SEA BREAM LIVER:HISTOLOGICAL AND ULTRASTRUCTURE STUDIES (II) BILIARY SYSTEM AND HEPATOPANCREAS.

M. A. El-Shammaa; S. M. El-Shafey; G. H. El- Bargeesy and M. I. Abd-Rabou

Department of Cytology & Histology,Faculty of Veterinary Medicine, Cairo University.

ABSTRACT

The liver was covered by a thin fibrous connective tissue capsule, and a single layer of mesothelial cells. The parenchyma of the liver was not divided into lobules. The biliary channels were dispersed throughout the parenchyma. The biliary tree was formed of canaliculi, preductules, ductules and ducts. The bile canaliculi were formed by the apposition of 2-3 hepatocytes and the plasmatic membrane at this level projected numerous microvilli into the lumen. Adjacent cells were joined by apparent tight junctions, followed by desmosomes. Bile ductules were lined by 2-3 cuboidal cells with rounded nuclei and prominent nucleoli, its outer contour was surrounded by dark stained cell with flat nucleus. These cells were attached by apparent tight junctions, desmosomes and interdigitations. Additionally they were joined to the hepatocytcs basally by desmosomes. Few short microvilli projected into the lumen.

Bile ducts showed a clear basal lamina. The small sized duct was lined by 4-6 cuboidal cells, with spherical vesicular nuclei, prominent nucleoli and with clear apical brush border. It was surrounded by connective tissue. Ultrastructurally, the bile duct was encircled by fibrocytes and myocytes, up to 4 layers. Few microvilli were observed on the apical border. The large bile ducts were lined by more than six cells and had a much wider lumen than the smaller ducts. Its lining was columnar cells with slightly elongated basal nuclei. Dark stained

nuclei of migratory cells were observed in the epithelium of large bile duct. Rodlet cells were found among the biliary epithelial cells.The large bile duct was surrounded by abundant fibrous connective tissue formed mainly of collagen fibers and few reticular fibers. The bile duct had apical brush border which reacted positively with PAS.

Exocrine pancreatic tissue was observed intrahepatically around the large branches of the portal vein and found also along its course and tributaries. The pancreatic cell appeared pyramidal to columnar in shape with spherical basal nuclei and prominent nucleoli. The cytoplasm was differentiated into two parts; basal around nucleus which was deeply basophilic and apical contained acidophilic granules. The acidophilic (zymogenic) granules directed toward the lumen of acini and the nucleus faced the portal vein and adjacent hepatocytes. The exocrine pancreatic cells were arranged in two rows. The intralobular pancreatic duct appeared between acini and contained secretion. They were lined by cuboidal cells with spherical nuclei and peripheral heterochromatin. Ultrastructrally the pyramid-shaped cells had basally located nuclei and prominent nucleoli; both were some of characteristic features of highly active cells. The basal cytoplasm was packed with dilated profiles of rough endoplasmic reticulum, among which elongated mitochondria were scattered. Smaller irregular microvilli were seen projected into the lumen.

INTRODUCTION

Fish have some unique anatomical and physical characteristics that are different from mammals; however, they still possess the same organ systems that are present in other animals. Organ systems of fish vary to some extent from that of mammals due to the aquatic environment they live in.Gilthead sea bream *(Sparus aurat*a) is a species of great economical importance for the Mediterranean marinculture industry. Rapid growth rates, good productivity per unit volume of water and

economic food conversion makes sea bream a suitable fish for the needs of modern aquaculture. This group of fish has long been extensively bred in lagoons in Egypt, France, Greece, Tunisia, Turkey, Spain and in Italy. But since the 1970s (when artificial reproduction techniques were established), growing of these fish has become more intensive in ponds, tanks, raceways and cages. Nowadays there are only a few land-based farms while the major part of the on growing takes place in sea cages ***(Stephanis, 1996).***

Marked attentions are focused on liver of fish. This can be attributed to the fact that, the liver plays a major role in the process of vitellogenesis of oocytes (production of yolk protein) and in the energy production during spawning ***(Wallace, 1978; Quebral, 1991; Toru & Shozo, 1998 and Arukwe & Goksoyer, 2003).***With the escalating threat of pollution, the search for histological indicators of environmental quality and physiological stress achieves increasing importance. The liver is an integrator in physiological and biochemical functions and thus alterations, for example in its structure, might be expected under certain toxic circumstances ***(Hinton & Laurèn, 1990 and Rocha, Monteiro and Pereira, 1994a).*** Besides it is used as biomarker for determinig environmental quality ***(Ashley, 1972; Malins & Haimanot, 1991 and Gonzàlez, Crespo and Bruslè, 1993).***

As a preliminary test for the evaluation of pathological reactions, it is necessary to establish the normal structure of the liver; so that, morphological changes associated with age, sex, season or nutritional stage, may be confused with toxic induced lesions. Additionally, because no soild basic studies are carried out, the nomenclature used for the liver of mammals is frequently applied to fish liver, generating misinterpret-

ations in several occasions ***(Hampton, McCuskey, McCuskey & Hinton, 1985).*** From the above, and because of the advantages of these fish as a food source, sea bream deserve some attention. This work concerned the normal histological and cytological organization of the liver in sea bream fish species in addition to the histochemical remarks involving its biochemical functions. In order to clarify the percise organization of liver in these important species.

