

gegen Bisse in isolierende mit Wasser gefüllte Gefäße und daß eine solche Jagd, wenn günstig, 12—20 Liter Iças ergebe.

Es ist sodann zu erwähnen, daß es beliebter Scherz ist, solche Iças wie Puppen zu bekleiden. Auf diese Weise mögen oft Iças von Passagieren mitgenommen und unfreiwillig verschleppt werden. Ein zweiter Punct des Küstengebietes von S. Paulo, wo die Sauva seit Kurzem vorkommt, ist Bairro de S. Francisco, nahe bei S. Sebastião. Hier scheint die Verbreitung eine natürliche vom Innern des Staates her erfolgte gewesen zu sein. Jedenfalls datiert auch da das Auftreten der Sauvas erst seit 5—6 Jahren. Sie werden nun successive das Küstengebiet des Staates in Besitz nehmen. Wenn wir also hier wie in Rio Grande do Sul ein stetes Vordringen der *Atta sexdens* vom Westen zum Küstengebiet hin beobachten können, so dürfen wir wohl auch rückwärts schließend annehmen, daß auch die jetzt von den Sauvas eingenommenen Gebiete Südost-Brasiliens erst relativ spät successive von ihnen in Besitz genommen worden sind.

São Paulo (Brasil), 25. Jan. 1895.

### 3. On the Maturation of the Pelagic Eggs of Teleostean Fishes.

By T. Wemyss Fulton, M. D., F. R. S.E. Scientific Superintendent, Fishery Board for Scotland.

eingeg. 3. März 1895.

It is well known that the mass of the ovaries of an osseous fish, whose eggs are pelagic, is, just before spawning begins, made up for the most part of opaque whitish eggs nearly uniform in size; and that when spawning has commenced other eggs are found scattered throughout the substance of the organ, conspicuous by their clear, glassy appearance and larger size. Some years ago in investigating the fecundity of certain species, I was impressed by the differences between the two kinds of eggs, and the rarity of intermediate stages, and I then stated that the change seemed "to be due to sudden accession of fluid from the ovarian follicles, which increases the bulk of the ovum and renders the opaque contents clear by dilution"<sup>1</sup>.

Further investigation has shown this view to be correct, and that the final change in the maturation of the ovarian egg of Teleostean fishes is associated with the entrance of watery fluid, which dissolves the germinal vesicle and renders the yolk clearer. In demersal eggs (such as those of *Clupea harengus*, *Cyclopterus lumpus*, *Cottus scorpius*, *Lophius piscatorius*, etc.) the change in size and appearance is less marked,

<sup>1</sup> »The Comparative Fecundity of Sea Fishes«. Ninth Annual Report, Fishery Board for Scotland. Part III. p. 247. 1891.

although quite distinct; the ovum becomes more pellucid, the yolk-spherules more separated, perhaps in part dissolved, and the germinal vesicle disappears. In pelagic eggs the process is carried further. The yolk-spherules are entirely dissolved, as is also the interstitial protoplasm (except in certain cases where strands remain between masses of fluid yolk); the egg is distended to three or four times its previous volume, the zona radiata being stretched and thinned out; the contents are rendered of crystalline transparency and the specific gravity is reduced so that the egg floats in ordinary sea water. The egg is at the same time freed from the follicle.

The detailed results of the investigation, which was made on a large number of species, will appear in the forthcoming Report of the Fishery Board for Scotland, but the chief features may be stated here.

