Experiment	Sudden temperature change (°C) From	To	No. of rings on strobila Initial	Maximum	Average time[b] (days)	No. of rings on strobila which were tentaculate on conclusion of observations
1964 1 and 2	11	15	1	..	5 ± 1	0[a]
			1	–		0[a]
			1	2		1[a]
			3	4		3[a]
			7	9		5
			8	10		8[a]
			11	12		8
1965/1	8	21	13	13	5	2
1966 1, 2, and 3	5	15	1	2	4 ± 2	2
			2	2		2
			3	4		4
			3	3		3
			3	4		4
			3	4		4
			4	7		0
			4	7		2
			6	8		7[a]
			6	8		2
			6	7		2
			9	14		7
			12	22	14.5 ± 3.5	1
			12	12		0
			13	16		0
			16	18		0
			17	22		1
			18	24		2
1966/4	5	7	2	19		0
			3	20		0
1966 5 and 6	5	10	3	5	16 ± 2	4[a]
			3	7		7
			5	5		3[a]
1966 7 and 8	5	10	5	(14)		0
			7	(22)		0
			8	(18)		0
			8	24		0

[a] These specimens exhibited resorption of the proximal rings to some extent; in other specimens the number of rings remained unchanged.
[b] These figures show the range in time between the start of the experiment and the first appearance of tentacles in place of lappets on the ephyral rings. Numbers in brackets indicate values at termination of the experiment.

the Kiel Fiord describes this form which he refers to as a 'polyp-strobila', and records that their appearance was subsequent to a drop in temperature to below 1 °C for over 3 months. More recently SPANGANBERG[6] has shown that this effect can be induced by a transference of strobilae from natural to artificial sea water (though it should be noted that her samples were from a population of *Aurelia* that strobilate at a much higher temperature than the European examples). These results emphasize the plasticity of form found in the tissues of the *Cnidaria*, the phylogenetic significance of which has been discussed by THIEL[5] and WERNER[7]. The apparent dependence on environmental conditions also raises the problem of how strobilation is regulated within the scyphistoma. It can be noted that there appear to be 2 distinct stages, the formation of ephyral rings as a result of the buckling of the body wall of the scyphistoma, and the morphogenesis of these into ephyra larvae. The reversion to polyp form begins in the most proximal rings; this could be interpreted on the basis of a gradient of activity controlling the metamorphosis of the scyphistoma into the strobila operating along the proximal-distal axis, and which has its centre at the distal end of the polyp. Recent work on the morphogenesis of *Hydra* by BURNETT[8] and LENTZ[9] has shown that neurosecretory material in the hypostome may be involved in the organization of body form. In *Aurelia* maintenance of strobilation could be controlled by a similar mechanism, dependent in this case on environmental changes[10].

Zusammenfassung. Scyphistomalarven von *Aurelia aurita* strobilieren normalerweise bei Temperaturen unter 10 °C. Die Strobilen können sich bei stetigem Ansteigen der Temperatur bis zu 14 °C normal entwickeln, ein plötzlicher Anstieg über 5 °C führt jedoch zur Entwicklung polypoider Tentakeln anstelle der Ephyra-Arme. Diese Formplastizität veranlasst die Frage nach der Regulation der Metamorphose des Scyphistoma zur Ephyra.

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[6] D. B. SPANGANBERG, J. exp. Zool. *160*, 1 (1965).
[7] B. WERNER, Ann. N.Y. Acad. Sci. *105*, 461 (1963).
[8] A. L. BURNETT, N. A. DIEHL, and F. DIEHL, J. exp. Zool. *157*, 227 (1964).
[9] T. L. LENTZ, Science *150*, 633 (1965).
[10] This work was supported by a grant from the Royal Society's Fund for Scientific Research in Schools.

Deposition of Fat in the Liver Following Administration of Caffeine

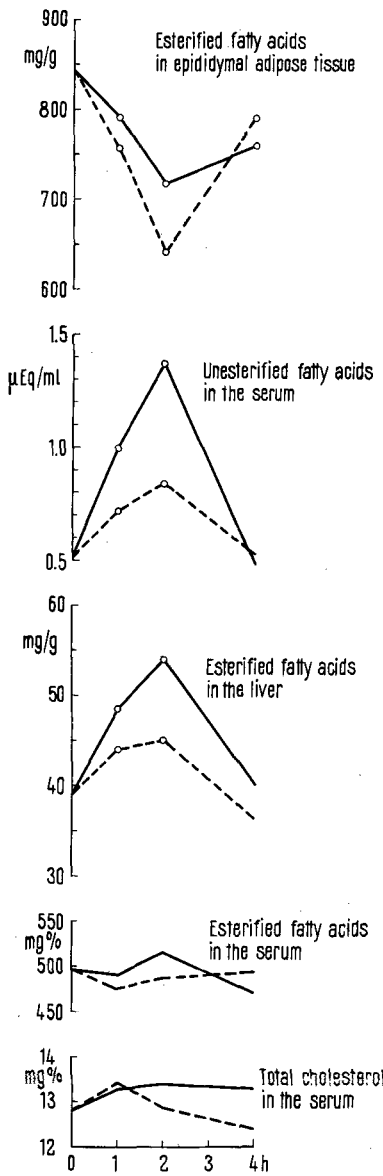
During the last years there have been many reports on the increase in non-esterified fatty acids in the blood with subsequent deposition of fat in the liver under the influence of numerous pharmacologically active substances, the effect of which depends partly on a direct attack on the lipolytic enzyme system of the adipose tissue, and partly on being supposed to come into action through the mediation of hormonal and neural carrier mechanisms (PAOLETTI[1]). Purine derivatives likewise belong to the fat-mobilizing drugs. However, as far as we know, no investigations have been made up to the present as to whether caffeine will also produce an accumulation of fat in the liver.