MATERIAL AND METHODS

The material used in this study was Gilthead sea bream obtained from some private fisheries near the Mediterranean sea in EL-Manzala lagoon. Forty adult hermaphrodite fish ranged between 250-400 gms in weight were collected over the period from September 2003 to April 2004.The samples for light microscopic investigation were dissected finely from the fish. Pieces of about 1cubic cm. were taken quickly from liver, then transferred to 10% neutral buffered formalin, Bouins fluid, Sussa fixative, Zenker formol and Gender's fixatives. The tissue blocks were processed and embedded in paraffin wax. Tissue sections of 5-6 µm thicknesses were prepared and staind with Harris haematoxylin and eosin(H&E), Crossmon's trichrome stain, Gomori's reticulin and PAS techniques. The fixatives and staining methods were used as outlined by ***Crossmon (1937); Cook (1974); Drury &Wallington (1980) and Bancroft & Stevens (1982).*** For the ultrastructural purposes, small pieces of about $1mm^3$ of the liver were immediately fixed in a mixture of paraformaldehyde/glutaraldehyde buffered with 0.1 M phosphate buffer PH 7.2 conc. 2.5% ***(Karnovesky, 1965).*** Specimens were post fixed in 2.0% buffered osmium tetroxide and finally embedded in Spurr. Thin

sections were obtained and were double stained using uranyl acetate and lead citrate then examined by electron microscope.

RESULTS

The biliary tree was formed of canaliculi, preductules, ductules and ducts. The bile canaliculi were formed by the apposition of 2-3 hepatocytes and the plasmatic membrane at this level projected numerous microvilli into the lumen. The cytoplasm immediately adjacent to the canaliculi was denser than the general cytoplasmic background (Fig.1). Adjacent cells were joined by apparent desmosomes. Microvilli were projected from the hepatocytes into the canalicular lumen, except in the smaller one according to level of section. Bundles of filaments formed a circular network around the pericanalicular cytoplasm. Canaliculi that were formed by the same number of hepatocyts but had quite different diameters were also observed (Fig.1). In several sections there were darkly stained nuclei of bile preductular cell between hepatocytes. With ultrastructural examination, these cells form a single duct cell (preductule). These cells were distinguished from the hepatocytes itself by its elongated nucleus with euchromatin and heterochromatin arranged at the periphery and by a paucity of cell organells (Figs.1 & 2). They had no basal lamina and were linked to hepatocytes by the intercellular junctions. Nearly all luminal microvilli belonged to the hepatocytes. At the canaliculo-ductular junction biliary passage become gradually formed by two bile preductular epithelial cells, which appeared pyramidal in shape (Fig.3).

Bile ductules begin to be formed when the wall of the biliary channels was internally constituted by biliary epithelial cells (up to 4 in number). The bile ductule was lined by 2-3 cuboidal cells, with rounded clear nuclei and prominent nucleoli, its outer contour was surrounded by

dark stained cells, with flat nuclei, (Figs.4 & 5) and with connective tissue. Some canaliculi open directly into ductules. Bile ducts showed a clear basal lamina. The small sized duct was lined by 4-6 cuboidal cells, had spherical vesicular nuclei with prominent nucleoli and clear apical brush border. It was surrounded by haemopoitic tissue as well as connective tissue (Figs.4 & 6). The bile duct was encircled by fibrocytes and myocytes, up to 4 layers. Biliary epithelial cells were cuboidal in shape with spherical basal nuclei and heterochromatin arranged on the periphery. Microvilli were observed on the apical border (Fig.7). Apical junctional complexes were also present.

The large bile ducts were lined by more than six cells and had a much wider lumen than the smaller ducts. It was lined by columnar cells with slightly elongated basal nuclei. Dark stained nuclei of migratory cells were observed in the epithelium of large bile duct (Fig.8). In some areas; the small, medium and large sized bile ducts were observed in between the hepatic cells. The transition between biliary ductule and bile duct occurred with the appearance of a connective tissue sheath surrounding the duct, a hepatic arteriole, and usually one or more pigment laden melanomacrophages (Fig. 8).Some sections showed rodlet cell among the biliary epithelial cells. These cells appeared lightly stained with clear dark outline and a nucleus occupying one pole (Fig.9). The large bile duct was surrounded by abundant fibrous connective tissue formed mainly of collagen fibers and few reticular fibers (Figs.10&11). The bile duct had apical brush border which reacted positively with PAS.

The parenchyma of liver contained an association between bile duct that was lined by cuboidal to columnar cells with acidophilic cytoplasm and an arteriole which was called Biliary arteriolar structure (BA). This association was enclosed by connective tissue fibers. The sizes of this type of association were very variable. The proportions of

the components were also variable, and so, it was fairly common to find these associations with two or three biliary ducts and only one arteriole (Fig.8). Also bile duct may be associated with a venous vessel and exocrine pancreatic tissue (Fig. 10) or may be appeared alone in the parenchyma as biliary tract (BT) . Veins and venules are seen scattered in the parenchyma.Venous-biliary-arteriolar structure (VBA) was seen intermingled with connective tissue; veins (or venules), biliary ducts, and arterioles (Fig.10). Generally, they were formed by only one vein, one or two arterioles, and one to two biliary ducts. Groups of melanomacrophages tend to be dispersed in the connective tissue of these structures . Some of the large VBA associated with hepatopancreas (Figs 8 &10).

