ORIGINAL ARTICLE

A comparative histochemical/histomorphologic assessment of the hepatic architecture in mammals

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ABSTRACT

Introduction: The mammalian liver is a functional and morphological complex organ with common histological features across different species. The mild variations in the mammalian liver were attributed to speciesadaptational changes, dietary habit, specific metabolic activities, and phylogenic organizational peculiarities.The previous studies on mammalian liver were not based on the macromolecules of the liver and could not provide detailed histological associations between herbivores, carnivores and omnivores of mammalian class. This study aimed to provide detailed histological association between the livers of six mammalian species.

Methodology: Ethical approval for this study was obtained from the Research and Bioethics Committee, Faculty of Basic Medical Sciences, Delta State University, Abraka (RBC/FBMS/DELSU/18/26). Six animals of mammalian species were utilized for this study in line with the guidelines for the care and use of animals for research.

Observations: A common trend was observed in the histological features of the liver across the six mammalian species studied: common histo-architectural features, distribution of glycoprotein, glycolipid and collagen type III were observed in the liver across the six mammalian species. Intermittent distribution of glycogen was observed in the liver of cow and high presence of glycogen was also observed in dog liver on oppose to their dietary classification.

Conclusion: This research work has established a strong histological association in the liver of three groups of mammals, based on their diet. The work has been able to provide histochemical evidence of the existence of a near phylogenetic relationship between the six species of mammals, studied.

Keywords: Mammal; Liver; Histoachitectural; Adaptation; Histology, Phylogeny

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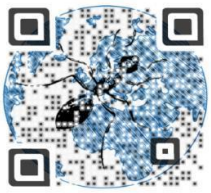

INTRODUCTION

Comparative anatomical study, establishes the anatomical association that exist in various species of organism.[1]It showed that different organisms share a common ancestor. Comparative anatomy assist scientists in classifying organisms based on their anatomical relationships[2] and serves as evidence for evolution.[3]

Animals are classified into three categories based on their physical structures and dietary habit; herbivores, omnivores and carnivores. All these animals are facilitating each other for their survival.[4]

Index inquisition into the histological features of the vertebrate liver indicated that the histoarchitecture of the liver was similar in various vertebrates, however there were marked differences, which were attributed to species adaptational changes, dietary habit, specific metabolic activities, and phylogenic organizational peculiarities.[5-7] The previous studies on mammalian liver were not based on the macromolecules of the liver and could not provide detailed histological associations between herbivores, carnivores and omnivores of mammalian class. This study was aimed at providing detailed histological association between the livers of six mammalian species by specifically staining and studying the histoarchitecture and histochemical staining pattern in the liver of the six mammalian species and comparing their histoarchitecture and histochemical staining pattern.

MATERIALS AND METHOD

Ethical approval for this study was obtained from the Research and Bioethics Committee, Faculty of Basic Medical Sciences, Delta State University, Abraka (RBC/FBMS/DELSU/18/26).

Six animals of mammalian species were utilized for this study in line with the guidelines for the care and use of animals for research.

Six mammalian species were utilized in this study and were grouped into three, based on their dietary habbits. Each of the group has two mammalian species present: Herbivorous group (*Bostaurus* and *Capra aegagrus*); Carnivorous group (*Canis l upus* and*Feliscatus*); Omnivorous group (*Rattusnorvegicus*and*Susscrofa*).

The animals were euthanized and the tissue harvested, fixed in 10% formal saline[8] and processed using standard techniques. (9) While Hematoxylin and Eosin stain was used to study the histoarchitecture, Periodic Acid-Schiff reagent highlighted the glycogen distribution. Periodic Acid-Schiff reagent with diastase demonstrated the glycoprotein and glycolipid distribution and Gomori's method for demonstration of reticulin was used to study the reticulin distribution. A light microscope (Olympus binocular microscope) at x40 x 100 and x400 magnifications facilitated the interpretation of the processed tissues.

