

**Ultrastructure of the Chorion and its Micropyle  
Apparatus in the Mature Discus  
(*Symphysodon* spp.) Eggs**

**Olgun Diskus (*Symphysodon* spp.) Yumurtalarında  
Chorion ve Mikrofil'in Ultrastrüktürel Yapısı**

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**Abstract**

The surface ultrastructure of discus (*Symphysodon* spp.) eggs was investigated by a scanning electron microscopy technique. The telolecital, oval and pale yellow colored mature discus eggs were possessed a micropyle and consisted of relatively smooth-sided, funnel-shaped vestibule at the bottom. The outer surface of the eggs were covered with pile of short fibrils.

**Keywords:** Ultrastructure, Chorion, Micropyle, *Symphysodon* spp.

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**Introduction**

In egg laying fish the reproductive materials form in the gonads of the fish and the materials have to be transported to the outside of the body either as an egg or a spermatozoa (Brown, 1957; Franchi et al., 1962; Ball, 1960; Demir, 1992; Lagler et al., 1962).

Great majority of teleost fishes are oviparous, therefore fertilization and larval development occurs externally in those fishes (Franchi, 1962; Hoar, 1962).

Fish eggs are either pelagic (buoyant eggs) or demersal character so, sticky demersal eggs are adhere to any suitable surface in the water (Baran and Timur 1983). Fish egg cell has a tender membrane when ready to leave the ovary. As the eggs of oviparous species, pass through the oviduct, they go through the glands which secretes the shells about them (Bond, 1979). This secreted layer is called the chorion (*Zona radiata*) or primary envelope which contains a single micropyle opening (Franchi et al., 1962; Lockwood, 1972; Hibiya, 1982). The chorion is a major protective coat of an ovum, and the outer surface of the chorion is covered with a jelly sticky fibrils in some fishes (Ginzberg, 1972).

The presence or absence of this layer and its structure in variety of teleost fish have been the source of controversy (Boyd and Simmonds 1974; Shanklin, 1959). Most of the problems arisen from light microscopic studies which is unable to detect the plasma membrane or a jelly coat.

Shanklin and Armstrong (1952) described the ultrastructural formation of the chorion externum, with numerous pores, and protein fibrils in the *Fundulus heteroclitus* (Shanklin and Armstrong 1952).

Ultrastructural studies of Kaighn (1964) found that four to five layered chorion in the *Fundulus heteroclitus* with 0.56  $\mu\text{m}$  pores (Kaighn, 1964).

Limited number of fine structural studies were existing on a mature teleost ova (Hurley and Fisher 1966; Hagstrom and Lönning 1968; Yamamoto and Onozato 1965), and those do not shedding any light on the condition of the surface of the discus (*Symphysodon* spp.) eggs.

Purpose of this study was to obtain background information in the mature discus eggs, particularly the structure associated with fertilization phenomena.

## Materials and Methods

In this study two adult discus fish were brought from a professional aquarist, and maintained at 26-28 °C in an aquaria (Altinköprü and Altinköprü 1976). The fish laid their eggs on the surface of a flower-pot which was put in the aquarium previously. The fertilized eggs were then picked from the surface of the pot (Figure 1) (Köhler, 1998).



Figure 1. Fertilized eggs are on the surface of the flower-pot

Freshly obtained fertilized eggs were fixed in 0.25 % glutaraldehyde for 2 hours at 4 °C, and then rinsed with 0.1 M phosphate buffer solution (PBS) for 1/2 hour at pH 7.4, and post fixed in the 2 % osmium tetroxide (OsO<sub>4</sub>). The samples were washed by PBS for 1/2 hour, and treated by the mixture of various graded alcohols and amyl-acetate (Culling, 1963; Roberts et al., 1971). Eventually to be dried at 40 °C by the treatment of CO<sub>2</sub>. The egg samples either as a whole or a divided into two pieces were stucked on the grids and covered by carbon and gold. Cutting prosedure of the eggs was done by means of a razor blade under a stereo microscope. The samples were studied in a JSM-5400 scanning electron microscope (Henrikson and Matoltsy 1968; Postek et al., 1980).

### Results

Under the scanning electron microscope it was observed that the fertilized eggs of a discus fish have an advanced telolecital, oval, and pale yellow colored ova with a thick wrinkled shell.

The eggs have had a long axis of  $0.880 \pm 0.080$  mm; and the shorter axis of  $0.680 \pm 0.040$  mm. Counted wet weight was  $0.0020 \pm 0.0002$  g.

The outer surface of the ovum was covered by a jell-like coat, comprising of a short fibrils (Figure 2). Several thick and longer fibrils were present at the periferal area of the micropyle (Figure 3).

The micropyle apparatus was consisting of relatively smooth-sided, funnel-shaped vestibule at the bottom and the canal sides were quite irregular (Figure 4).