Exocrine pancreatic tissue was observed intrahepatically around the large branches of the portal vein and found also along its course and tributaries. The pancreatic tissue was surrounded by thin layer of connective tissue containing fibroblasts. This layer was formed of collagen and reticular fibers. The pancreatic cells appeared pyramidal to columnar in shape with spherical basal nuclei and prominent nucleoli. The cytoplasm was differentiated into two parts; basal around nucleus which was deeply basophilic and apical contained acidophilic granules. The exocrine pancreas was formed of tubulo-alveolar secretory end-pieces. The acidophilic (zymogenic) granules were directed toward the lumen of acini and the nucleus faced the portal vein and adjacent hepatocytes (Fig.12). The exocrine pancreatic cells were arranged in two rows, the basal region of the inner row of cells was in contact with the basement membrane of the vein, while the basal region of the outer row came in contact with the outer layer of connective tissue. Within this tissue**,** intralobular pancreatic duct appeared between acini and contained secretion. Few collagen fibers seemed to be extended between pancreatic

acini (Fig. 13). The pancreatic duct appeared lined by flattened to cuboidal cells with spherical dark stained nucleus and surrounded by few collagen fibers (Figs.5&12&13). Ultrastructrally the pancreatic acini were observed around its central lumen . The pyramid- shaped cells had basally located nuclei and prominent nucleoli; both were some of characteristic features of highly active cells. The basal cytoplasm was packed with dilated profiles of rough endoplasmic reticulum among which elongated mitochondria were scattered (Fig.14). Mature zymogenic granules were seen as electron dense granules, which aggregate in the apical cytoplasm. Smaller irregular microvilli were seen projected into the lumen (Figs. 14 & 15).

Exocrine pancreatic tissue appeared associated with bile ducts at different sites (Figs 4& 8). Exocrine pancreatic tissue was enclosed from out side by a single layer of flat cells with elongated nucleus and dispersed chromatin . In the center of acini we found elongated cell with elongated nucleus directed toward the pancreatic duct;it was the centro acinar cell (Fig.16). The pancreatic duct appeared embedded between the pancreatic tissues. They were lined by cuboidal cells with spherical nuclei and peripheral heterochromatin (Fig. 17).Melanomacrophage centers appeared as group of macrophage with colored material and melanin pigments. They were found associated with hepatopancreas (Fig.18**).**

DISCUSSION

With the exception of a study on rainbow trout ***(Hampton, Lantz, GoldBlatt, Lauren and Hinton 1988),*** a detailed description of the intrahepatic biliary passage in teleosts was lacking. Here we reported the biliary tree of sea bream which begins with the bile canaliculi, preductules, ductules and ducts. This findings came in accordance with

several authors ***(Bonates & Ferri; 1980, Chapman; 1981, Rocha et al.;1994a, El-Habback; 1995 and Konsowa & Abd El-gawad, 2001)*** in several species of fish**.**

Bile canaliculi of sea bream was formed by the close apposition of two to three cells, and involve cell to cell contact by tight junctions followed by desmosomes. These junctions obliterated the intercellular spaces between the liver cells separating the bile canalicular lumen from the blood plasma space. This description seems in harmony with that of ***Hinton & Pool (1976), Bonates & Ferri (1980) and Chapman (1981).*** The former author stated that the number of cells involved in bile canaliculi may include at least four cells and complete junctional complexes were rarely encountered. On the contrary ***Ferri (1982),*** in fresh water teleost, mentioned that zonula occludens located along the bile canaliculus obliterated the intercellular spaces between hepatocytes. This explains the existence of a permeability barrier interposed between the lumen of the canaliculi and the interhepatocellular space which had a continuous access to the blood stream. The bile canaliculi in the adult goldfish liver possessed a smooth wall ***(David, 1961)***, and a similar picture was found in the liver of young sea trout embryos ***(Byczkowska-Smyk, 1967)***. While, in this study, the microvilli were projected from the hepatocytes into the canalicular lumen, except in the smaller one. Bundles of filaments formed a circular network around the pericanalicular cytoplasm. This result seemed to be in agreement with that of ***Byczkowska-Smyk (1967)***. It Had been suggested that closely packed microvilli in the bile canaliculi may be contractile and may be responsible for the control of the bile flow in the bile canaliculi in goldfish ***(Yamamoto, 1965)***, and crucian carp ***(Tanuma, 1980)***. We suggested that the microtubules inside microvilli were responsible for their contractile function in the bile canaliculi of sea bream liver.

Bile preductular cells with dark nuclei and light cytoplasm located in the center of the hepatic cords were mentioned in this study as the beginning of the duct system. These cells were elongated and found mainly associated with bile canaliculi, and in fact, showed some desmosomal junctions with them. The cells had long elongated nuclei with euchromatin and heterochromatin arranged at the periphery. The cytoplasm contained a paucity of cell organelles. These cells appeared to be similar to those noted in goldfish ***(Yamamoto, 1965),*** in channel catfish ***(Hinton & Pool, 1976)***, in rainbow trout ***(Hacking, Budd, and Hodson, 1977& Hampton et al., 1985),*** in Atlantic salmon ***(Robertson & Bradley, 1992)*** and in Tilapia ***(El-Habback, 1995).*** Further, the morphological evidence associated with the liver of crucian carp that the preductular cells contained a large number of dense bodies was indicative that they may be histiocytes or macrophages. Thus it was presumed that the population of preductular cells includes two types of mesenchymal cells, fibroblasts and histiocytoid cells ***(Tanuma, 1980)***. This study confirmed other studies by ***Yamamoto (1965), Hinton & Pool (1976)*** *and* ***Robertson & Bradley (1992)*** in regard to some aspects of the morphology of the bile preductular cells. It was observed that the number of organelles per cell was few when compared to the hepatocytes, and moreover these cells were contributing to the ducts of Hering as in mammals, as the initial part of the duct system connecting the canaliculi to the intraparenchymal duct. These cells may be originated from the intraparenchymal fibroblasts recorded in this study.

