A Further Note on the 'Unmasking' of Lipids in the Cell

Since the author[1] reported on the 'Unmasking' effect of 1% phenol on the lipids in the oocytes of *Chrotogonus*, some more observations[2,3] have become available on the subject which prompt a further comment on the phenomenon of 'masking' in lipids.

BERENBAUM[2] has reported on the use of sudan black *B* in his 'Acetone-Sudan black', 'Burnt Sudan black', and 'Ethanolic Sudan black at 60°C' techniques for colouring the firmly bound lipids of the cell nuclei, reticulin and epithelial brush borders, etc. CLAYTON[3] has investigated the 'unmasking' influence of a number of lipid solvents on the acroblast in *Acheta domesticus*, and has found 90% ethanol and 5% hydroquinone to be the most effective 'unmasking' agents.

Both these papers deal with the techniques which 'unmask' lipids bound firmly with proteins to form lipoproteins. The influence of lipid solvents in breaking the bonds between lipoproteins, and consequently releasing the lipids, has been widely recognized by the biochemists[4]. In this laboratory also, the 'unmasking' effect of acetone and ethanol, especially on the lipoproteins of the mitochondria in oocytes, has very often been observed[5–7].

NATH, GUPTA *et al.*[5–7] have observed in the oocytes of a number of insects three kinds of lipid bodies viz. (1) L_1 bodies having a phospholipid or lipoprotein nature, (2) L_2 bodies having a phospholipid sheath surrounding a triglyceride core, and (3) L_3 bodies of a pure triglyceride nature.

The L_2 bodies show a duplex appearance with a completely sudanophobe core and a sudanophil cortex, in all the variants of sudan black *B*[8–10], even at 60°C. After a treatment in 1% phenol, either of the material or the gelatine sections, these L_2 bodies colour homogeneously in sudan black. Their duplex appearance is again restored after a simple treatment of the 'unmasked' sections in cold acetone for 12–24 h. The cores colour pink in nile blue[11] while the sheaths are blue. Further, the cores of the L_2 bodies are negative to acid hematein[12], Schultz's variants[13], mercuric-bromophenol blue[14] (for proteins), and PAS[15] (for carbohydrates). All these tests establish the presence of triglycerides in them.

Now, the triglycerides are not known to form lipoprotein complexes[16,17]. It is implicit, therefore, that some other phenomenon is involved which keeps the triglycerides in a 'masked' condition in such mixed lipid bodies, widely occurring in oocytes[7].

SCHMIDT[18] had suggested that the phospholipid spheres probably have a series of concentric shells of water in between the bimolecular layers of the phospholipid molecules. He has also drawn a 'water vacuole' surrounded by a bimolecular layer of phospholipids[19]. This observation has been fully confirmed recently by Ross and CHOU[19]. HIRSCH[20] and NATH[21] have often pointed out that such 'osmiophobic' parts of duplex vesicles act as the sites where cell secretions, including fat (triglycerides), are condensed. To the author, it appears that in the L_2 bodies mentioned above the triglyceride cores and the phospholipid sheaths remain separated by a thin layer of water molecules which form an impermeable membrane for the water-insoluble physical lipid colorants like sudan black *B* or sudan III and IV, etc. The ethanol or acetone of sudan solutions should be able to break the water barriers, but they dissolve away the triglycerides also in prolonged treatments and therefore are unable to reveal their 'unmasking' influence. Phenol, on the other hand, might attack the water molecules without destroying any lipids. It is interesting to note that in OsO_4 preparations like Lewitsky-saline[22] (unstained), these L_2 bodies always appear as homogeneously black spheres: but if such sections are bleached in H_2O_2 and then coloured with sudan black[22], the L_2 bodies again appear as rings[7,23].

It is clear, therefore, that in such cases as the L_2 bodies in oocytes, the 'masking' is a purely physical phenomenon involving technical difficulties in colouring. This would add another possibility to CIACCIO's[24] undefined[16] term of 'masked' lipids.

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Résumé

Les triglycérides se trouvant à l'intérieur de quelques corps lipides, dans les oocytes paraissent masqués lorsqu'on utilise des colorants pour lipides. C'est peut-être à cause de la présence d'une couche de molécules d'eau entre les molécules phospholipides de la gaine et les molécules des triglycérides du cœur. Cet écran interposé par l'eau est détruit d'une manière ou d'une autre par une solution d'1% de phénol et ainsi le «masque» disparaît.

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Intraspecific Sexual Preferences in *Drosophila prosaltans* Duda and in *Drosophila equinoxialis* Dobzhansky

By direct observation[1,2] of courtship and mating behaviour of neotropical strains of *Drosophila prosaltans* Duda

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