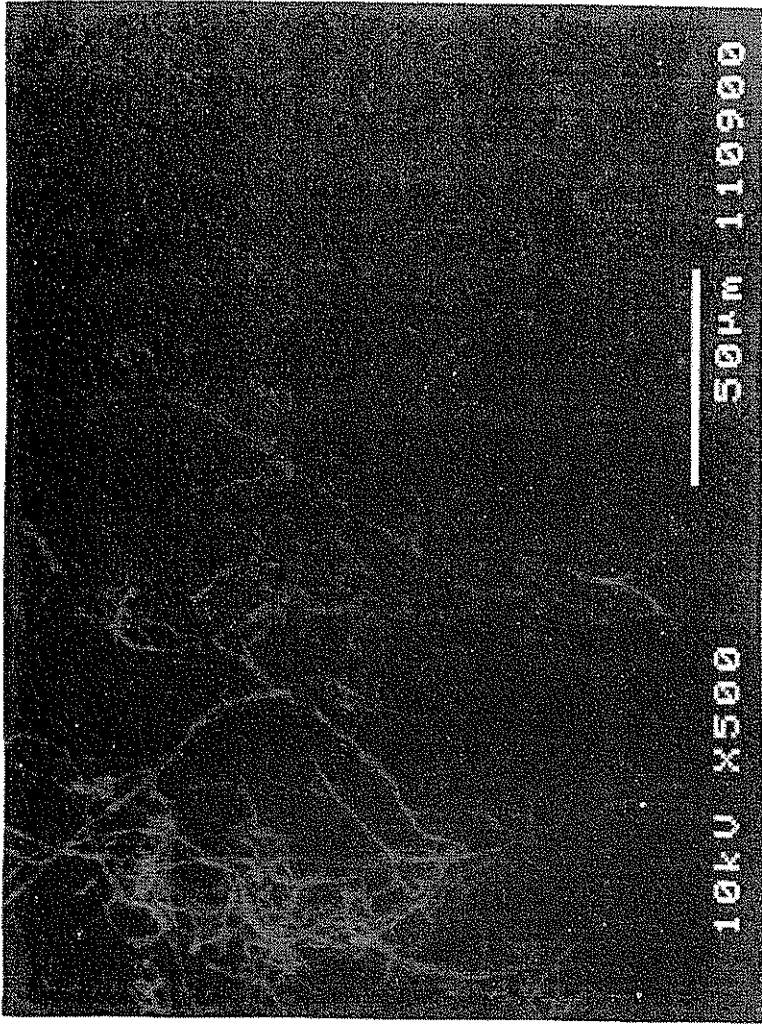


Figure 2. Chorion lamellar zone showing fibrillar matrix

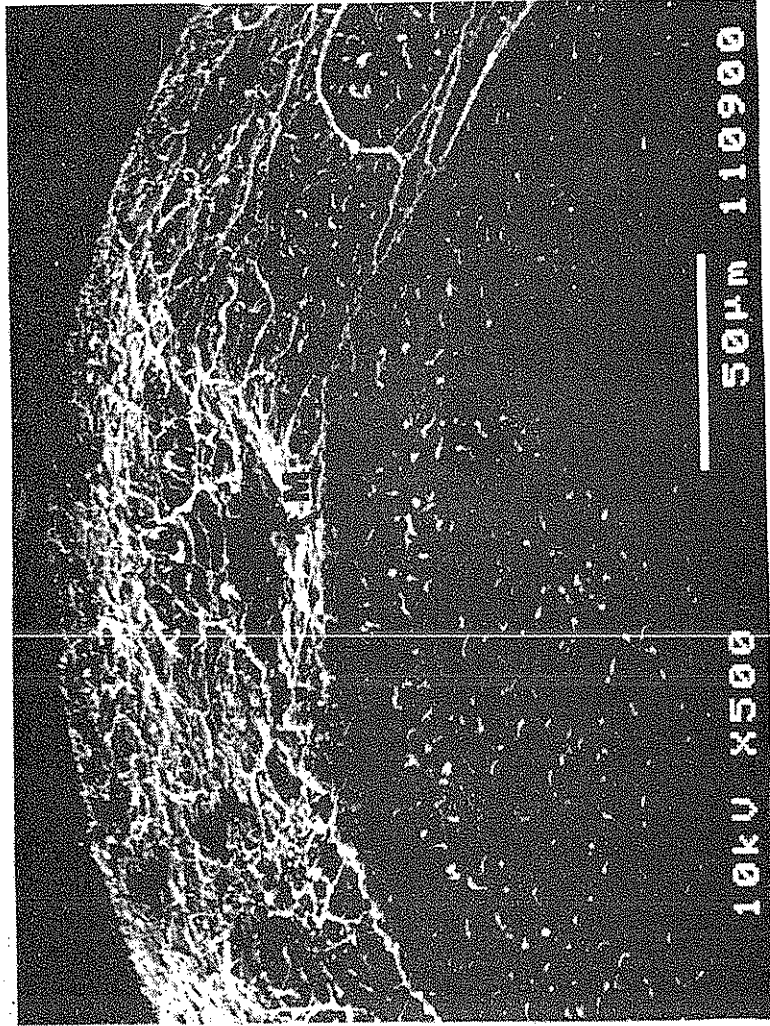


Figure 3. Surface view of ovum showing micropyle (M)

