

Observations on the Retinal Structure of a Fresh Water Carp *Cyprinion macrostomus* (Heckel)

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ABSTRACT. The structure of the retina of the light adapted fish *Cyprinion macrostomus* has been studied by light microscopy, and this revealed that the basic plan of retinal structure resembles that of other fresh water fishes, except for certain specific differences and specialization which are also noted in this study. The pigmented epithelium is composed of a single layer of cuboidal cells with long processes filled with extensive melanin pigments. The contact of the epithelial cell processes with the outer segments of the visual cell are also noted and correlated with the retinomotor responses. The slender rod cells are distributed singly or in clusters among the cone cells. The latter are of five types: short and long singles, equal and unequal doubles and occasional triple cones. These cones are often arranged in a well defined square mosaic pattern in tangential sections. This pattern may reveal the degree of retinal development. The lipid-like droplets have been observed in the scleral part of the cone ellipsoid. The present finding has been discussed in the light of existing knowledge about the occurrence of different cone types and oil droplets in the retina of different fresh water fishes.

Light microscopical investigations of the retina of the eye in some cyprinid fishes have shown incomplete knowledge and contradictory results of various structural aspects. These include i) the cone types and the cone arrangement in the retina, ii) the presence or absence of oil droplets in the cones. The distribution of different kinds of cones in the cyprinid fishes is very little known and has been reviewed by Ali and Anciau (1976). Wunder (1925) did not find double cones in 12 cyprinids, but double cones were observed by Engstrom (1960) in thirteen cyprinids (including those studied by Wunder). Even triple cones have been found in *Leuciscus cephalus*, *Tinca tinca*, *Cyprinus carpio* (Vrabec 1955) and *Rutilus tricolor* (Al-Mehdi *et al.* 1983). It has also disputed as to whether equal double cones exist within this family or whether only unequal double cones or both types are to be

found there. In connection with that question, the possible existence of a regular cone pattern in the retina of the cyprinid fishes has been described by Muller (1952) but denied by Lyall (1957). The studies of Engstrom (1960) and Al-Mehdi *et al.* (1983) support the findings of Muller (1952).

The oil droplets, which consist of a dense globular mass usually found at the scleral end of the cone ellipsoid in some Cyprinid fishes (Anctil and Ali 1976) were long considered absent in the teleosts (Munk 1968) until Berger (1966) reported it in the guppy at the ultrastructural level. More recently their occurrence in the cyprinid retinas has still been disputed (Crescitelli 1972, Rodieck 1973, Anctil and Ali 1976, Al-Mehdi *et al.* 1983).

The aim of the present study, therefore, is to provide more information concerning the above contradictory records of the eye retina. A part of the present study describes also the structure of the eye retina in a fresh water fish *Cyprinion macrostomus* at the light microscopical level which was not reported previously.

Material and Methods

The carp, *Cyprinion macrostomus*, with body length ranged from 64 mm to 101 mm, was collected from the small rivers of Sulaimaniyah district in northern Iraq. Eyes were taken from light adapted fishes and fixed for 6 hr in 2% glutaraldehyde, 4% paraformaldehyde mixture in 0.1 M cacodylate buffer at pH 7 including 1% sucrose (Karnovsky 1965) and post fixed for 2 hr in 1% osmium tetroxide buffered with 0.1 M cacodylate. The eyes were dehydrated in graded ethyl alcohol and propylene oxide and embedded in Araldite Cy 212. They were cut transversely and tangentially at one micrometer thickness and stained with Azure II-methylene blue.

Results

The retina of the *Cyprinion macrostomus* (Heckel) conforms to the morphological type commonly found in the fresh water fishes. The pigment epithelial layer composed of a single layer of cuboidal cells. The cells contain a large round nucleus usually located sclerally near the basement membrane (Fig. 1). The vitreal (apical) regions of the cells extend numerous long cytoplasmic processes filled with extensive melanin pigments and enclose the outer segments of the visual cells. The melanin pigment was less abundant in the cell body.

The rod cells are less numerous and distributed singly or in clusters among the cone cells. They are slender cells with long myoid and cylindrical outer segment that reach the epithelial cells (Fig. 1).

