Adv. Odonatol. 5 : 59-65 December, 1991

UTILIZATION OF DIETARY FATTY ACIDS BY LARVAL *AESHNA CYANEA* (ANISOPTERA : AESHNIDAE)

H. KOMNICK

Institut für Cytologie
Rheinische Friedrich-Wilhelms-Universität Bonn,
Ulrich-Haberland-Str. 61 a,
5300 Bonn 1, Bundesrepublik Deutschland

Unsaturated C14 through C24 fatty acids were readily absorbed by *Aeshna* larvae irrespectively of the number, position and cis-trans configuration of their double bonds. They caused a heavy accumulation of triglyceride droplets in the enterocytes. Saturated C8 through C15 fatty acids were absorbed in the same way but to a lesser extent, while saturated short chain fatty acids ($C < 8$) were absorbed in free form without triglyceride synthesis, and absorption of saturated long-chain fatty acids ($C > 15$) was not detected with the methods used.

Aeshna larvae feed on various vertebrate and invertebrate animals, whose lipid content can amount to one third of their dry weight (HANSON *et al.*, 1985). Fatty acids of the dietary lipids prevail in the form of esters, mainly as triglycerides.

We have tested a large number of fatty acids with chain lengths ranging between C4 and C24 in order to examine the utilization of defined lipids and the mechanism of intestinal absorption.

Thin layer chromatography revealed that all esterified fatty acids tested such as glyceryl-, ethyl-, cholesteryl-, wax esters and phospholipids were hydrolysed in the midgut lumen. This is documented in Figure 1 for the ethylesters of medium and long-chain saturated fatty acids which were hydrolysed regardless of their fluid and crystalline states. Such and similar digestion experiments suggest that absorption takes place in form of free fatty acids after esterolysis of dietary lipids.

Short-chain fatty acids of chain lengths below C8 are transported unesterified across the midgut epithelium into the hemolymph, while those of longer chains are re-esterified mainly to triglycerides in the absorptive cells.

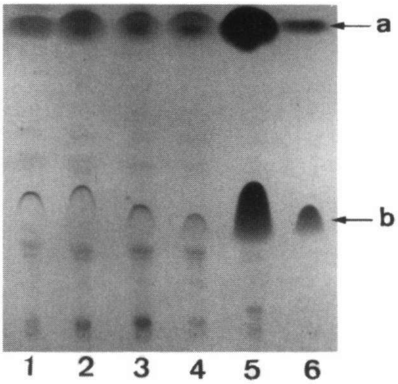

Fig. 1. Thin layer chromatogram of the midgut luminal contents isolated 2 days after infusion of : — 1, lauric acid ethylester (C12 : 0) ; — 2, myristic acid ethylester (C14 : 0) ; — 3, palmitic acid ethylesterr (C16 : 0) ; — 4, stearic acid ethylester (C18 : 0) ; — 5, oleic acid ethylester (C18 : 0) ; — 6, standard : a, oleic acid ethylester ; b, oleic acid.

This is shown in the next two figures. Six hours after tricaproin ingestion, there is a high increase in free fatty acids of the midgut epithelium when compared to the fasting controls, followed by a decrease after 24 hours (Fig. 2A). These changes have no effect on the esterified fatty acids of the epithelium (Fig. 2B). This contrasts to the results obtained after tricaprylin ingestion. In this case the esterified fatty acids increase after 24 hours, apparently at the expense of the free fatty acids which decline (Fig. 2A, B).

Thin layer chromatography reveals that the increase in esterified fatty acids is largely due to the formation of triglycerides which represent the major lipid class of the epithelial lipid droplets (Fig. 3).

The triglycerides resulting from fatty acid absorption are accumulated in the enterocytes and transiently stored in the form of lipid droplets. Fig. 4 shows that ingestion of the unsaturated myristoleic acid leads to a heavy accumulation of large, osmiophilic lipid droplets, while ingestion of the corresponding saturated myristic acid causes only a moderate accumulation of small, osmiophobic lipid droplets.

This difference is even more pronounced in the case of the next pair of even-numbered fatty acids. Again, tripalmitolein induces a heavy accumulation of lipid droplets, whereas tripalmitin, the saturate of equal chain length does not (Fig. 5a and b).