RESULTS

Table 1.0: Histology and histochemical features of the six mammalian species.

	Herbivores		Carnivores		Omnivores	
Features	Cow	Goat	Dog	Cat	Rat	Pig
1. Central vein	+	+	+	+	+	+
2. Portal triad	+	+	+	+	+	+
3. Hepatocytes arrangement	σ, μ, τ	ϒ, μ	τ, σ	Σ	σ, μ	μ
4. Fibroconnective tissue stroma	+	+	+	+	+	+
5. Glycogen distribution	-	+	+	-	+	+
6. Glycoprotein	+	+	+	+	+	+
7. Glycolipid	+	+	+	+	+	+
8. Collagen type III (reticulin)	+	+	+	+	+	+

+= present; - = absent; σ = sheet; μ = tubule; τ = cluster; ϒ = cord

The table 1.0 shows the histological and histochemical features of the six mammalian species. Portal triad and the content (portal vein, bile ductule and hepatic artery) were present, enmeshed in fibromyxoid connective tissue stroma in all the mammalian species studied.

Variable occurrence of central veins of different sizes were also observed. Hepatocytes arrangement varied in various species as cow; sheet, tubules, and clusters, goat; cords and tubules, dog; clusters and sheets, cat; sheets, pig; tubules and rat; sheets and tubules. Fibroconnective tissue stroma enclosed all the liver tissue section studied.

Glycogen was well distributed in the parenchyma of all the mammalian species studied except the cow and cat liver tissue section with a sparsely distributed glycogen. The cow and cat liver tissue cells showed vacuolated cytoplasm which portends the absence of glycogen in the species liver parenchyma.

Individual hepatocytes of all the studied mammalian species showed dark pigmented intracytoplasmic vesicles, although which were small in size but demonstrates round to oval plomorphism. These vesicles also demonstrate some degree of birefugence.

The cytoplasm as well as the nuclear membrane of individual hepatocytes responds to the reticulin stain. The connective tissue in the sinusoidal wall and the fibromixoid connective tissue stroma in which the portal structures are enmeshed were also intensely stained with reticulin stain which indicates presence of collagen type III (reticulin).

Hematoxylin and Eosin tissue stain sections

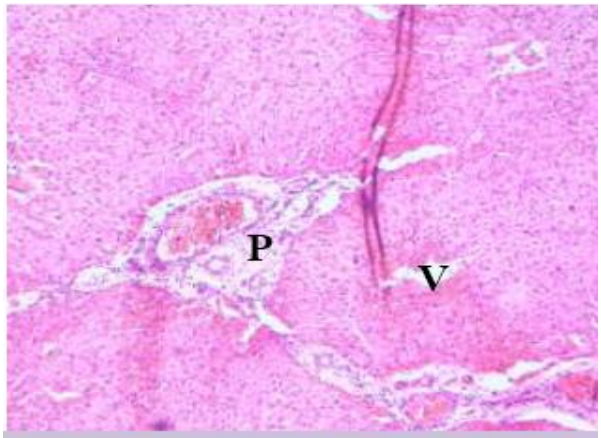

Fig 1.1: H & E tissue section of cow liver. P (portal triad), V (central vein). ×100

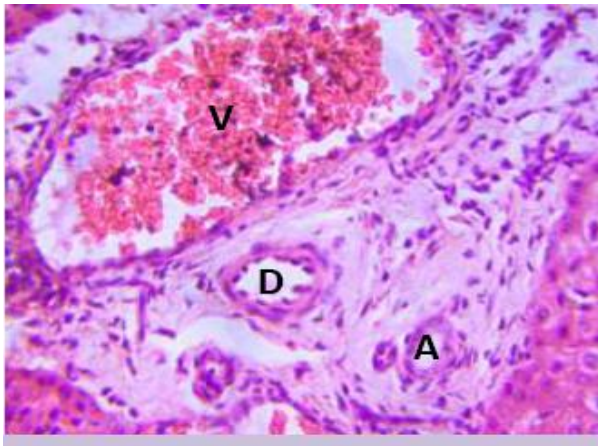

Fig 1.2: H & E tissue section of cow liver. V (portal vein), D (bile ductule), A (hepatic artery).