The structural feature at the zone of transition from the bile canaliculi to the bile ductule was of particular interest, since many differences existed between species. Thus, ***Yamamoto (1965)*** described that in goldfish each liver cell was provided by a single intracellular bile canaliculus, and each canaliculus joins independently a terminal bile duct

which was formed by a single duct cell. In fresh water telesots ***(Ferri, 1982),*** it had been clearly demonstrated that the canalicular-ductular junction,where the intercellular spaces were formed only by grooves in adjacent hepatocytes, was formed by a duct cell. Gradually 2 or more duct cells were added with a concomitant decreasing participation of hepatic cells. The auther had pointed out that the duct cells were very poor in organelles and contained abundant cytoplasmic filaments, it might be supposed that they had a very low metabolic activity and solely play some role in the bile transport. The similarity of structure may be the case in sea bream as well.

The liver of sea bream showed a well developed intraparenchymal bile duct system.The bile ductule was surrounded by 2-3 cuboidal epithelial cells in a single layer. The small ducts were lined by 4-6 cuboidal cells, while the large ducts had more than six cells. The lateral wall of this cells showed tight junctions, desmosomes and interdigitations, while the lumen of these intercellular biliary passages was patent and contained microvilli. The cytoplasm of these cells had sparse organelles and microfilaments. Cells increased in number with increasing of the diameter of the duct. The small sized duct was surrounded by a moderately developed basal lamina formed of collagen and reticular fibers, and containing one or two fibroblasts while the large duct had a well developed basal lamina. The basal lamina was absent in ductule. This results seemed in agreement with ***Yamamoto (1965), Nopanitaya, Carson, Grisham, and Aghajanian, (1979), Tanuma (1980), Hampton et al. (1988) and El-Habback (1995).*** The possibly contractile microfilaments may be contributed to the transport of bile through the intraparenchymal biliary passage by bringing about peristaltic movement, as well as by the maintenance of the tonus of the biliary passage, as ***Tanuma (1980)*** had suggested that case in crucian

carp. On the other hand, intracytoplasmic microfilaments (cytoskeleton) in the intraparenchymal bile ducts may serve as the internal framework for the hepatic tissues as suggested by ***Tanuma, Ohato and Ito, (1982)***. The intracytoplasmic microfilaments might perform both functions in sea bream and could help in the passage of bile as well as support the hepatic parenchyma.The bile ducts of most vertebrates were composed of a transporting epithelium which was involved in secretion and absorption of water and bicarbonate ions ***(Goldfarb, Singer & Popper, 1963).*** The apical surfaces of the bile duct of sea bream had numerous microvilli, which might be reflective for their absorptive nature.

The present work revealed some large oval cells with well defined borders present between the cells of biliary epithelium of large duct. These cells contained dark stained nuclei that occupy one pole of the cell. These cells may be resembled the rodlet cell that described by ***Smith, Caceci, Marei and El-Habback (1995)***, in angel fish and ***Koponen & Mayers (2000),*** in freshwater bream. These cells were moderately reacted to PAS. This result appeared in agreement with that described by ***Smith et al. (1995),*** who emphasized that the rodlet cell produces secretory material of proteinceous nature. Rodlet cells were found in the intrahepatic biliary channels of the following six species: the minnow, the guppy, the brook stickleback, ***(Morrison & Odense, 1978),*** the Atlantic croaker, ***(Eurell & Haensly, 1982),*** Cabrilla sea bass, ***(Gonzàlez et al., 1993)*** and the brown trout, ***(Rocha, Monteiro and Pereira 1994b)***. We did not conclude much about the task of these strange cells and in our opinion a comprehensive study was needed to clarify their exact nature and functions.

It was opportune to state that the term BAT (biliary-arteriolar tract) was first introduced for fish liver histology by ***Hampton et al. (1988)***,

and the terms VBAT (Venous-biliary-arteriolar tract) and VAT (venous-arteriolar tract) were subsequently proposed by ***Rocha et al. (1994a).*** In the rainbow trout, Oncorhynchus mykiss; larger-diameter veins were occasionally associated with small bile channels and arterioles ***(Hampton, Lantz and Hinton, 1989 and Schar, Maly and Sasse 1985).*** The same fish also contained ***BAT (Hampton et al. 1985& 1988).*** Such triple associations also exist in the Atlantic salmon ***(Robertson & BradIey, 1992).*** Although those entities were occasionally observed in the Atlantic croaker, their appearance was not consistent enough to establish definite triads ***(Eurell & Haensly, 1982)***. In the liver of English sole, the biliary ducts and vascular elements were scattered irregularly throughout the parenchyma rather than organized into triads ***(Myers, Rhodes and McCain, 1987).*** Their absence was also reported in the cabrilla sea bass, ***(Gonzalez et al., 1993).*** In this fish, however, two-by-two associations among blood vessels and biliary ducts sometimes occur. ***Rocha et al. (1994a)*** reported that in brown trout, there were associations between venous, arterial, and biliary elements; the **VBAT**. Moreover, it was suggested that the **VBAT** should not be regarded as scarce. In the present paper, our data unequivocally confirm that these tracts **VBAT** occupied a considerable proportion of the areas in the stroma.

