

ABSTRACT

BOOK

Molecular Biology Week '91
Poster Session

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N201-N202 Memorial Union

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Analysis of phospholipids and fatty acids in heads of normal and visual mutant *Drosophila* from the standpoint of cellular signalling.

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Visual receptors survive the attack of an armada of deadly toxins by turning over their membranes and their associated lipids and proteins. We genetically dissected the phospholipids and fatty acids in the heads of wild type (+) and mutant *Drosophila* (*ora*, *norpA*, and *pinhead*). The compound eye dominates the head and contains visual membranes in a large rhabdomere, which is composed of closely packed microvilli. *norpA* is of interest because it lacks PLC, a membrane associated protein involved in transduction (cleavage of PIP₂ to IP₃ and DG). The *ora* mutant has rhabdomeres that degenerate due to a rhodopsin defect. Because of the degeneration, *ora* lacks R1-6 rhabdomeres (the largest receptor subset). *pinhead* is an allele of *eya* and lacks compound eyes altogether, providing for a good control for testing the effects of visual mutants. Alterations in feeding (chronic vs. acute and vitamin A deprived (which eliminates opsin) vs. replete) and age were also used to dissect the turnover and incorporation process.

The phospholipids were labeled by feeding flies with ³²P. Organic extracts of heads broken off after freezing in liquid nitrogen were separated by 2 or 3 solvent system HPTLC. Spots on TLC plates were counted. The profile of labeling after 24 hr of feeding was similar for + and *norpA* and was approximately as follows: phosphatidylethanolamine=PE - 47%, PE plasmalogen=PE_{pl} - 1.5%, phosphatidylcholine=PC - 24%, PC_r [an unknown, likely PC, but with different fatty acids] - 4%, phosphatidylinositol=PI - 12%, lysophospholipids LPE - 2.5%, LPC - 1.6%, LPI - 1.4%, poly-PI's PIP₂ - 0.2%, PIP - 0.2%, phosphatidic acid=PA - 0.4%, phosphatidylserine=PS - 1.6% and the cardiolipins CL₁ - 2.5% and CL₂ - 1.1%.

A fraction of a percent of cerebroside is likely present. Relative to rat brain (Sun and Lin, 1989) the most notable differences were that in *Drosophila* heads PE is higher, PC is lower and PC_r is present, and there is probably no sphingomyelin. There are no major differences between *norpA* and + nor between light and dark. We compared acute with chronic feeding and found profound alteration in several major lipids (PI decreases, while PS, PC_r, Lysophospholipids and poly-PI's increase.) In carotenoid deprived flies, PS, PI and PE are higher while LPE and PC are lower. Aging increases PS, LPE and PC while decreasing PI and PE. A time course (3, 7, 16, 22 and 34 hours) showed major changes with lipids; PA and PI decrease, PS and PC increase, and the Cl's go down then up.

The fatty acids that compose PS, PI, PE and PC were analyzed by Gas Chromatography. The results showed that *Drosophila* fatty acids have the following distribution: 14:0 - 2%, 16:0 - 9 to 34%, 16:1 - 2 to 10%, 18:0 - 8 to 25%, 18:1 - 22 to 33%, 18:2 - 18 to 28% 18:3 - 1 to 15%. Triglycerides comprise around 70% of head lipids and have high 14:0 and 14:1. There are no fatty acids longer than 18 carbons in fly heads, but neither are there any in the food. Using medias which differed in fatty acid composition, it was shown that the fatty acid composition of the heads could be readily manipulated. We then supplemented the fly food with manhaden oil, a rich source of long chain fatty acids. We found that the ultrastructure of the photoreceptors was normal and that there was a small but significant increase in the electrophysiological sensitivity, especially in the UV. Although docosahexaenoic acid [22:6] is high in vertebrate photoreceptors and nervous system (Bazan, 1989), *Drosophila* heads had none. We confirm the finding of Yoshioka (1985) that PI lacks arachidonic acid [20:4], a precursor for many signalling molecules.