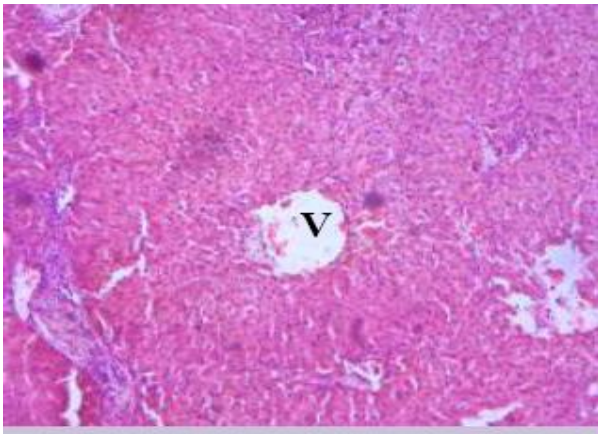

Fig 1.3: H & E tissue section of cow liver. V(central vein). ×100

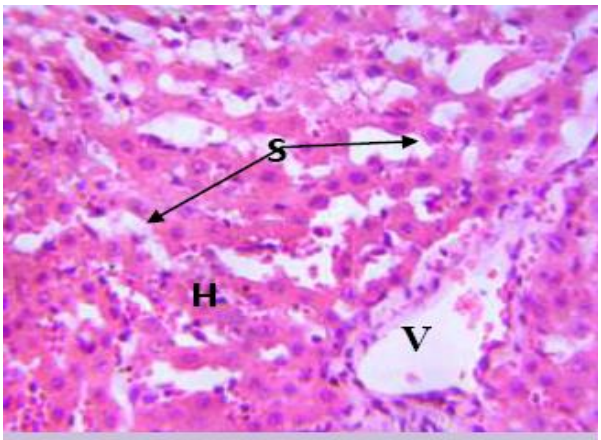

Fig 1.4: H & E tissue section of cow liver. V (central vein), S (sinusoids), H (hepatocyte). ×400

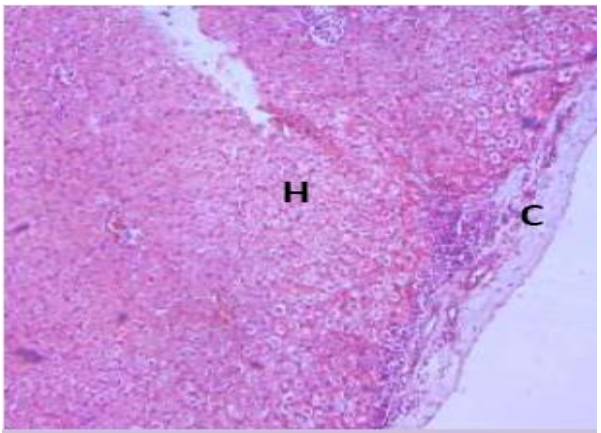

Fig 1.5: H & E tissue section of cow liver. C (capsule), H (hepatocyte). ×100

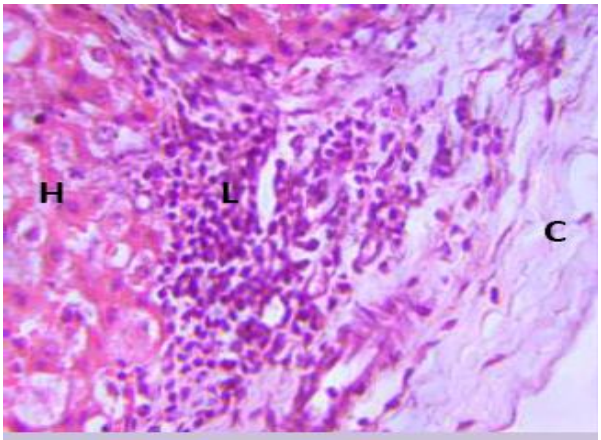

Fig 1.6: H & E tissue section of cow liver. H (hepatocyte), L (lymphoid tissue), C (capsule).