In sea bream the exocrine pancreatic tissue was observed intrahepatically around the large branches of the portal vein and found also along its course and tributaries. Intrahepatic pancreas in fish was studied by several authors ***(Weis; 1972, Kendall & Hawkins; 1975, Hinton & Pool; 1976, Timoshava; 1981, Groman; 1982, Hampton et al.; 1988, El-Habback; 1995, Konsowa & Abd El-gawad; 2001 and Vicentini, Franceschini-Vicentini, Bombonato, Bertolucci,Lima and Santos; 2005).*** The pancreatic cells were differentiated from the

hepatocytes by the presence of large rounded secretory zymogen granules. The latter occupied large portion of the pancreatic cells. However, the liver of Antractic notoheniids usually lacked pancreatic exocrine tissue ***(Eastman & DeVries, 1981).*** The structure of exocrine pancreatic acini of sea bream resembled that described in other teleosts ***(Eurell & Haensly; 1982, El-Habback; 1995 and Abd-Elfatah, 1999).*** The acini contained pyramidal to columnar epithelial cells with spherical basal nuclei and prominent nucleoli. The cytoplasm was deeply basophilic basally around the nucleus while apically it contained acidophilic granules.These zymogenic granules contained various digestive enzymes which were released directly to the digestive tract. Some of these enzymes catalyze the breakdown of protein to amino acids, while others catalyze the breakdown of fats and charbohydrates ***(Harder, 1975).*** It might be that they perform the same function in sea bream. Other types of cells were detected in the hepatopancreas of sea bream. These cells were arranged as single layer of flat cells, with a thin subepithelial layer, which resembled the covering epithelium. The same cells were observed in sea bass ***(Diaz & Connes, 1988 and El-Habback, 1995). Beccaria, Diaz & Connes (1992)*** decided that these cells may facilitate the transfer of molecule from the blood by channeling their movement in the interstitial substance of the connective tissues. In addition to this role, these cells might transfer some substance in bulk across the cell without going through the cell.

One type of pancreatic duct was recorded in anguillaris joining the bile duct near its opening in the duodenum ***(Macky, 1929)***, while ***Khalilov (1968)*** noticed a small pancreatic duct that discharge directly into the pyloric caeca of teleosts. On the other hand, ***Harder (1975)*** mentioned two types of ducts, a thin intercalary duct which

communicates with the secretory acini and was lined by flat cuboidal epithelium, and a larger excretory duct, lined by 2-3 cuboidal cells forming narrow irregular lumens devoid of basal lamina. These ducts joined to form larger interlobular ducts lined by more than three layers of high cuboidal epithelial cells and surrounded by a well developed basal lamina. The latter joined to form the main excretory duct, which had the same structure as the extrahepatic bile duct. In this study we reported only the intralobular pancreatic duct with similar structure. The duct system in the pancreas had been shown to transfer the homogenous substance produced by the acini to the intestine in many fish species ***(Harder, 1975).*** Hepatopancreas of sea bream appeared associated with bile ducts and melanomacrophage at different sites. Their appearance was not consistent enough to establish definite triads. Such speculation came in agreement with ***Eurell & Haensly (1982)*** description in Atlantic croaker.

LIST OF THE FIGURES

Fig. (1): Electron micrograph of sea bream liver showing different levels of bile canaliculi (b) between hepatocytes (h) which join together by desmosomes (arrow). Notice bile preductular cell (P) between hepatocytes. (Uranyl acetate-Lead citrate X5000)

Fig. (2): Electron micrograph of sea bream liver showing bile preductular cell (p) with elongated nucleus abut on bile canaliculi (b) obscured by Microvilli. (Uranyl acetate-Lead citrate X15000).

Fig. (3): Electron micrograph of sea bream liver showing two bile preductular cells (p) lead to bile ductule (d) between hepatocytes (h). Large quantity of glycogen (g) peripherally situated.(Uranyl acetate-Lead citrate X2000).

Fig. (4): A section in the liver of sea bream showing bile ductule (arrow) and small bile duct (b) associated with pancreatic acini (a) that was surrounded by haemopoietic tissue.(H&E X 410).

Fig. (5): A section in sea bream liver showing pancreatic duct within the pancreatic tissue. Notice bile ductule was surrounded by dark stained cell (arrow). (H&E X320).

Fig. (6): Higher magnification of Fig. (4) Showing small bile duct (b) was lined by 4 – 6 biliary epith. Cells and was surrounded by C.T. Notice pancreatic duct (p); pancreatic acini and haemopoietic tissue. (H&E X1025).

Fig. (7): Electron micrograph of bile duct showing biliary epithelial cell had spherical basal nucleus (n) with peripheral heterochromatin. Notice the lumen contained few microvilli (arrow) and the duct was surrounded by fibroblasts (f). (Uranyl acetate-Lead citrate X3000).

Fig. (8): A section in the liver of sea bream showing bigger biliary – arteriolar structure holding 2 bile ducts and one arteriole. Notice melano-macrophage (arrow), migratory cells and exocrine pancreatic tissue. (H&E X320).

Fig. (9): High magnification of large bile duct showing columnar epith. (e), microvilli (arrow)on the apical border and surrounding C.T. & smooth m. Notice rodlet cell (r) intraepithelial position. (H&E X800).

