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The present module is from a Developmental biology paper from Bachelor of Science, Third Year. The unit is an introduction to developmental biology and the module name is Activation and Amphimixis. The outline of this module is first, there will be an approach of spermatozoa to egg will be explained followed by the activation of egg and finally the Amphimixis.

And the learning outcome expected after the end of the module is, the student will be able to understand the approach of spermatozoa to the Ovum during the process of fertilization. Students should be able to know how the activation process occurs during fertilization, and thus students will be able to learn the concept of Amphimixis.

To introduce the activation and amphimixis during the process of fertilization. As we have already seen in the previous module. Fertilization is nothing but the Fusion of two gametes from male and female. The fertilization triggers the developmental program. And the moment of activation saves the life of an egg because at the time of fertilization as the sperm approaches the egg, the egg gets activated so that it can be further developed into a cyclic process of fertilization finally accomplishes activation followed by or amphimixis. And amphimixis is nothing but the Fusion of the two nuclei. The approach of spermatozoon to the egg is one of the important steps in the formation or in the process of activation of fertilization. First and foremost, let us see how the approach of spermatozoa to the egg takes place as we have already mentioned, there are two important types of fertilization. One is the external and the other one is the internal. In external fertilization the eggs and the sperm are shed in the external environment, specially the aquatic environment and it becomes a great challenge for this gametes to approach to its species specific egg. So therefore this has been taken place by two important strategies. One is by means of chemo taxis. The other one by means of Agglutination or also known as fertilizin, anti fertilizin interaction, which has been explained by two different methods. One is antigen antibody type of interaction and the other one is key and lock type of interaction. To begin with, chemotaxis,
Chemotaxis is the ability of the sperm to detect a difference in concentration of some chemicals or the substance released into the water to move from an area of lower concentration of this chemical to
the greater of that substance, and they Orient and swim towards a putative attractant, and this has been observed in case of coelenterates, tunicates and chitons.

The Chorion or envelope around the egg is usually perforated by narrow canals called micropyles. In certain aquatic Organism animals through which the sperm finds its way to enter inside the egg cytoplasm and it has been observed in fish egg examples, Sturgeon, even in case of lampreys where each of the eggs is enclosed in hard chorion, you can see a structure over here.

This is the micropyle opening which leads to the opening into the egg cytoplasm and there are chemicals found in this region which have been detected by the sperm and it enters through that opening.
This is a structure of a micropyle in the egg of some teleost fish, that is Sturgeon, that allows the sperm entry fast through the outer covering. Another way by which a spermatozoon approaches the eggs by

means of agglutination or the fertilizin and anti fertilizing interactions, eggs release into the surrounding medium.
Substances that bring about agglutination of the sperm and the approach of the sperm to the egg is the main concept.
In this type of interaction the factor, called fertilizin, came from the egg Jelly. Which is coated and gets dissolved as the egg shed in sea water and for sperms to be agglutinated by this fertilizin, they have a receptor site on their surface. Which is called the anti fertilizin and the binding of fertilizin with the anti fertilizin which is very species Specific leads to the process of Fusion of the gametes. The anti fertilizin have been extracted from the spermatozoa by heating, freezing or throwing or acidifying the water in the laboratory. This is a structure depicting the fertilizin-anti fertilizing reaction and which was studied in sea urchin.

You can see the sperm having a receptor site, which is binding with a fertilizing molecule from the egg.
This is a particle of the fertilizin.

Now, regarding the mode of fertilizin interaction of fertilizin and antifertilizin Balansky in 1981 has forwarded the following two analog mechanisms. That one it could be because of the antigen antibody type of interaction, wherein the fertilizing in the egg membranes behave as an antibody, combining with the antigenic site that is the anti fertilizin on this sperm surface.
Another analogy used is the key and lock type of interaction. Wherein or this same recognition has been seen. And this is because an addition depends on the bonding that happens between the fertilizin and the anti fertilizin or complementary molecules. So as this permit sperm approaches the egg, there is activation of the egg that takes place, so the process of initiating development in an egg is usually known as activation, and which is initiated by this form. So during activation there are certain changes that take place in the egg. Such as the surface of the egg forming the fertilisation Cone. The vitelline membrane, which is a membrane surrounding the plasma membrane of the egg, gets lifted up and is converted into another important membrane called fertilization membrane. The cortical granules which are found inside the peripheral region just below the plasma membrane of the egg explode, releasing certain contents and the cytoplasm exhibits movement. The permeability of the plasma membrane increases so that certain ions get entry. Just changing the voltage of the membrane. The coenzyme such as NAD is phosphorylated. The rate of protein synthesis increases, mitosis gets initiated.The breakdown of polysaccharide occurs and the enzyme dehydrogenase also increases.

Many theories have been proposed to explain the activation of eggs by sperm by various researchers. For example, the theory of Boveri in (1887, 1895), which says that the mature egg has no division center. This means there is no centriole which is introduced by sperm and thus the egg gets activated to divide. Another theory by Loeb in 1913 says that sperm brings in the cortical cytolysis, which leads to the sudden increase in the oxidation process of the egg. The theory of Battaillon in (1910 to 1916) says tha fertilization leads to release of certain substances to the eggs exterior, which leads to the accumulation of perivitelline fluid and an elevation of fertilization membrane. Theory of fertilizin-anti fertilizin by F.R., Lillie, where the egg is activated by the fertilizin anti- fertilizin reaction. Another theory is changing the viscosity theory, which was postulated by Heilbrunn, where in the activation leads to calcium release in the egg cytoplasm. thus,

increasing the viscosity and initiates the development. The theory of Tyler in 1967 says that protein synthesis is initiated immediately after fertilization and the dissolution of the masking protein is brought about by an enzyme liberated in egg by spermatozoon and theory of the inhibition by Renstrom and Brackett says that the maturation of the metabolic inhibitors accumulate in the oocyte and this is eliminated and the egg is deinhibited.

Amphimixis is a process wherein there is a fusion of male and the female pronuclei. In sea Urchin and in all vertebrates, the two nuclei come in contact. The nuclear membrane at the point of contact disappears and the contents of the nuclei Unite into one mass surrounded by a common nuclear membrane. Please remember these are the two different gametes, one from the male one from the female having its own nuclear membrane, which disappears at the point of contact, and the nucleus unite into one and surrounded by a common nuclear membrane to perform the act of amphimixis. The sperm nucleus has to perform two activities, one it has to become pronuclei and one has to migrate from the site of fertilization to the site of amphimixis. Before Amphimixis the nucleus of the egg also undergoes certain changes like the sperm nucleus. After the completion of the second meiotic division. The haploid nucleus of the egg occurs near the surface of the egg in the form of several vesicles known as karyomeres, and in fertilizing the egg. These karyomeres fuse to form a female pronucleus. An increase in volume becomes vesicular. It also migrates towards the sight of amphimixis.

Thus, the process of activation leads to an amphimixis and the fertilization gets over, but at the same time there are certain biochemical changes that take place which we will be studying in the next module.

These are the references that have been used to prepare this module.

Thank you.