The micrograph sections, figure 1.1- 1.6 shows the hepatocytes of cow liver disposed predominantly in sheets, tubules and clusters separated by a loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids). Individual hepatocytes possess an indistinct cell border, vacuolated granular cytoplasm, which is mildly eosinophilic and a centrally placed round to oval vesicular nucleus; a nucleolus is prominent in most nuclei. Also present were portal triad in which bile ductules, hepatic artery and portal vein were embedded or enmeshed within a fibromyxoid connective tissue stroma. Occasional solitary veins of varying sizes (central veins) were also seen. The liver was enclosed by a dense

fibro-connective tissue stroma with occasional lymphoid aggregates obvious in the subcapsular space.

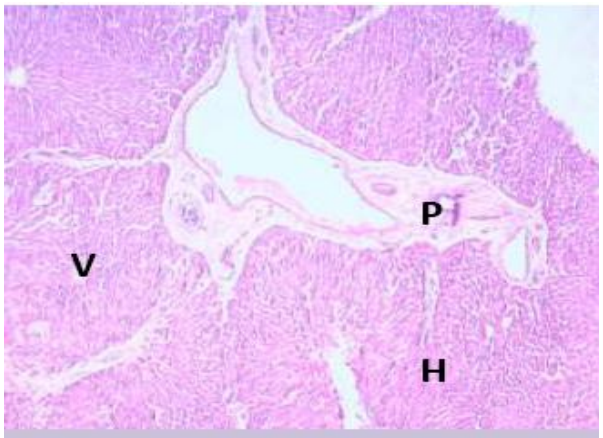

Fig 1.8: H & E tissue section of goat liver. P (portal triad), H (hepatocyte), V (central vein). ×100

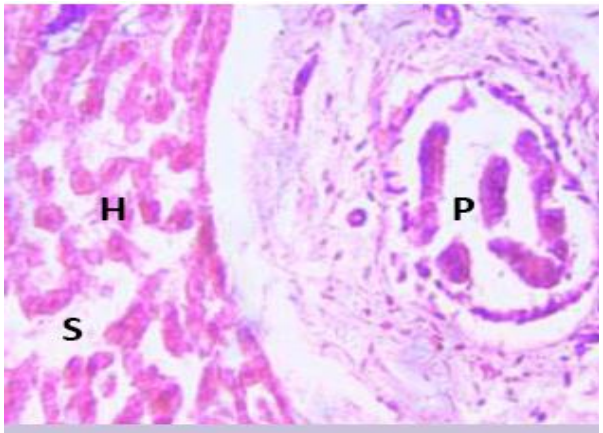

Fig 1.8: H & E tissue section of goat liver. P (portal vein), H (hepatocyte), S (sinusoid). ×400

The histological sections figure 1.7 and 1.8 shows a fibromyxoid connective tissue stroma in goat liver with hepatic artery, portal vein and bile ductule enmeshed in it. Hepatocytes with pale cytoplasm, indistinct cell borders and centrally placed vesicular nuclei were observed disposed within hepatic parenchyma, existing majorly in tubules and cords separated by loose fibroconnective tissue stroma which enclosed thin spaces (sinusoids) and converge into the central venules.