The cone cells are usually seen as singles, doubles and rarely triples (Fig. 1 and 2). The single cells are of two types: short and long cells, whereas the double

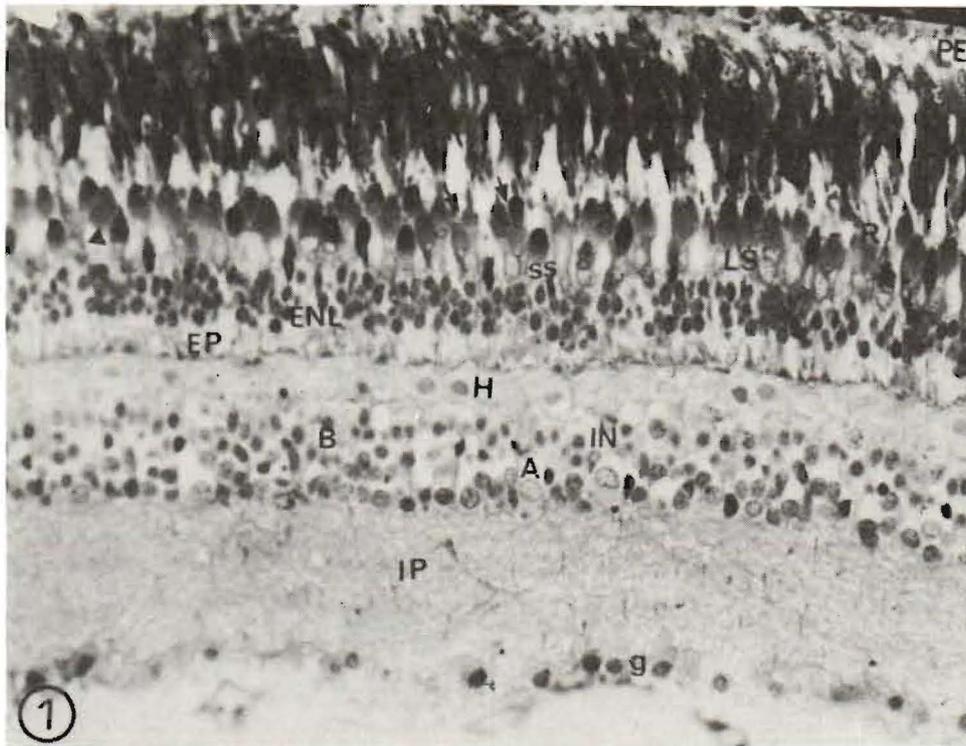


Fig. 1. Transverse section through the retina. The pigment epithelium (PE) with their processes filled with heavy pigment granules. R = Rod cells, LS = Long single cone, SS = short single cone, unequal double (small arrows) and triple cone (arrow head). ENL = External nuclear layer, EP = External Plexiform layer, IN = Internal nuclear layer, IP = Internal plexiform layer, H = Horizontal cell, B = Bipolar cell, A = Amacrine cell, g = ganglion cell. $\times 1260$.

cones are unequal and equal cells. The long cones are generally tall cells with bulky ellipsoid, and their nuclei are usually found outside or straddling the external limiting membrane (Fig. 2 and 3). The nuclei of the short single cones are situated across the external membrane and sclerally to the external nuclear layer. It has been observed that most of the short single cones are without a myoid region in their inner segments.

In tangential sections, the cone cells are arranged in a well developed square mosaic pattern with the short single cones at the corners and a long single cone at the center (Fig. 4).

The cone cells possess distinct globular structures or droplets at the scleral tip of the ellipsoid giving a coarsely granular texture to the inner segment of the cones. These lipid-like granules are found in all cone types (Fig. 2 and 3).