Fig. (10): A section in the liver of sea bream showing large bile duct (b), arteriole (a) and venule (v) forming venous-biliary-arteriolar

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Fig. (11): A section in the liver of sea bream showing reticular fibers in the C.T. sheath around bile duct. (Gomori's tech. X320).

Fig. (12): A section in the liver of sea bream at level of hepatopancreas showing spherical basal nucleus and apical acidophilic granules. Notice portal vein filled with RBCs and intralobular pancreatic duct (arrow). (H&E X410).

Fig. (13): A section in sea bream liver showing pancreatic duct containing secretion (p).Arteriole (a). (Crossmon's stain X200).

Fig. (14): Electron micrograph of pancreatic acinus showing zymogenic granules (z) of variable size, basal nucleus (n) with prominent nucleolus (ne) and dispersed heterochromatin, mitochondria (m) and great amount of rough endoplasmic reticulum. (Uranyl acetate-Lead citrate X6000).

Fig. (15): High magnification of pancreatic acini showing zymogenic granules (z)directed toward the lumen before its releasing by exocytosis. Notice lumen contained microvilli (mv). (Uranyl acetate-Lead citrate X6000).

Fig. (16): Electron micrograph showing a portion of exocrine pancreatic acini. Notice exocrine pancreatic cell (p) containing zymogenic granules (z) and elongated mitochondria (m) and was surrounded by elongated cell (f) of epithelial cover of pancreatic tissue. Centroacinar cell (c) and pancreatic duct (d). (Uranyl acetate-Lead citrate X2000).

Fig. (17): High magnification of pancreatic duct showing large nucleus (n) with dispersed heterochromatin on the periphery and narrow lumen (I). (Uranyl acetate-Lead citrate X6000).

Fig. (18): A section in sea bream liver showing melano-macrophage associated with pancreatic tissue. (PAS X410).

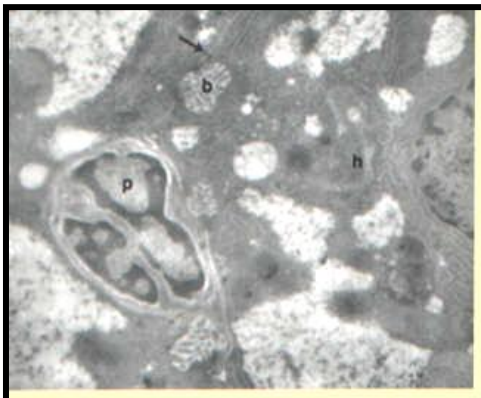
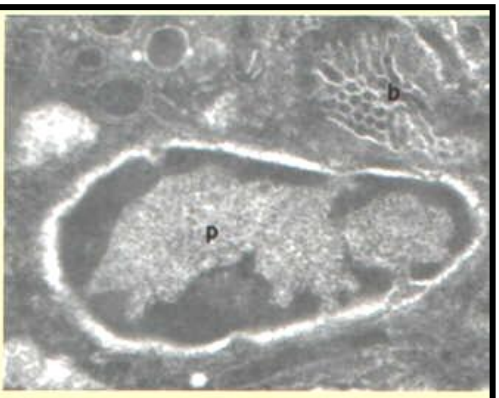


Fig 1



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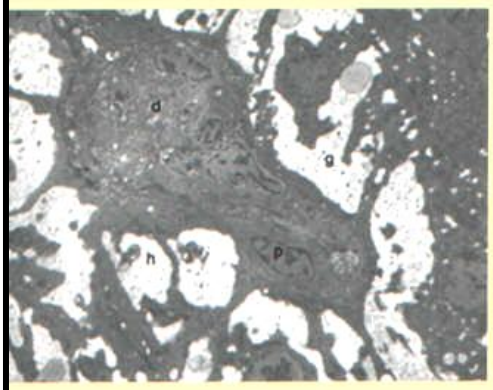

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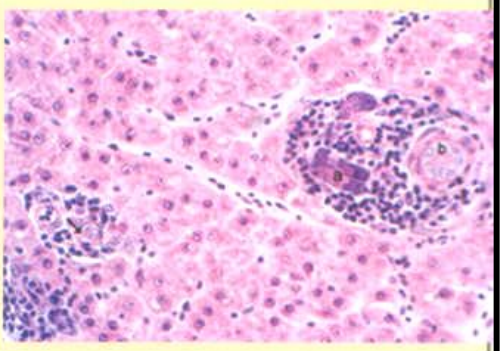

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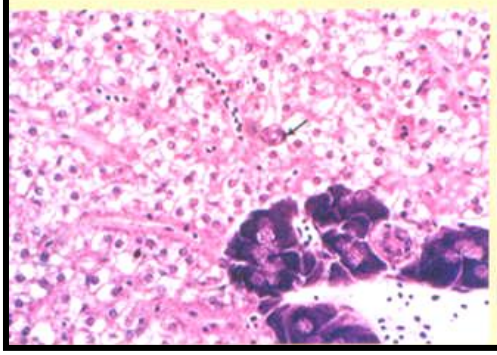
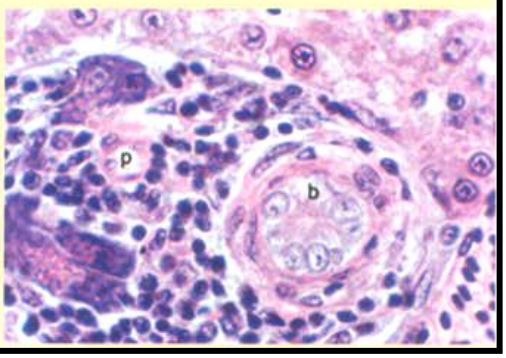