The amount of lipid droplets can be used for a rough estimate of absorption. The following tables summarize the results of such estimates (KOMNICK, 1988 ; BAUERFEIND & KOMNICK, 1989). All unsaturated fatty acids tested of chain lengths between C14 and C24 were equally absorbed. The

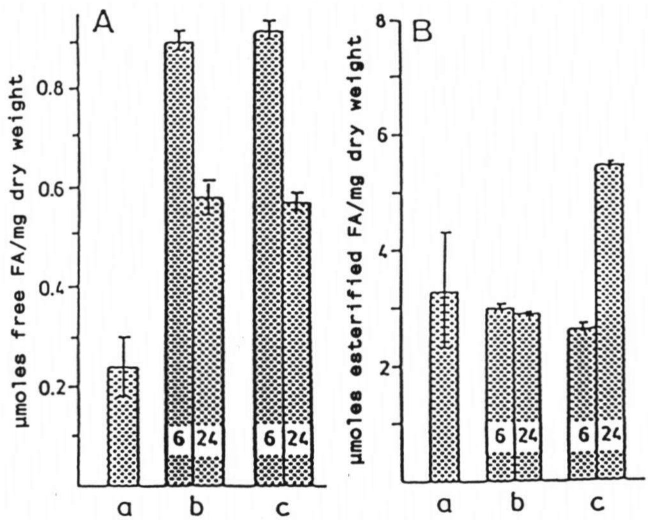

Fig. 2. Free (A) and esterified (B) fatty acids in the midgut epithelium of: (a) fasting control larvae and 6 and 24 hours after ingestion of (b) tricaproin (C6 : 0) and (c) tricaprylin (C8 : 0).

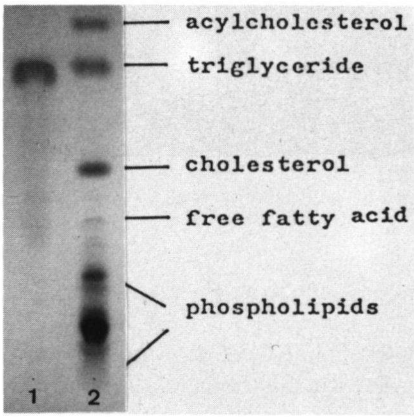

Fig. 3. Thin layer chromatogram of the midgut epithelium 2 days after the oral infusion of a mixture of lauric acid ethylester and monolaurin (lane 1 : lipid droplets isolated from the midgut epithelium by homogenization and subsequent centrifugation : lane 2 : pellet of midgut epithelium after lipid droplet isolation. Triglycerides are the predominant lipid class of the lipid droplets. Remnants are still present in the tissue pellet due to incomplete removal of lipid droplets).