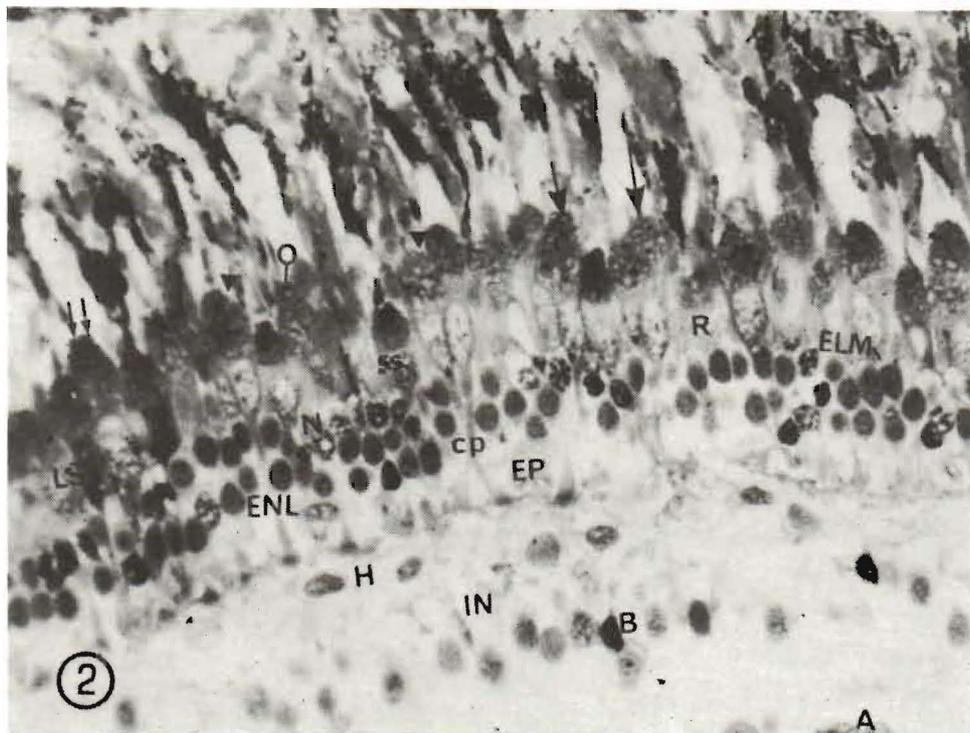


Fig. 2. Transverse section showing the various cone types: LS = Long single, SS = Short single. An equal double is seen at the parallel arrows, unequal double at the blank arrow head and the triple cones are indicated by large arrows. ELM = External limiting membrane, R = Rod cell, N = Nucleus of short single cone, cp = cone pedicle, ENL = External nuclear layer, IN = Internal nuclear layer, H = Horizontal cell, B = Bipolar cell, A = Amacrine cell. The oil droplets (O) are shown at the scleral part of the cone ellipsoids. $\times 2016$.

The external nuclear layer composed of 2-5 profiles of dense and small rod nuclei and few large and pale nuclei which belong to the cone cells and are situated sclerally near the external limiting membrane (Fig. 1, 2 & 3).

The internal nuclear layer composed of 6-7 profiles of neuronal cells which are separated from the external nuclear layer by a thin external plexiform layer. There is a layer of horizontal cells with large ovoid to round nuclei followed by several cell layers composed of bipolar and amacrine cells (Fig. 1 and 2). The bipolar cells are spindle or globular in shape and their dendrites synapse with the foot pieces of the photoreceptor cells while their axons are in contact with the dendrites of the ganglion cells. The amacrine cells are fewer and larger than the bipolar cells and present deeply in the internal nuclear layer. Each amacrine cell contains a large, round and lightly stained nucleus (Fig. 1).

The ganglionic layer is composed of a single profile of small or large ganglion cells (Fig. 1 and 2). Their axons constitute the nerve fiber layer which is thick near the optic disc, where the fibers emerge to form the optic nerve.

