

tion of many alkaloids may be appreciably influenced by the presence of certain types of salts such as alkali carbonates.

Who knows but that the toxicity of caffeine may be influenced by the mineral matter of the diet and by the quantity of water the animal drinks?

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Catalogue of the Lepidoptera Phalaenæ in the British Museum; Catalogue of the Noctuidæ in the Collection of the British Museum. By SIR GEORGE F. HAMPSON, Bart. Volume X., 1910, Volume XI., 1912. London (England).

The two volumes comprise 2,140 species, in the subfamilies *Erastriniæ*, *Euteliinæ*, *Stictopteriniæ*, *Sarrothripinæ* and *Acontiinæ*. They are illustrated by two volumes of colored plates, Nos. CXLVIII. to CXIX., inclusive. There are also many cuts in the text illustrating typical species in each genus, giving both the general appearance and structural characters. Keys to the genera in each subfamily and to the species in each genus are given. There are also genealogical trees for each subfamily, showing the author's ideas of the evolution of the genera. The treatment is the same as in previous volumes of this work, which we have had occasion to notice. The genera are arranged upon adult structural characters, selected by the author. Resort has been had to many minor characters, such as modifications of the tuftings of the vestiture, tubercles on the front of the head, spines on the legs, etc. These characters are in many cases of little phylogenetic importance, so that the classification is to a large degree arbitrary and artificial. This appears distinctly in the arrangement of species within the genus also, where primary groups are made on modifications of antennal structure in one sex and other secondary sexual characters, so that really closely allied species are often widely separated. It would be rather difficult, however, to have avoided this and still keep the keys in a workable condition, especially where the vast majority of the early

stages and life histories are unknown, as is the case with these insects. The nomenclature of the North American species included in the book is greatly changed from that familiar to us. This appears to be unavoidable, as the classifications of different authors based on restricted faunal regions are here combined. The names here established will probably tend to be permanent, as it will be long before any one attempts to treat the Noctuidæ of the world on new lines with material equal to that afforded by the British Museum.

The British Museum collection, rich as it is, does not make a practise of retaining long series of specimens of common species. Consequently the author of these catalogues occasionally suffers from lack of sufficient material. We notice in the genus *Iscadia* (vol. XI., p. 362) some errors due to this cause. The subfamily *Sarrothripinæ*, to which *Iscadia* is assigned, is defined by the presence of a bar-shaped retinaculum on the fore wing of the male. In *Iscadia aperta* Walker and *I. duckinfieldia* Schaus this is absent. Sir George notes its absence in *I. aperta*, but having only one male he supposes it may have been broken off. Furthermore, *I. aperta* has simple flattened antennæ in the male, not bipectinate, as given in the table. *I. duckinfieldia* is abundantly distinct from *I. aperta*, not possibly an aberration, as suggested, for it has pectinated antennæ in the male and differs in markings, the double black line above the reniform-mark being absent. The separation of *I. duckinfieldia* on the brown costal shade is ineffective, as this shade is as often absent as present. These imperfections would have been obviated by larger series of specimens of these rather common species.

HARRISON G. DYAR

SPECIAL ARTICLES

THE NATURE OF THE FERTILIZATION MEMBRANE
OF THE EGG OF THE SEA URCHIN
(*ARBACIA PUNCTULATA*)

MANY widely held hypotheses, *e. g.*, on the dynamics of cell division, etc., are based on

the nature of the fertilization membrane of the sea urchin egg. The methods which have been used heretofore in the study of this so-called membrane have been inadequate to determine its exact nature. The writer has made some observations, by the use of methods which are apparently new to this field, which seem to throw a new light on the structure of the various coatings on this egg. As is well known, there has been no agreement among cytologists concerning the number and nature of the coverings of this egg. A few investigators have recognized the presence of a thick jelly surrounding the egg, but have given us no methods of demonstrating its true extent or nature. Further, no one has used methods by which the physical characteristics of the vitelline membrane could be determined in the living egg, and the relation of this membrane to the cytoplasm on the one hand and to the egg-jelly on the other.

The normal unfertilized egg is covered by a soft invisible jelly about 23 microns in thickness. Beneath this and closely stuck to the surface of the cytoplasm is a tough somewhat elastic vitelline membrane. Morphologically, this latter is the only membrane on the egg of *Arbacia*. It is literally glued to the outer surface of the cytoplasm. The average of a number of measurements shows that the vitelline membrane is about 1.9 microns in thickness. In the living egg the inner part of this structure is seen as a light line on the outer surface of the cytoplasm when the light is sufficiently stopped down. The refractive index of the outer part is so nearly that of sea water that this portion is invisible.