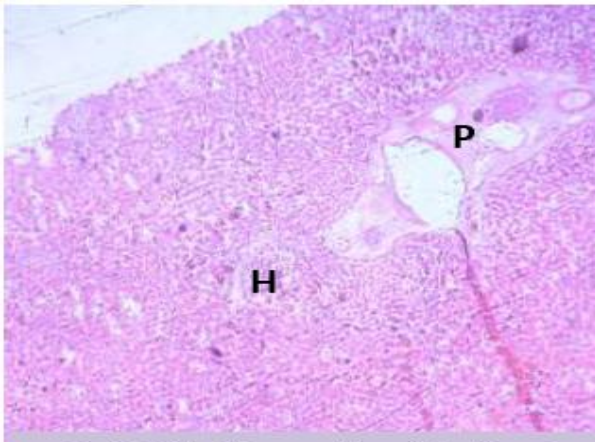

Fig 1.9: H & E tissue section of dog liver. P (portal triad), H (hepatocyte). ×40

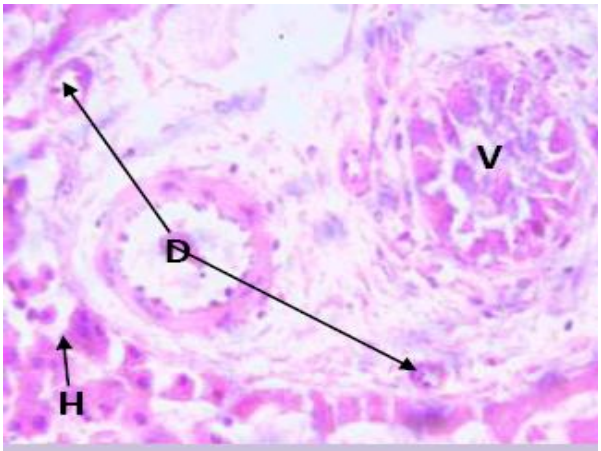

Fig 1.10: H & E tissue section of dog liver. H (hepatocyte), V (portal vein), D (bile ductule). ×400

The liver tissue sections figure 1.9 and 1.10 shows hepatocytes disposed predominantly in clusters

and few tubules in dog liver separated by a loose connective tissue stroma in which were sinusoids. Individual hepatocytes possess a distinct cell border, stained granular cytoplasm, and a centrally placed round to oval vesicular nucleus; a nucleolus is prominent in most nuclei. Scanty portal triads were present in the liver parenchyma. Central vein tributaries were also present. The hepatic lobular system is rudimentary in this case. A thin connective tissue capsule obviously enclosed the liver.

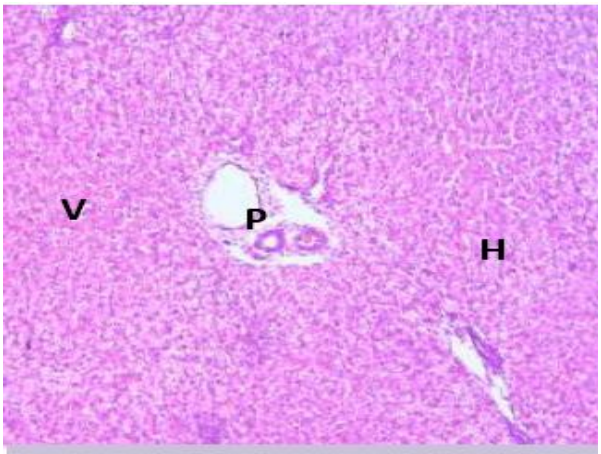

Fig 1.11: H & E tissue section of cat liver. P (portal triad), H (hepatocyte), V (central vein).