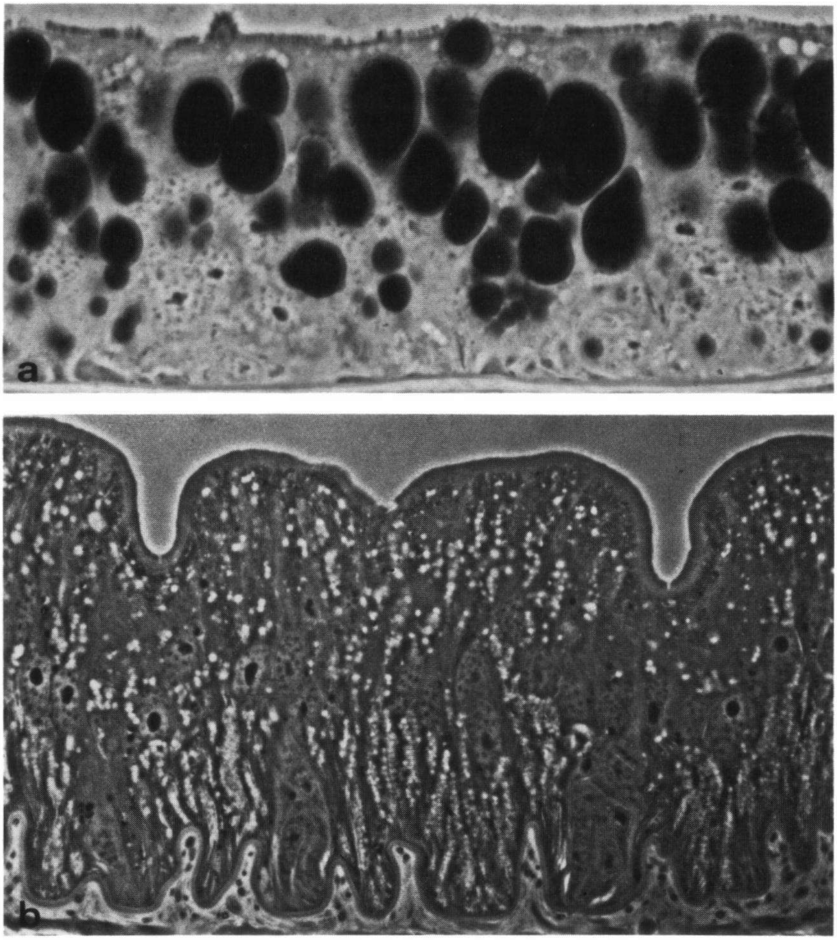

Fig. 4. Cross sections of the midgut epithelium fixed 2 days after the ingestion of (a) myristoleic acid (C14 : 1) and (b) myristic acid (C14 : 0). × 800.

number, position and cis-trans configuration of double bonds had no major effect. (Tab. I).

However, the results obtained with saturated fatty acids strikingly depended on the chain length (Tab. II). Short-chain saturates did not lead to epithelial lipid droplets, because — as already pointed out — they are transported as free fatty acids across the midgut epithelium. Medium-chain saturates from C8 to C15 induced a moderate and irregular accumulation of lipid droplets, while long-chain saturates including the most frequent palmitic and stearic acid failed to do so.

Table I

Intestinal absorption of mono- and polyunsaturated free fatty acids as judged from the accumulation of lipid droplets in the midgut epithelium

	Fatty acid	Characterization [1]	Accumulation of lipid droplets
1	Myristoleic	14 : 1 Δ 9 cis	+++
2	Palmitoleic	16 : 1 Δ 9 cis	+++
3	Petroselinic	18 : 1 Δ 6 cis	++
4	Oleic	18 : 1 Δ 9 cis	+++
5	Elaidic	18 : 1 Δ 9 trans	++
6	cis-Vaccenic	18 : 1 Δ 11 cis	+++
7	trans-Vaccenic	18 : 1 Δ 11 trans	++
8	Linolic	18 : 2 Δ 9, 12 cis	+++
9	Linoelaidic	18 : 2 Δ 9, 12 trans	+++
10	Linolenic	18 : 3 Δ 9, 12, 15 cis	+++
11	Eicosenoic	20 : 1 Δ 11 cis	+++
12	Eicosadienoic	20 : 2 Δ 11, 14 cis	+++
13	Eicosatrienoic	20 : 3 Δ 11, 14, 17 cis	+++
14	Arachidonic	20 : 4 Δ 5, 8, 11, 14 cis	++
15	Erucic	22 : 1 Δ 13, cis	++
16	Docosahexanoic	22 : 6 Δ 4, 7, 10, 13, 16, 19 cis	++
17	Nervonic	24 : 1 Δ 15 cis	++

[1] Number of C-atoms, number, position and configuration of double bonds.
+++ heavy, ++ intermediate accumulation (according to subjective estaminates).

Table II

Intestinal absorption of saturated lipids as judged from the accumulation of lipid droplets in the midgut epithelium

	Fatty acid	Chain Length	Lipid accumulation
1	Butyric	4 : 0	–
2	Valeric	5 : 0	–
3	Caproic	6 : 0	–
4	Heptanoic	7 : 0	±
5	Caprylic	8 : 0	±
6	Nonanoic	9 : 0	± (++)
7	Capric	10 : 0	± (++)
8	Undecanoic	11 : 0	± (++)
9	Lauric	12 : 0	± (++)
10	Tridecanoic	13 : 0	± (++)
11	Myristic	14 : 0	± (++)
12	Pentadecanoic	15 : 0	±
13	Palmitic	16 : 0	–
14	Heptadecanoic	17 : 0	–
15	Stearic	18 : 0	–
16	Arachidic	20 : 0	–
17	Behenic	22 : 0	–

– Questionable, not clearly distinguishable from the control.
± Slight, but irregular (i.e. not in all larvae or not at all places of the epithelium).
(++) In some larvae or at some places of the epithelium intermediate accumulation, but well below the heavy accumulation observed after the ingestion on unsaturated lipids.