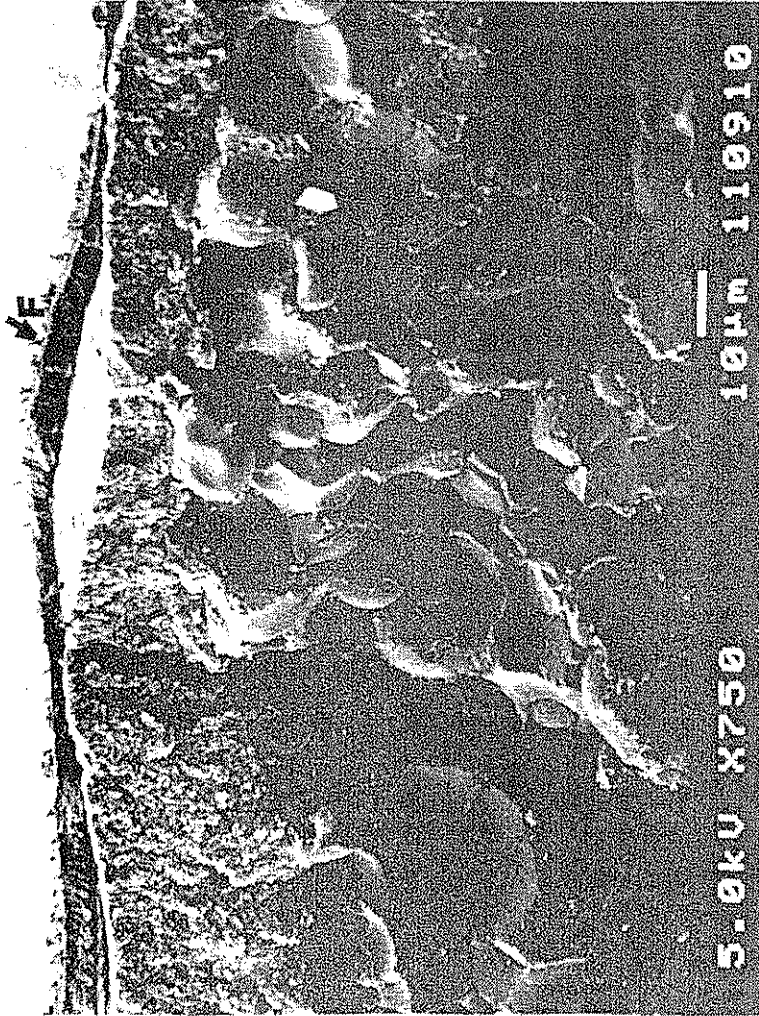


Figure 4. Transverse view of lamellar zone of chorion (F)

## Discussion

In fish, egg cell itself has a tender membrane when ready to leave the ovary. Discus fish eggs after fertilization become sticky and adhere to any suitable surface in water. Since sticking of the some fish eggs on any object is important to fishery scientists. Therefore it was decided to study the matter in more detail.

The present scanning electron microscope study was undertaken to elucidate general appearance of discus eggs, particularly the structures associated with fertilization phenomena.

The present study of general appearance of fertilized eggs showed a similar structures to that found by the other workers in *Fundulus heteroclitus* (Shanklin and Armstrong 1952; Kaighn, 1964). The particularly significant feature of the outer surface of the egg is covered by a jelly-like coat, comprised of fibrils. In addition the micropyle apparatus which was consisting of relatively smooth-sided, funnel-shaped vestibule at the bottom and the canal sides were quite irregular. This feature was also noted in *Fundulus heteroclitus* by Kaighn (Kaighn, 1964).

## Özet

Döllenmiş diskus balık yumurtalarının yüzey yapısı elektron mikroskop ile incelendiğinde; oval, telolecital ve açık sarı renkteki yumurta kabuğunun (chorion) üzerinde kısa fibriller bulunan jel benzeri bir katmanın yer aldığı görülmüştür.

Baca şekilli mikrofil deliğinin tabana uzanan iç yüzeyinin, düzgün olduğu ve yumurta dış yüzeyinin fibril yığını ile kaplı olduğu görülmüştür.

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