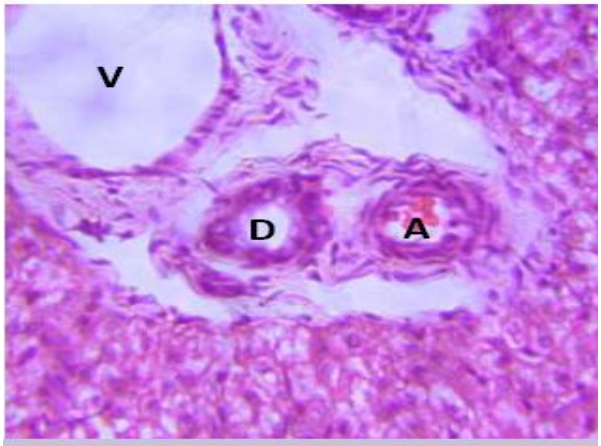

Fig 1.12: H & E tissue section of cat liver. V (portal vein), D (bile ductule), A (hepatic

The micrograph tissue sections of cat liver figure 1.11 and 1.12 shows veins of varying sizes (central veins) with hepatocytes disposed predominantly in sheets, separated by a loose fibro-connective tissue stroma which enclosed scanty thin spaces (sinusoids). Individual hepatocytes possess a distinct cell border, vacuolated granular cytoplasm, which wwere mildly esinophilic and a centrally placed round to oval vesicular nucleus; a nucleolus is prominent in most nuclei. Also present were portal triad in which bile ductules, hepatic arterioles and venules were embedded or enmeshed within a fibromyxoid connective tissue stroma. The liver

was enclosed by a dense fibro-connective tissue stroma.

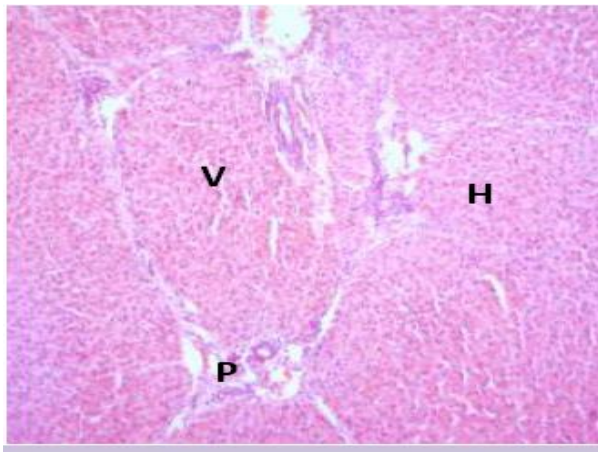

Fig 1.13: H & E tissue section of pig liver. P (portal triad), H (hepatocyte), V (central vein). ×100

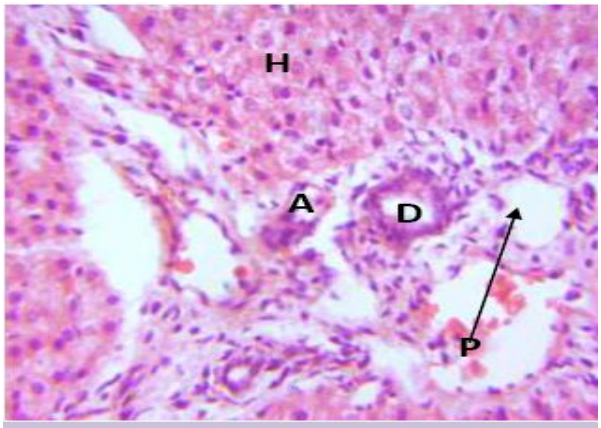

Fig 1.14: H & E tissue section of pig liver. P (portal vein), D (bile ductule), A (hepatic artery). ×400

The histological tissue sections of pig liver figure 1.13 and 1.14 shows distinct hepatic lobules with prominent intercellular fibroconnective tissue delineations. Hepatocytes disposed predominantly in tubules, separated by a loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids). Individual hepatocytes possess a distinct cell border, vacuolated granular cytoplasm, which was mildly esinophilic and a centrally placed round to oval vesicular nucleus; nucleolus was prominent in most nuclei. Also present were portal triad in which bile ductules, hepatic artery and portal vein were embedded or enmeshed within a fibromyxoid connective tissue stroma. Veins of varying sizes (central veins) were also seen. The liver was enclosed by a dense fibro-connective tissue stroma.