In the reaction of the egg to the spermatozoon, striking morphological changes occur in the vitelline membrane and the surface of the cytoplasm. The change in form of the egg-jelly is slight. About one and one half minutes after active spermatozoa are mixed with the eggs, a definite swelling of the vitelline membrane occurs. The extent and location of the swelling varies greatly in different eggs. In some eggs the swelling is complete in one minute, but three to five minutes are usually required for the maximum swelling of

this structure. During the swelling of the vitelline membrane its refractive index is so changed that it usually becomes distinctly visible by the usual microscopical examination. When swelling is complete the thickest portion of this structure frequently measures as much as nine microns. About six minutes after insemination of the eggs the surface of the cytoplasm swells and changes its refractive index. In one minute the swelling is complete and measures about one micron in thickness. This is the well-known hyaline plasma-layer. By the time the swelling of the vitelline membrane has become well advanced, a change occurs in the refractive index of the inner part of the egg-jelly and this becomes visible. As seen by the usual microscopical examination the so-called fertilization membrane of the egg of *Arbacia* consists of three parts, viz., the inner part of the egg-jelly which has undergone a change in its refractive index, the swollen vitelline membrane and the thin highly refractive surface layer of the cytoplasm. This hyaline layer is still very adherent to the vitelline membrane. The edematous vitelline membrane is softer and more elastic than it is in the unfertilized egg. If this structure is partially dissected from the fertilized egg it frequently contracts to a glutinous mass on one side of the cytoplasm. The relation of the hyaline plasma-layer to the vitelline membrane and the cytoplasm is brought out very clearly in fertilized eggs which have been plasmolyzed by adding cane sugar to sea water. The protoplasm shrinks and the hyaline layer frequently takes on the appearance of pseudopodial-like processes of the cytoplasm. When the vitelline membrane is dissected from the egg, the hyaline layer remains as an organic part of the cytoplasm. These methods leave no doubt as to the nature of this structure; it is the swollen surface of the cytoplasm and enters into the formation of the larval sea urchin. The water-holding power of the hyaline layer and of the edematous vitelline membrane is striking. These structures do not show an appreciable shrinkage in quite concentrated solutions of cane sugar in sea water.

Three methods have been used singly and in various combinations in this study. By the plasmolytic method a separation of the hyaline plasma-layer from the vitelline membrane is easily affected. Vital staining differentiates clearly the various structures on the surface of the egg. Janus Green (dimethylsafraninazodimethylalanin) in dilute solutions stains the egg-jelly light blue. It is also beautifully demonstrated by a number of other vital stains. In concentrated solutions of janus green the jelly shrinks to a mere hull. Slightly concentrated solutions of isamin blue, dissolved in sea water by boiling, stain the swollen vitelline membrane a deep blue while the hyaline layer is much lighter in color. Toluidin blue stains the hyaline layer of the cytoplasm and the vitelline membrane, but as a differential stain it does not equal isamin blue.

The removal of the egg-jelly and vitelline membrane from the fertilized and unfertilized eggs was affected by dissection with glass needles made from very hard Jena glass tubing about 5 mm. in diameter. The points on many of these needles measured less than one half micron. The needles were held in a Barber pipette-holder and the dissections made under a magnification of five hundred and sixty-two diameters.

It seems that the type of reaction described for the egg of *Arbacia* is a somewhat common one, since essentially the same changes occur in the eggs of *Chaetopterus* and the mollusk *Cumingia*. In these two forms the maximum swelling of the vitelline membrane does not occur until about twenty to thirty minutes after insemination of the eggs.

An analysis of the reaction of the egg of *Arbacia* to the spermatozoon has been attempted. Puncture of the vitelline membrane has failed to produce the reaction. Doses of from one to five spermatozoa have been injected into the egg-jelly and the relation between the time required for the penetration of the vitelline membrane by the spermatozoon and the extent and location of the swelling of this structure have been studied. By injecting

spermatozoa into the egg-jelly, in a small percentage of cases a single spermatozoon becomes attached to the vitelline membrane and produces the reaction that has been described. The passage of the spermatozoon through the vitelline membrane has been observed in a number of eggs. It has been found possible to remove the spermatozoon from the vitelline membrane after it has caused the reaction. The real difficulty in this type of experiment is not the size of the spermatozoon, but the fact that when even four or five spermatozoa are injected into the egg-jelly they usually swim out and away from the egg. This necessitates the making of many injections in order to get a single spermatozoon to attach itself to the vitelline membrane and start the reaction.

As far as my evidence goes at the present time it seems that the primary function of the much discussed reaction of the egg of *Arbacia* to the spermatozoon is the prevention of polyspermy.

The details of this study will appear later.

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A SIMPLE METHOD OF MAKING ARTIFICIAL CELLS
RESEMBLING SEA URCHIN EGGS IN CERTAIN
OF THEIR PHYSICAL PROPERTIES

SEVERAL years ago Robertson showed that if chloroform was shaken with egg-albumen solution, the droplets would not reunite even when washed in water, because of the formation of a proteid film on the chloroform surface. It can be readily observed that such droplets shrink in volume, owing to the passage of chloroform into the water outside.

While studying the penetration of alkalies into lecithin in various solvents, I noticed that if lecithin is dissolved in chloroform and the solution shaken with proteid solutions, the chloroform of the resultant globules is in time completely replaced by water. Eventually, then, instead of lecithin in chloroform, we may obtain small cells of lecithin in water sur-