The opaque ovarian eggs immediately before the change referred to occurs have the following structure, as shown by examination in the living condition and in mounted sections. The egg-capsule is soft and elastic, its thickness varies in different species but is greater than in the mature state; usually homogeneous in section, or indistinctly fibrillated, it may show distinct fine radial striation (cg. *Solea vulgaris*, *S. lutea*). On surface view two patterns are usually exhibited (1) very fine, minute punctation or dotting (2) a much coarser wrinkled, mesh-work or basket-work appearance. The former which may simulate striation in optical section, or the opening of pores on surface view, is limited to a delicate superficial layer, whose presence may sometimes be demonstrated by the rupture of the subjacent zona under pressure, the escaping yolk bulging it and stripping it off. In macerated mature eggs it may separate spontaneously as a delicate film. There is also within the zona an extremely delicate transparent homogeneous membrane, closely applied to the vitellus, such as has been described by Vogt in the demersal eggs of *Coregonus palea* and *Salmo umbla*<sup>2</sup> by Ransom in those of the Stickleback<sup>3</sup> and by Oellacher in those of the trout<sup>4</sup>. Scharff also found it in sections of the eggs of *Trigla gurnadus*<sup>5</sup>; but its existence as a distinct membrane is usually denied. Its presence may occasionally be revealed by pressure on the fresh ovum. Usually it is ruptured with the capsule, but sometimes it remains intact, and may then be seen as a delicate follicle bounding and retaining the protruding yolk, and emitting it with a rush when the pressure is increased, or, when the pressure is diminished and the yolk-

<sup>2</sup> Embryologie des Salmones p. 29.

<sup>3</sup> Phil. Trans. Vol. 157. p. 433.

<sup>4</sup> Zeitschr. f. wiss. Zool. Bd. XXII. Hft. 4. 1872.

<sup>5</sup> Quart. Journ. Micr. Sc. Vol. XXVIII. p. 53.

spherules are sucked back it may remain protruding from the rupture. In other cases it may be traced within the zona. In the living fertilised eggs of *Pleuronectes flesus* I was able to trace it up over the edge of the segmenting blastodisc; in some, after death, when the cortical protoplasm began to retract, the delicate membrane was carried with it and was thrown into wrinkles in the neighbourhood of the blastodisc. The membrane therefore continues to cover the vitellus after the perivitelline space is formed.

Immediately beneath this membrane is the thin cortical layer of protoplasm or periblast, most readily seen in mounted sections, with the minute shining granules or vesicles imbedded in it. These resist stains, dilute acetic acid, alcohol and ether; they persist in the mature eggs for a long time after death, even when the cortical layer has shrunk and partially disintegrated; and (apart from oil globules when these are present) they are the only definite bodies which can be recognised to continue unchanged within the pelagic egg throughout the stage of maturation. The cortical protoplasm is continuous internally with the scanty matrix between the yolk-spherules which make up the great mass of the egg. The yolk-spherules in pelagic eggs differ from those in demersal eggs in being smaller and less dense. The largest measure about 0,02 mm; they may be clear or may contain fine granules; after treatment with osmic acid they show concentric lamination. In demersal eggs the spherules may reach 0,05 mm (*Zeus faber*) or 0,07 mm (*Gobius niger*, *Lophius piscatorius*); they are markedly granular or may be made up of solid-looking refringent bodies, presenting sometimes a mulberry-like appearance. In eggs possessing in the mature condition one or more oil globules, numerous oil-droplets are arranged around the germinal vesicle, which fuse together and collect on the upper surface of the yolk when the watery fluid enters.

The germinal vesicle is central, typically spherical, and relatively large, occupying from about one-fourth to nearly one-third of the diameter of the ovum. It can readily be isolated from the living egg; it is soft and plastic, with a distinct membrane, finely dotted on the surface (probably from rupture of threads of linin) and it contains numerous, conspicuous, refringent nucleoli, arranged more or less regularly around the periphery, and which stain deeply. The chromatin network cannot well be made out except in the young eggs.

The structure of the fully matured pelagic egg, ready for fertilisation, is simple. It consists of the envelopes described, the cortical layer of protoplasm (which may in some forms become aggregated at one pole to form the blastodisc) and a homogeneous transparent fluid

yolk. The germinal vesicle, nucleoli, and yolk-spherules have disappeared, and the egg is swollen to three or four times its former volume. The exact changes that occur in the germinal vesicle have not been determined satisfactorily. The elimination of two polar bodies from unfertilised pelagic eggs has been described by Cunningham and Kingsley, while Agassiz and Whitman carefully describe the elimination of two polar bodies shortly after fertilisation. Kingsley admits the possibility of sperms having been present; and there seems little doubt that the polar bodies are not formed until after fertilisation. But I have found no trace of the germinal vesicle or of its contents in the translucent mature egg. The membrane and nucleoli are dissolved and the karyoplasm diffused, and although the chromatic substance must be transferred to the cortical protoplasm near the micropyle I have not been able to detect it. This phase in the ovarian development of the pelagic egg is a striking one, marking the conclusion of growth and the preparation for fertilisation.