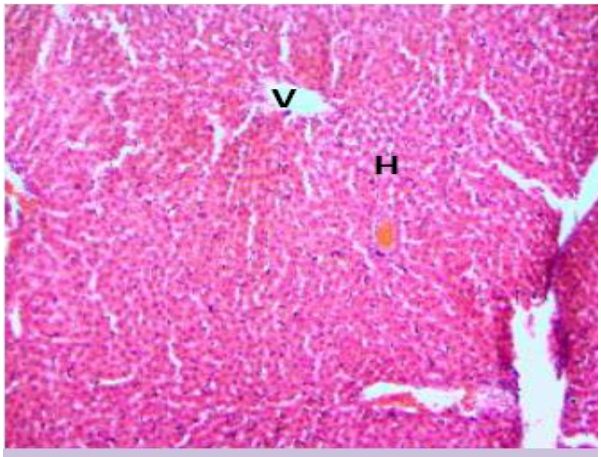

Fig 1.15: H & E tissue section of rat liver. H (hepatocyte), V (central vein). ×100

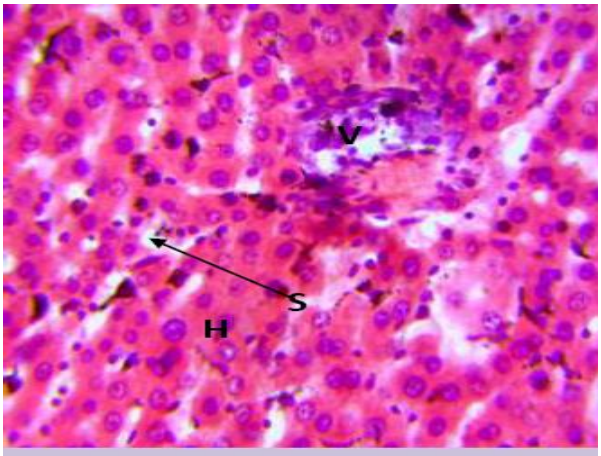

Fig 1.16: H & E tissue section of rat liver. H (hepatocyte), V (central vein), S (sinusoids). ×400

The liver tissue sections of rat figure 1.15 and 1.16 shows the liver enclosed by fibro-connective tissue stroma and the fibromyxoid connective tissue in the portal triad, enmeshed by the portal triad content; portal vein, bile ductule, and hepatic artery. Esinophilic granular cytoplasm and a centrally placed round to oval vesicular nucleus with one or two prominent nucleolus in the nuclei were also observed. The hepatocytes were disposed predominantly in sheets and tubules separated by a loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids). Individual hepatocytes possess a distinct cell border, granular cytoplasm, centrally placed round to oval vesicular nucleous; a nucleolus is prominent in most nuclei.

Periodic acid-Schiff reagent tissue stain sections

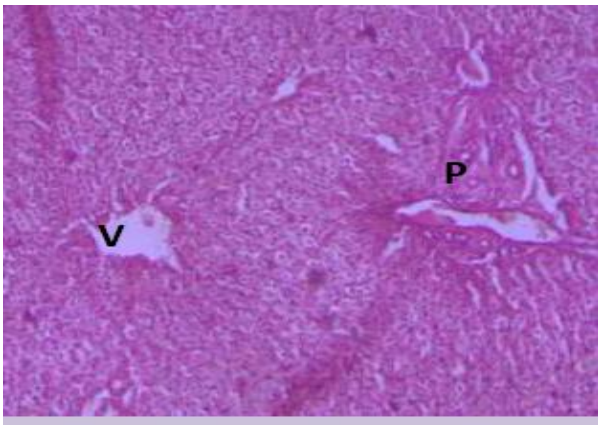

Fig 2.1: PAS tissue section of cow liver. H (hepatocyte), V (central vein) P (portal triad). ×100