For this reason, white mice (NMRI breed) were given caffeine-sodium-benzoate in doses corresponding to 25 and 50 mg/kg of caffeine s.c. Following this, the content of esterified fatty acids in the liver and in the epididymal

[1] R. PAOLETTI, *Lipid Pharmacology* (Academic Press, New York, London 1964).

adipose tissue, as well as the content of esterified and non-esterified fatty acids and cholesterol in the serum, was determined. These determinations were made by the following methods: Determination of esterified fatty acids according to FRIED and HOEFLMAYR[2] (in the tissues following adequate modification), non-esterified fatty acids according to DUNCOMBE[3] (some values for comparison according to DOLE and MEINERTZ[4]), and the cholesterol determination being effected by following the method of RICHTERICH and LAUBER[5]. For each test 10–15 animals were used. All animals were kept at their usual ambient temperature of 25 °C, having free access to food (Altromin standard diet) and water.

The experiments disclosed (see Figure) that under the influence of caffeine the content of esterified fatty acids in the adipose tissue, equal to the content of neutral fat, had decreased until the second hour and increased thereafter. There was no evidence of quantitative differences between the 2 caffeine doses. Simultaneously, there was a rise in the non-esterified fatty acid level of the serum, dependent on the dosage, with a maximum after 2 h.



Lipid content of epididymal adipose tissue, liver and serum of white mice after administration of 25 mg/kg (····) and 50 mg/kg (——) caffeine s.c. o = Value significantly different from the controls. $P \leqq 0.05$.

After a duration of 4 h, the non-esterified fatty acids had reached normal limits again, in conformation with the results obtained by KHAN et al.[3]. Hence it follows that under the influence of caffeine a mobilization of the depot fats takes place due to the fact that purine derivatives are able to inhibit the enzyme phosphodiesterase in the adipose tissues and thus the breakdown of cyclic 3′,5′-AMP (BUTCHER and SUTHERLAND[7]). The accumulating cyclic 3′,5′-AMP produces an activation of the lipase, the latter decomposing neutral fats into fatty acids and glycerol being released into the blood (RIZACK[8]).

The non-esterified fatty acid level in the blood is a result of the delivery of fatty acids from the adipose tissues and of their absorption in the organs. Above all, the heart and skeletal muscle cover an essential part of the energy requirements by oxidation of fatty acids. Since caffeine has a stimulating effect on the heart and the motor activity of the animals (HEIM and HAAS[9]), the increased liberation of fatty acids from the depots might be partly compensated by an increased withdrawal and oxidation, which may also account for the inconsistency of the blood level in the first hour.

Concomitant with the mobilization of fat, there is an increase in the content of esterified fatty acids in the liver with a maximum after 2 h, depending on the dosage. This means that a considerable percentage of fatty acids is absorbed in the liver where they undergo esterification. This agrees with the experience that the incorporation of fatty acids into the liver and their subsequent esterification into neutral fat and phospholipids are proportional to the content of fatty acids in the blood (HAVEL[10]). According to ROBINSON[11], part of the triglycerides formed in the liver will be delivered again to the blood and retransported to the adipose tissues. No rise in the content of esterified fatty acids in the serum could be seen in our experiments. It seems, therefore, that under the influence of caffeine the fatty acids taken up and esterified by the liver are retained in the liver to a larger extent or that the uptake of esterified fatty acids from the blood by the organs is enhanced. The cholesterol level also remained unchanged.

Zusammenfassung. Koffein in Dosen von 25 und 50 mg/kg s.c. führte bei weissen Mäusen als Folge einer gesteigerten Lipolyse im Depotfett zur Abnahme des Fettgehalts des Fettgewebes, zur Zunahme der unveresterten Fettsäuren im Serum und zur Einlagerung von veresterten Fettsäuren in die Leber. Die Hauptwirkung fand sich nach 2 h. Der Gehalt des Serums an veresterten Fettsäuren und Gesamtcholesterin blieb unverändert.

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[2] R. FRIED and J. HOEFLMAYR, Klin. Wschr. *41*, 727 (1963).
[3] W. G. DUNCOMBE, Clin. chim. Acta *9*, 122 (1964).
[4] V. P. DOLE and H. J. MEINERTZ, J. biol. Chem. *235*, 2595 (1960).
[5] R. RICHTERICH and K. LAUBER, Klin. Wschr. *40*, 1252 (1962).
[6] A. U. KHAN, R. B. FORNEY, and F. W. HUGHES, Arch. int. Pharmacodyn. *151*, 466 (1964).
[7] R. W. BUTCHER and E. W. SUTHERLAND, J. biol. Chem. *237*, 1244 (1962).
[8] M. A. RIZACK, J. biol. Chem. *239*, 392 (1964).
[9] F. HEIM and B. HAAS, Arch. exp. Path. Pharmak. *226*, 395 (1955).
[10] R. J. HAVEL, in *Lipid Pharmacology* (Ed., R. PAOLETTI; Academic Press, New York, London 1964).
[11] D. S. ROBINSON, in: *Proc. Internat. Symp. Lipid Transport* (Ed., H. G. MENG; Thomas, Springfield 1964). p. 194.