That it is due to the comparatively rapid imbibition of watery fluid is shown by the following circumstances.

1) The rarity of eggs between the two stages described. Many hundred may be examined before intermediate stages are found: these are usually in the opaque condition, but somewhat larger and clearer and they contain the germinal vesicle.

2) The great difference in the volume of the largest opaque and the mature translucent eggs. Thus the mean diameter of the largest opaque eggs of the plaice (*P. platessa*) just before the change occurs is about 1,21 mm and the mean diameter of the translucent eggs is about 1,88 mm (see Figure); the respective volumes are therefore 0,9275 cubic millimetres and 2,479 cubic millimetres — nearly a fourfold increase. So likewise the increase in the volume of the ovum of the haddock (*Gadus aeglefinus*) is from 0,5236 cmm to 1,5962 cmm; in the cod (*G. morrhua*) 0,3817 cmm to 1,3706 cmm; in the turbot (*Rhombus maximus*) from 0,1795 cmm to 0,5556 cmm; in the halibut (*Hippoglossus vulgaris*) from 8,18 cmm to 28,732 cmm; and so forth. In as much as the eggs, both opaque and translucent, vary a little in size, and are not perfect spheres, these measurements are necessarily approximate; but they are founded on numerous measurements, and they clearly indicate the great increase in volume which occurs. In demersal eggs the augmentation of the size is much less. The large opaque eggs of the herring (*Clupea harengus*) for example, which still exhibit the normal germinal vesicle, have an average diameter of about 1,18 mm and a volume of 0,8602 cmm; while the mature egg, in which



the germinal vesicle has vanished, has an average diameter of about 1,33 mm and a volume of 1,2318 cmm.

3) The egg-capsule becomes thinner, and the pattern on the surface stretched out, from the mechanical distension of the ovum. Thus the thickness of the zona of the opaque egg of *P. platessa* is about 0,054 mm; that of the translucent egg 0,025 mm. On the zona of the opaque egg of *Brosimius brosme* seven or eight punctations lie between two lines 0,05 mm apart, and only two or three in the same space in the mature egg. The thinning of the capsule is however less than calculation shows should occur in the stretching of a homogeneous elastic membrane to the same extent, a circumstance accounted for by its structure.

Fig. A.

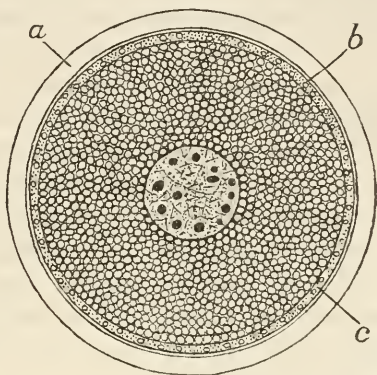


Fig. B.

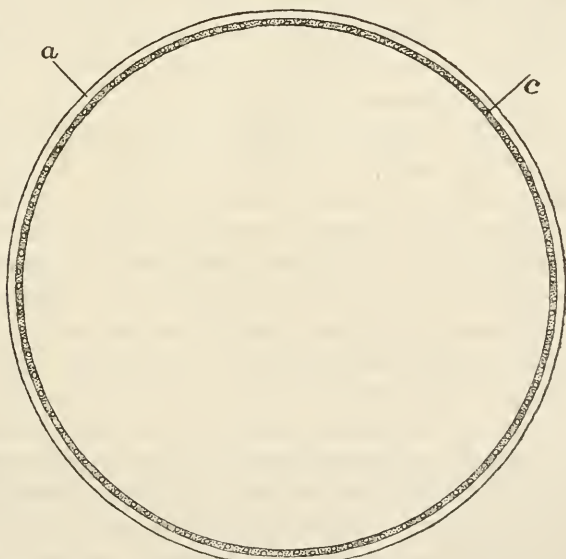


Fig. A, B. Sections of the large opaque and of the mature translucent eggs of the plaice (*Pleuronectes platessa*)  $\times 40$  (slightly diagrammatic). *a* zona radiata; *b*, internal vitelline membrane; *c*, cortical protoplasm. Diameter of *A* 1,21 mm, specific gravity 1,070; diameter of *B* 1,88 mm, specific gravity 1,025.