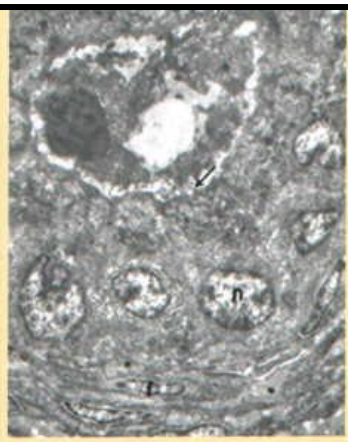


Fig. 7

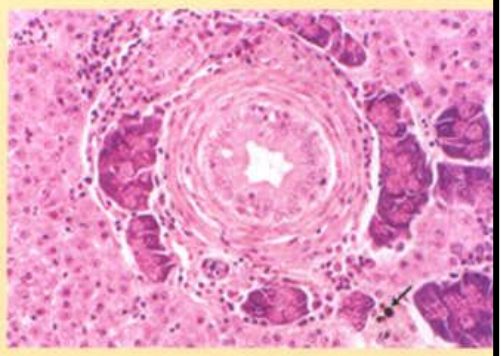


Fig. 8

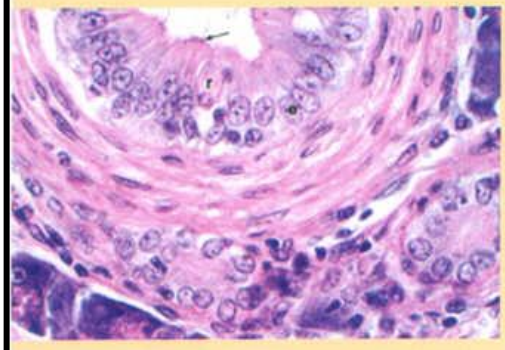


Fig. 9

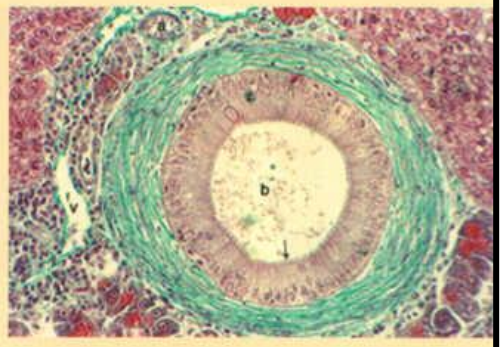
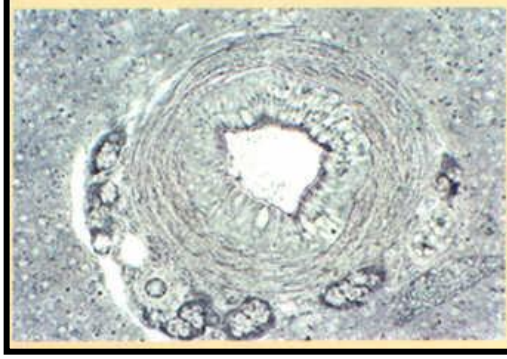
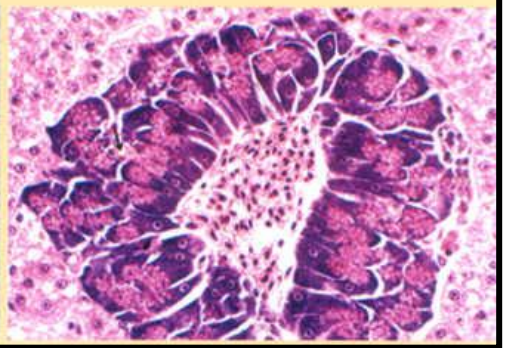


