor e, the error becomes as large as 110,000 years. Already a change of 38 percent causes an error of 52,000 years. This would explain the low rates of sedimentation obtained by Miyake and Sugimura.

In general, it can be concluded that the Th²³⁰/Th²³² methods will not give correct ages or rates of sedimentation unless the basic assumptions are carefully controlled and investigated. In fact, in the Caribbean cores dated by the ratio of Pa²³¹ to Th²³⁰ (9), the ratio of Th²³⁰ to Th²³² was shown to give conspicuously discrepant ages (10). F. F. Koczy

Marine Laboratory,

University of Miami, Miami, Florida

References and Notes

- 1. E. Picciotto and S. Wilgain, Nature 173, 632 (1954).
 E. D. Goldberg and M. Koide, Science 128,
- 1003 (1958). 3. V. I. Baranov and L. A. Kuzmina, Geochem-
- istry 2, 131 (1958); I. Almodovar, thesis Carnegie Institute of Technology, Pittsburgh Pa. (1960). Y. Miyake and Y. Sugimura, Science 133,
- 4. 1823 (1961). 5. P
- N. Hurley et al., U.S. Atomic Energy Comm. Rept. NYO-3939 (1958), p. 114;
 P. M. Hurley et al., U.S. Atomic Energy Comm. Rept. NYO-3941 (1960), p. 267.
 E. Picciotto, Ciel et terre 3-4, Mar.-Apr. (1960)
- 6. E.
- (1960).
 7. F. F. Koczy, Deep-Sea Research 3, 93 (1956); —, E. Picciotto, G. Poulaert, S. Wilgain, Geochim. et Cosmochim. Acta 11, 103 (1957).
 8. W. S. Broecker, K. K. Turekian, B. C. Heezen, Am. J. Sci. 256, 503 (1958).
 9. J. N. Rosholt et al., J. Geol. 69, 262 (1961).
 10. This report is contribution No. 340 from the Marine Laboratory University of Miami
- 10. Marine Laboratory, University of Miami.
- 18 July 1961
- 15 DECEMBER 1961

Sex as Regulator of Triglyceride Metabolism in the Mosquito

Abstract. The female mosquito, in contrast to the male mosquito or the male and female house fly, synthesizes triglycerides when maintained on glucose; after 7 days, the amount of triglycerides in the female may be 50 times that in the male. Polyunsaturated fatty acids are absent from the newly synthesized triglycerides.

Labeled precursors of metabolic products facilitate the study of biosynthesis, since newly formed products can be differentiated from products already present. In living animals, however, quantitative estimation of synthesis from labeled precursors requires a steady state, detailed knowledge of specific activity-time relationships between precursors and product, or knowledge of the distribution of the product over many metabolic pools. It is therefore customary to carry out studies of biosynthesis in vitro, where the specific activity of the environment can be kept constant, but where the physiological control mechanisms of the intact animal are no longer functional. To study synthesis of triglycerides in vivo, we have used the adult female mosquito because it can be starved until virtually no triglycerides remain. Therefore, the triglycerides that appear after subsequent feeding on a lipid-free diet are due entirely to new synthesis.

Adult mosquitoes [Aedes sollicitans (Walker) and A. taeniorhynchus (Wied.)] were obtained from larvae reared at 25°C on rabbit pellets and yeast. House flies (Musca domestica L., susceptible strain) were obtained as pupae from the U.S. Department of Agriculture, Orlando, Florida. Within 2 hours after emergence, males and females were selected and kept at 25°C in glass jars provided with a moist cheesecloth pad and a feeding vial containing a cheesecloth wick soaked in 10-percent glucose solution. The glass jars and the feeding solution were changed daily. Duplicate samples of ten mosquitoes or three flies each were killed at various intervals by brief exposure to chloroform vapor. This pooling of samples reduced biological variation between duplicates to 10 percent or less. The insects were homogenized, and the homogenate was extracted twice with 1 ml of methanol and chloroform (1:1), with centrifugation after each extraction. Chloroform (2 ml) was added, methanol and water-soluble impurities were removed by washing twice with 1/2 ml of water, phospholipids were absorbed by shaking the chloroform eluate with 100 mg of silicic acid, and triglycerides were determined from the glycerol moiety (1). Phospholipids were determined (2) after elution from the silicic acid with 2 ml of methanol.

On emergence, triglyceride levels of males and females were quite similar. Subsequently, triglycerides of the male and female house flies and of the male mosquito diminished gradually. By contrast, the female mosquito (both Aedes sollicitans and A. taeniorhynchus) showed a constant net synthesis of triglycerides from the first until the sixth or seventh day after emergence (Fig. 1), ranging from 70 to 115 μ g/day in different experiments. At the maximum, the amount of triglycerides may exceed the lipid-free dry weight. In the same interval, the triglycerides in the male dropped to 10 to 20 μ g (Fig. 1A). The observed differences were not due to





differences in food intake. Both sexes fed freely on glucose, as evidenced by distended abdomens. When maintained on water, the female catabolized 20 μ g/day from emergence until death, so that additions to the pool from the different metabolic compartments and utilization by tissues take place at a constant rate.

At emergence, phospholipids and triglycerides comprised more than 90 percent of the total lipids. Free fatty acids and hydrocarbons made up the balance. Phospholipid levels remained virtually unchanged during the entire experiment $(46 \pm 3 \ \mu g \text{ per female},$ $36 \pm 4 \,\mu g$ per male mosquito; 240 ± 20 μg per house fly). Sterols (free cholesterol, including 10 to 35 percent dehydrocholesterol) were found in amounts of only 0.1 μ g per mosquito.