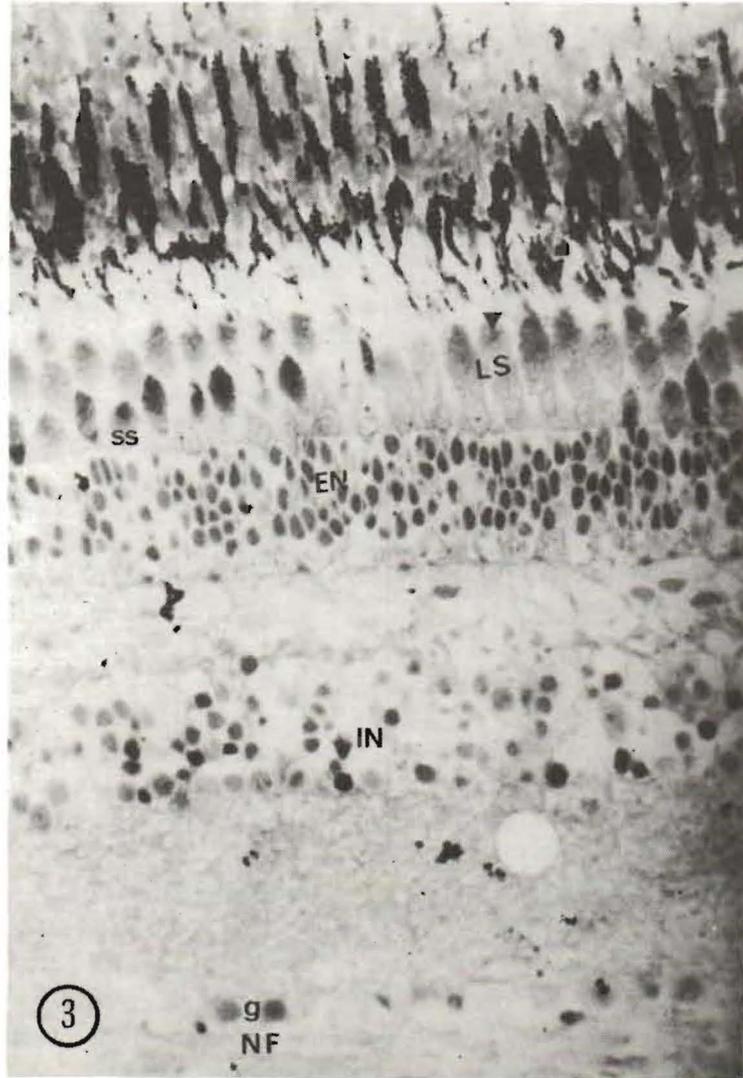


Fig. 3. Transverse section showing the long single cones (LS) and the short single cone (SS). Oil droplets are also shown at the blank arrow heads and the ganglion cell (g) in the Nerve fiber layer (NF), EN = External nuclear layer, IN = Internal nuclear layer. $\times 2016$.

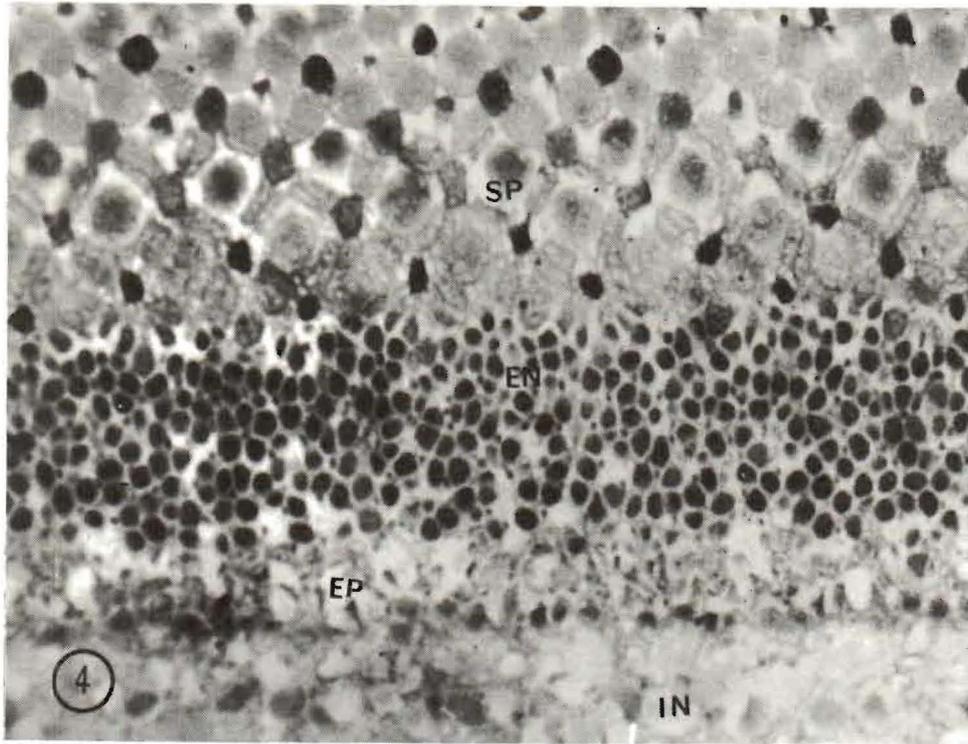


Fig. 4. Tangential section through the retina showing the cone arrangement in the form of square mosaic pattern (SP). EN = External nuclear layer, EP = External plexiform layer, IN = Internal nuclear layer. $\times 3200$.

Discussion

Although certain specific differences and specializations are noted, the morphology of the *Cyprinion macrostomus* (Heckel) is quite typical of that described for most other cyprinids (Engstrom 1960, Ali and Anctil 1976, Al-Mehdi *et al.* 1983). The specific differences and specialisations are:

- (1) The presence of single, double and triple cones.
- (2) The absence of the myoid in the short single cone.
- (3) The cone arrangement as square pattern is seen in the tangential sections.
- (4) The localisation of the lipid like droplets in the scleral tip of the ellipsoid of the cone cells.

The pigment epithelial layer consists of a profile of cuboidal cells with numerous long cytoplasmic processes arising from the apical portions of the cells which

intimately enclose the photoreceptor cells. It is suggested that these apical processes are active in phagocytosis (Steinberg and Wood 1974, Young 1978) as well as imparting architectural stability to the photoreceptor outer segments (Bernstein 1961). In addition, the contact of epithelial cell processes with the outer segments of the rods and cones indicates the intimate relationship between the epithelial cells and the visual cells. This relationship has been correlated with the retinomotor movement (Eckelbarger *et al.* 1980).