Fig. 10





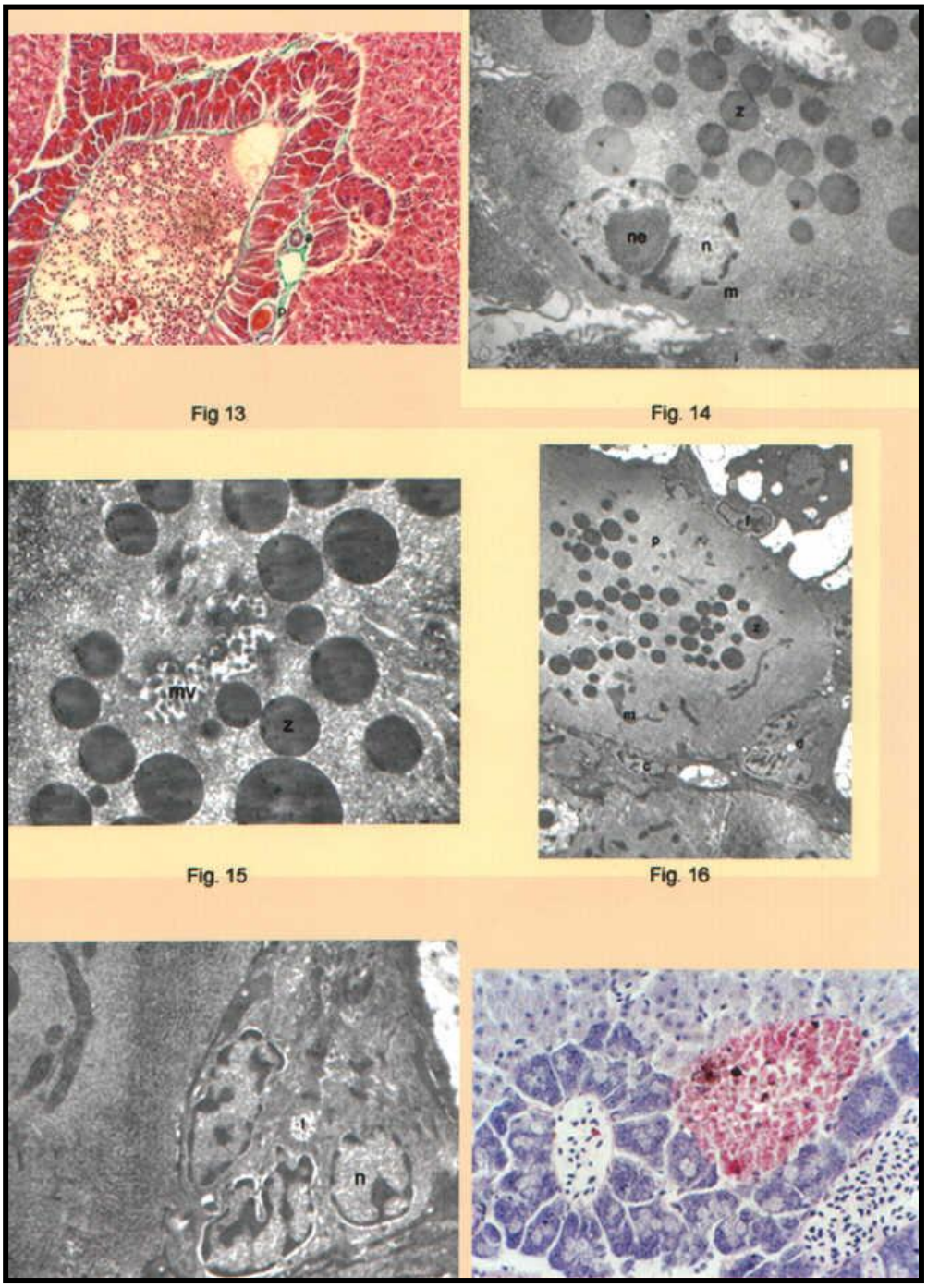


Fig 13
Fig. 14
Fig. 15
Fig. 16

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كبد سمك الدنيس :دراسات بالمجهر الضوئى والألكترونى (II) القنوات المرارية و نسيج البنكرياس

د./ ممدوح الشماع – د./ سمير الشافعى – د./ جهاد البرجيسى – د./ محمد عبد ربة

قسم الخلية والأنسجة –كلية الطب البيطرى –جامعة القاهرة

فى هذا البحث قمنا بدراسة التركيب الطبيعى لكبد سمك الدنيس ,باستخدام المجهر الضوئى و المجهر الألكترونى. وجد ان الكبد مغطى بمحفظة رقيقة من النسيج الضام الليفى و التى يغطيها صف واحد من الخلايا الحرشفية. لوحظ ان نسيج الكبد لا يتواجد فى صورة فصوص و أن المسارات المرارية

و العناصر الوعائية تنتشر بشكل عشوائى فى نسيج الكبد. تتألف الشجرة المرارية من القنيات المرارية ثم الأنابيب ثم القنوات. تتكون الأولى من مشاركه خليتين أوثلاثه من الخلايا الكبدية و يتزود الغشاء الخلوى بالخميلات. الخلايا المتجاورة ترتبط بوسائل ربط مختلفة. تبطن الانابيب بواسطة اثنين أو ثلاثة من الخلايا المكعبة. القنوات المرارية تبطن بخلايا مكعبة من (4–6) بها أنوية كروية باهتة و حافة فرشاتية. تحاط القنوات بنسيج ضام وخلايا مولدة الالياف وخلايا عضلية. الحافة الحرة مزوده بخميلات دقيقة بينما تكون القنوات المرارية الكبيرة مبطنه بأكثر من ست خلايا ولها فراغ داخلى واسع. الخلايا عمودية ذات أنوية بيضاوية قاعدية. تحاط هذة القنوات بكمية كبيرة من النسيج الضام به ألياف غروية و ألياف شبكية. الحافة الحرة تتفاعل ايجابياً مع كاشف شيف البيرأيودى.

النسيج البنكرياسى يتواجد فى داخل الكبد حول الأفرع الكبيرة للوريد البابى و على طول مساراتة. الخلية البنكرياسية هرمية الشكل تحتوى على أنوية قاعدية ذات نويات واضحة. البلازما الخلوية تنقسم إلى قسمين واضحين؛ قاعدى حول النواة و جزء قمى يحتوى على حبيبات حامضية. هذة الحبيبات تتجة إلى ناحية الفراغ الداخلى بينما تتوضع الأنوية فى اتجاه الوريد البابى و الخلايا الكبدية المجاورة. ينتظم النسيج البنكرياسى فى صفين. تظهر القناة البنكرياسية بين العنبات الافرازية و قد تظهر محتوية على افراز. تبطن هذة القنوات بخلايا مكعبة تحتوى على أنوية كروية. البلازما القاعدية تحتوى على العديد من الشبكة البلازمية الخشنة هذا بالاضافه الى الميتوكوندريا. تبرز فى التجويف خميلات دقيقة.