4) By the diminished specific gravity. The opaque eggs of all species sink in sea-water, while the mature pelagic float. For example,

the large opaque eggs of *P. platessa* have a specific gravity of about 1,070 while that of the translucent eggs ranges from 1,025 to 1,026. Considerable variation occurs in the specific gravity, as in the size, of mature eggs of the same species, or even the same individual. Empty capsules of mature pelagic eggs sink in sea-water; and when a pelagic egg dies it sinks owing to the fluid yolk diffusing out in consequence of the death and retraction of the periblast.

5) The action of heat and re-agents — the vitellus of the opaque egg becomes solidified and chalky white, that of the mature pelagic egg remains fluid and only becomes slightly milky or opalescent.

6) The proportion of water present. — Dr. T. H. Milroy, of the University of Edinburgh, who is making for the Scottish Fishery Board a detailed investigation of the chemical changes that occur in the growth and maturation of the eggs of Teleosteans, finds for instance that in the mature buoyant eggs of *P. platessa*, dried to a constant weight at 60° C., at 80° C., at 100° C., and finally at 105° C., the percentage of water present is the very high one of 91,86, while in the opaque eggs it is only 65,5.

The precise nature of the fluid which enters the eggs is being as far as possible ascertained by Dr. Milroy. It seems to be secreted by the granular layer of the follicle at the close of intraovarian growth, and it probably enters by endosmosis, but the immediate exciting cause has still to be determined. Its composition probably corresponds nearly to that of the ovarian fluid which is found bathing the ripe eggs in the cavity of the ovary, and which is expelled with them when they are shed. Very little appears to be known about normal ovarian fluids in any group (vide Hammarsten, Lehrbuch der Physiolog. Chemie, p. 364 [1895]). In Teleosteans with demersal eggs the ovarian fluid sometimes acts as a cementing substance, but in those with pelagic eggs it is watery, mingles with the sea-water and disappears, subserving only a lubricating function in spawning. Hensen<sup>6</sup> has described the free ovarian fluid of *Gadus morrhua* as having a specific gravity of 1,0115 at a temperature of 8,7° C., as being alkaline in reaction and containing an albuminoid; he regards it as probably originating in Graaf's follicles, and he found the quantity to range between 14 and 20 per cent of the total volume of eggs and fluid discharged. The quantity of free fluid present in immature ovaries is very small; it increases greatly when spawning begins, and the proportion is largest in ovaries nearly spent. In *P. platessa* 517 cc of eggs and fluid, gently pressed from ripe females and drained through silk-netting, furnished

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<sup>6</sup> Vierter Bericht d. Komm. z. wiss. Untersuch. d. deutschen Meere. 1884.

51,5 cc of very pale yellow almost transparent fluid, which was found by Dr. Milroy to possess a specific gravity of 1,0116, a practically neutral reaction, and to contain a large amount of chlorides, phosphates being absent; the amount of chloride of sodium present per 100 ccm was 0,766 grammes, the total albumins being 0,838 grammes; no mucin or pseudomucin was detected. It may be stated that opaque eggs placed in weak solution of common salt clear up considerably, so as to show the germinal vesicle, which persists.

It thus appears that the final stage in the development of the ovarian eggs of Teleosteans is characterised by the comparatively rapid imbibition of a watery, and probably saline, fluid of low density, which seems to be secreted by the follicle, and is associated with the dissolution of the germinal vesicle and the rearrangement of the chromatin. In eggs which become pelagic the volume of fluid imbibed is relatively very large, dissolving also the yolk-spherules and reducing the specific gravity of the whole ovum below that of sea-water.