The major types of the cone cells which have been revealed in the present study are singles, doubles and triples. The single cones are short and long types but they vary in shape and length with the absence of the myoid in the short single cone. This fact is coupled with the observation of their absence in other cyprinid fishes (Engstrom 1960, Al-Mehdi *et al.* 1983). An unusual feature that has been observed in the *Cyprinion macrostomus* retina is that both equal and unequal double cones are present. This feature has also been observed in the carp *Rutilus tricolor* (Al-Mehdi *et al.* 1983). On the other hand, equal double cones have been reported in *Cyprinus carpio* (Engstrom 1960), whereas the unequal double cones are found in some other species of fresh water fishes: *Leuciscus rutilus*, *Carassius auratus*, *Abramis vimba* (Engstrom 1960). The triple cones have been observed in the *Cyprinion macrostomus* as well as in the carp *Rutilus tricolor* (Al-Mehdi *et al.* 1983), in the *Abramis blicca* and in the *Cyprinus carpio* (Engstrom 1960). However, the triple cone is apparently absent in the retina of *Leuciscus rutilus*, *Phoxinus phoxinus* and *Alburnus alburnus* (Engstrom 1960). The different opinions on the distribution of the different cone types within the family Cyprinidae as presented above are attributable to a large extent due to the absence of any standard method and to the species specific differences.

The arrangement of the cones has been studied from tangential sections and has revealed a regular square pattern with the long cones in the center and the short cones at the corners of the square pattern. This pattern differs from the square pattern which has been observed in the carp *Rutilus tricolor* (Al-Mehdi *et al.* 1983). In the latter, the short cones are present in the center, whereas the long cones are located at the corners. The tendencies to form a square pattern found in some cyprinids, do not necessarily mean that these species have a divergent type of cone pattern, but only that it is a little more developed (Engstrom 1960). It has been suggested that the presence of the well defined cone pattern may reflect a sign of a high degree of retinal development (Al-Mehdi *et al.* 1983).

In the present study, the localisation of globular masses or lipid-like droplets in the scleral tip of the ellipsoid of the cone is similar to the finding obtained in other species such as the carp *Rutilus tricolor* (Al-Mehdi *et al.* 1983). These globules, however, are apparently absent in the retina of *Panchax lineatus* fish (Engstrom 1963). The differences in the accounts of the globular masses localisation may be attributed to the absence of any standard method. This morphological feature supposedly appear to be formed by mitochondrial transformations in a

vitreal-scleral gradient (Anctil and Ali 1976). The presence of the globular masses in the scleral tip of the cone ellipsoid probably reflects the mitochondrial maturation that occurs in the ellipsoid of the cone cells.

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ملاحظات حول تركيب الشبكية لعين سمكة المياه العذبة (سايرينيون ماكروستومس)

ميثم إسماعيل عباس المهدي

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الجمهورية العراقية

تمت دراسة تركيب الشبكية لنوع من أسماك المياه العذبة العراقية سايرينيون ماكروستومس باستخدام المجهر الضوئي . وقد وجد تشابه بالتركيب الأساسي لشبكية هذا النوع من الأسماك وأسماك المياه العذبة الأخرى ماعدا بعض الفوارق والتخصصات التركيبية التي أظهرتها هذه الدراسة .

لوحظ أن طبقة الخلايا الطلائية الخارجية تتكون من خلايا مكعبة ذات بروزات سيتوبلازمية تمتلئ بحبيبات صبغ الميلانين وتكون تلك البروزات ذات تماس متماسك مع الأجزاء الخارجية للخلايا البصرية . وقد عزي هذا الترابط بعلاقته مع الاستجابات الحركية الشبكية التي تحدث داخل الشبكية . احتوت الطبقة البصرية على خلايا عصبية نحيفة تنتشر بصورة انفرادية أو متجمعة ما بين الخلايا المخروطية . وهذه الأخيرة، تكون على خمسة أنواع : خلايا قصيرة وطويلة، مزدوجة وغير مزدوجة وأحيانا خلايا ثلاثية ؛ ولقد اتضح في هذه الدراسة أن المخاريط في المقاطع المماسية تترتب بأنماط رباعية بلاطية الشكل وقد تعطي هذه الأنماط انطباعاً عن مدى نمو الشبكية في العين . ولوحظ كذلك وجود

حبيبات أو قطيرات دهنية في قمة الأجزاء الداخلية
للمخاريط البصريه .

وقد تمت مناقشة النتائج في ضوء الاختلاف عن وجود
الأنواع المختلفة للمخاريط وكذلك الاختلاف عن وجود
القطيرات الدهنية في أسماك المياه العذبة بشكل موجز .