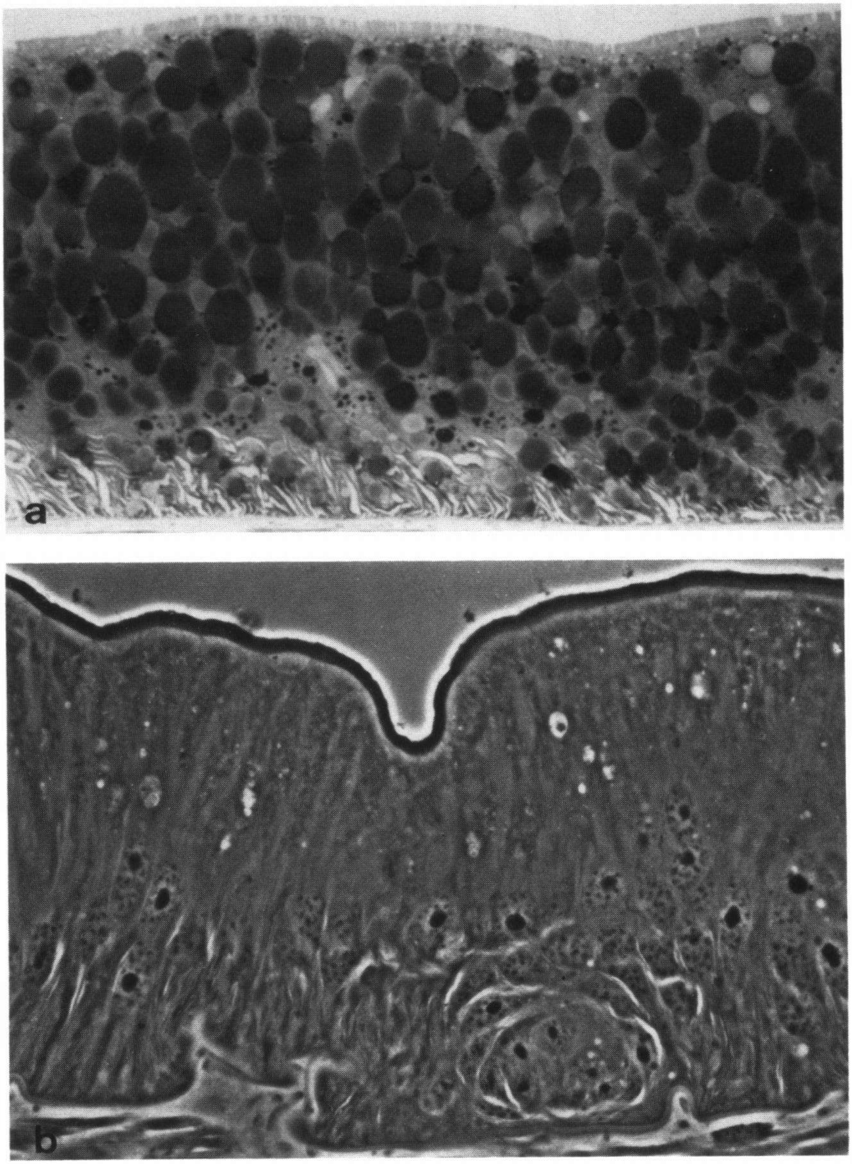

Fig. 5. Cross sections of the midgut epithelium fixed 2 days after the ingestion (a) of tripalmitolein (C16 : 1) and (b) tripalmitin. × 800.

These results suggest, that the absorption of saturated medium- and long-chain fatty acids by *Aeshna* larvae is greatly inferior to the uptake of unsaturates.

The demand for palmitic and stearic acids, the most frequent unsaturated acids of the body lipids, may be covered by chain-elongation of shorter-chain saturates or chain-shortening and saturation of longer-chain unsaturates. A further possible source are dietary wax esters. For recent results have shown that *Aeshna* larvae are able to absorb fatty alcohols released from wax esters including palmityl and stearyl alcohol.

REFERENCES

Bauerfeind, R. & H. Komnick, 1989. Intestinal absorption of defined lipids by the larval dragonfly Aeshna cyanea (Insecta, Odonata) : free and esterified saturated fatty acids. *J. Insect Physiol.* 35 : 155-164.

Hanson, B. J., K. W. Cummins, A. S. Cargill & R. R. Lowry, 1985. Lipid content, fatty acid composition and the effect of diet on fats of aquatic insects. *Comp. Biochem. Physiol.* 80B : 257-276.

Komnick, H., 1988. Intestinal absorption of defined lipids by the larval dragonfly Aeshna cyanea (Insecta, Odonata). Mono- and polyunsaturated free fatty acids and their homotriglycerides. *J. Insect Physiol.* 34 : 105-110.

Received October 3, 1989 / Accepted May 21, 1990