It is in virtue of this change that the eggs of certain marine species become buoyant or pelagic (those of fresh-water species could not by this device be enabled to float); but in as much as it also occurs in demersal eggs it is clear that its signification lies deeper. It is probable that not only among Teleosteans but among some other groups the apparent vanishing of the germinal vesicle and the rearrangement of chromatin is due to the absorption of fluid from without, and that this is the primary and essential meaning of the process. In certain marine Teleosteans Natural Selection has, so to speak, seized upon and exaggerated a normal phenomenon for another purpose, viz. to enable the eggs to float and be dispersed. The same genus may comprise closely-allied species whose eggs are respectively pelagic and demersal.

The fact that the yolk of pelagic eggs contain so large a proportion of water explains some other phenomena, such as the situation of the heavier blastodisc at the lower pole, the more rapid and less perfect embryonic development, the absence of vitelline circulation and the simple manner in which the yolk is absorbed. The initial nutritive value of a unit of yolk is less in a pelagic than in a demersal egg, owing to the less condensed, or more fluid, character of the spherules; but the nutritive value is further greatly diminished by the dilution of the yolk with three or four times its volume of water. For example, the demersal egg of *Cottus scorpius* is of about

the same size as the pelagic egg of *P. platessa*: the volume of yolk indeed is somewhat less. The former takes between three and four weeks to hatch, the larva is about 7,5 mm in length, has the mouth open, and is well developed; at the same mean temperature the latter hatches in about 16 days, the larva is 4,1 mm long, the mouth is not formed, and it is ill-developed.

It also explains the gradual, prolonged spawning. From the final distension of the ovum, the female is unable to carry simultaneously all the eggs in the mature condition; but the expansion takes place sporadically throughout the ovary, a comparatively small number undergoing the change and being expelled at one time. The gross volume of the eggs shed in one season may considerably exceed the volume of the body of the female. Thus a large flounder (*P. flesus*) examined a little before the spawning season had a volume of 951 ccm, the ovaries occupying 170 ccm and the body 781 ccm; the number of opaque eggs present was computed to be 2,733,800, with a volume in the mature condition of about 1114 ccm, of which nearly two-thirds is water.

Edinburgh, February 28th 1898.

#### 4. Neue Turbellarien der Bucht von Concarneau (Finistère).

(Vorläufige Mittheilung.)

Von Dr. O. Fuhrmann, Académie, Neuchâtel.

eingeg. 8. März 1897.

Während eines Aufenthaltes an der Zoologischen Station von Concarneau beschäftigte ich mich speciell mit der überaus reichen Turbellarienfauna der so thierreichen Meeresbucht. Ich fand 29 Arten, worunter 5 neu sind. Damit ist aber die Zahl der Turbellarienarten noch nicht erschöpft, denn es kamen mir noch mehrere Formen zu Gesicht, die ich wegen Mangel an Material oder aus anderen Gründen nicht bestimmen konnte.

Die gefundenen sind folgende:

*Proporus venosus* O. Sch.; *Convoluta flavibacillum* Jens.; *C. paradoxa* Oe.; *Macrostoma hystrix* Oe.; *Macrostoma rubromaculatum* Graff; *Macrostoma lucidum* nov. spec.; *Stenostoma Sieboldii* Graff; *Promesostoma marmoratum* Graff; *P. ovoideum* Graff; *Proxenetes flabellifer* Jens.; *P. gracilis* Graff; *Acorrhynchus caledonicus* Graff; *Macrorhynchus helgolandicus* Graff; *M. Nageli* Graff; *Macrorhynchus coeruleus* nov. spec.; *Hyporhynchus setigerus* Graff; *H. penicillatus* Graff; *Provortex balticus* Graff; *Plagiostoma Fabrei* nov. spec., *Plagiostoma violaceum* nov. spec., *P. dioicum* Graff, *P. Girardi* Graff, var. *major* Böhmig, *P. vittatum*



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