In order to establish the composition of the fatty acids synthesized solely from glucose, mosquitoes were maintained on water until the triglyceride level was down to 5 μ g and then fed 10-percent glucose for 5 days (Fig. 1B). At that point, methyl esters of the triglyceride fatty acids, 99 percent of which had been newly synthesized, were subjected to gas-liquid chromatography (Table 1). Palmitic, palmitoleic, and oleic acid comprised 92 percent of the new fatty acids, palmitoleic acid being one-third of the total. Whereas the 5 μ g of triglycerides left after starvation still contained 12 percent linoleic acid, this amount is represented only as a trace in the 400 μ g synthesized from glucose. It is therefore likely that the mosquito does not make polyunsaturated fatty acids.

A continuously falling level of triglycerides does not necessarily preclude all synthesis. Robbins et al. (3) showed that female and male adult house flies. when fed milk powder and sugar, in-

Table 1. Triglyceride fatty acids in female mosquitoes (5).

Chain length	Double bonds	Amount (µg per mosquito)	
		3 ¹ /2 days on water	3 ¹ / ₂ days on water plus 5 days on glucose
Short (6-12)		0.02	1
14	0	0.08	12
14	1	0.01	2
16	0	1.46	140
16	1	1.20	136
18	0	0.27	14
18	1	1.36	95
18	2	0.60	Trace
Long		Trace	Trace

corporate C14-labeled acetate in the saponifiable lipid fraction, but these workers did not separate phospholipids from triglycerides.

The ability of the female mosquito to build a huge fat body from glucose at a constant rate, as contrasted with the female house fly or the male mosquito, may facilitate the study of physiological factors that control lipogenesis in vivo (4).

> EMILE VAN HANDEL PATRICK T. M. LUM

Entomological Research Center, Florida State Board of Health, Vero Beach

References and Notes

- 1. E. Van Handel, Clin. Chem. 7, 249 (1961). 2. G. R. Bartlett, J. Biol. Chem. 234, 46 G. R. (1959). 466
- 3. W. E. Robbins, J. N. Kaplanis, S. J. Lou-loudes, R. E. Monroe, Ann. Entomol. Soc. Am. 53, 128 (1960).
- This work was supported by a grant (E-3112)
- from the National Institutes of Health. The data in Table 1 were provided by Dr. Leon Swell, Veterans Administration Center, Martinsburg, W. Va. 5.

10 August 1961

Licking Rates in Infant Albino Rats

Abstract. Two groups of neonatal rats raised from birth without the opportunity to drink, and tested at different age levels, exhibited the same general characteristics of licking rate that adult rats do. This suggests that licking in the rat is organized on a genetic-maturational basis.

The purpose of the experiment reported here was to determine whether constancies in licking rate reported for adult albino rats are found also in infant rats. Other workers (1, 2) have reported a mean licking rate in adults of 6 to 7 licks per second and a range from 5 to a little over 8 licks per second, with the mean rate constant in both sexes for varying levels of thirst, and for water, sucrose, saccharin, and saline solutions.

In the absence of evidence to the contrary, it might be argued that these constancies are the result of reinforcement of certain rates of licking. On the other hand, if rats raised from birth without an opportunity to drink displayed, when tested at different age levels, the same licking rate as adults, it would suggest that drinking in the rat is primarily a genetically and maturationally determined response.

We tested two litters of neonatal rats from the colony maintained by the University of Missouri. Each litter was composed of 3 males and 3 females. Each litter was reared with the mother

from birth until the first test day, then was separated from her and housed in a separate cage. The water supply for the mother was suspended from the ceiling of the cage, and thus the young rats were prevented from drinking prior to the first test day. Both litters had unlimited access to dry food prior to weaning and throughout all the testing and maintenance periods. The normal weaning age in the rat is between 21 and 28 days (3). The rats in group 1 were weaned at noon of the 21st day after birth; they were then tested daily, at 8 P.M. and 8 A.M., for 5 days. The rats in group 2 were weaned at noon of the 18th day and were tested at 4 P.M. and 8 P.M. on the first day and at 8 A.M., noon, and 8 P.M. on the four following days. All test sessions were 30 minutes long.

The apparatus consisted of six Hoeltge HB-11 cages with attached floormounted plastic drinking cups and 50ml glass tube reservoirs. The mouth of the cup was 10 mm in diameter. The test solution was water. Each time the animal's tongue came in contact with the water, an electronic circuit was completed, an $8-\mu a$ current passed through its body, and a record of the tongue lap was recorded on an Esterline Angus tape moving at 1.905 cm/sec. The experimental room was maintained at a temperature between 76° and 78°F throughout the experiment.

Four licking rates were measured: The rate at the time of contact with the drinking cup; the rate in the first burst of licking of at least 1-second duration in each session; the rate within sessions; and the rate within bursts of more than 1-second duration. The means and the range of responding in the initial contact with the drinking cup and in the first burst of licking of at least 1-second duration in the first four sessions are presented in Table 1. For both groups, the mean rate of licking on first contact with the cup parallels the mean rate of licking in adult animals. Individual rates show some variability. On its initial contact with the cup in the first test session, rat F2 in group 1 licked at the rate of 9.5 licks per second. In the second test session, rats F3 and M3 in group 1 licked at the rate of 11.4 and 9.5 licks per second, respectively. Similarly, in the first test session both F1 and F3 in group 2 licked at the rate of 9.1 licks per second, and M2 licked at the rate of 9.5 licks per second. No systematic relationship between sex and licking rate was observed in either group.