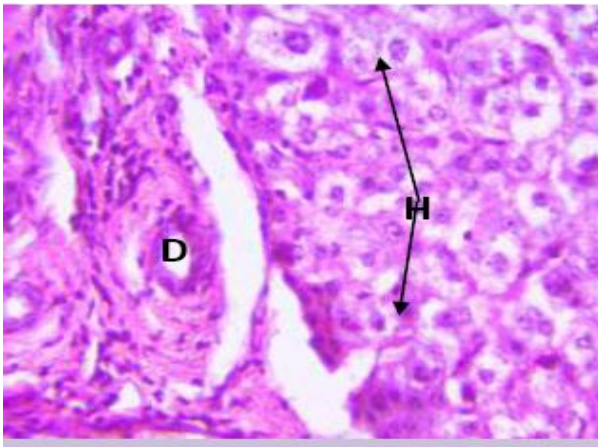

Fig 2.2: PAS tissue section of cow liver. H (hepatocyte), D (bile ductule). ×400

The micrograph sections, figure 2.1 and 2.2 shows the hepatocytes of cow liver disposed predominantly in sheets, tubules and clusters separated by a loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids) with the portal triad content; bile ductule, hepatic artery and portal vein embedded or enmeshed

within a fibromyxoid connective tissue stroma. Individual hepatocytes possessed an indistinct cell border, vacuolated granular cytoplasm with patchy stain response to PAS, some cells were barely stained with PAS and a centrally placed round to oval vesicular nucleus; a nucleolus was prominent in most nuclei.

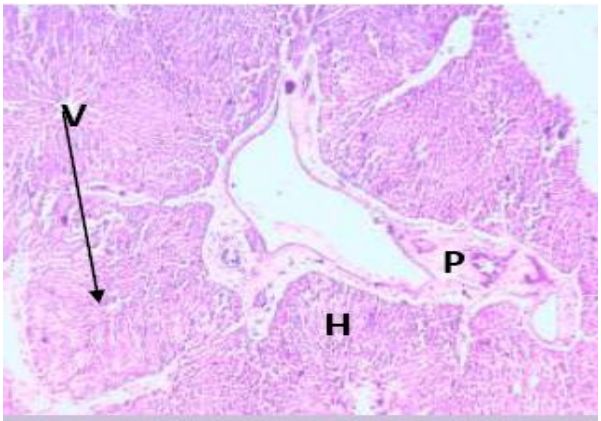

Fig 2.3: PAS tissue section of goat liver. H (hepatocyte), V (central vein) P (portal triad). ×40

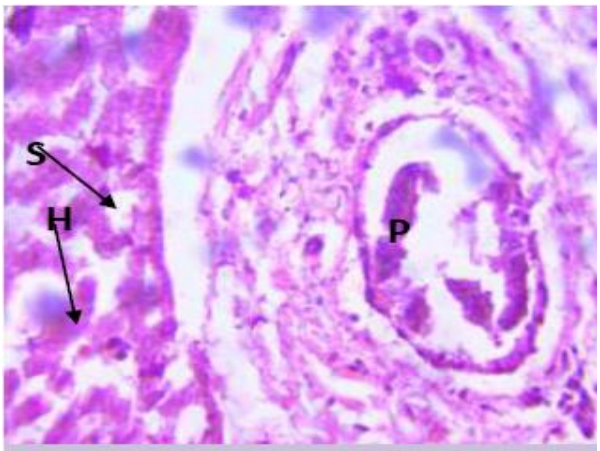

Fig 2.4: PAS tissue section of goat liver. H (hepatocyte), P (portal vein), S (sinusoid). ×400

The histological sections figure 2.3 and 2.4 shows a fibromyxoid connective tissue stroma in goat liver with hepatic artery, portal vein and bile ductule enmeshed in it. Hepatocytes with granular cytoplasm and an evenly intense stain response to PAS, indistinct cell borders and centrally placed vesicular nuclei were observed disposed within hepatic parenchyma, existing majorly in tubules and cords separated by loose fibroconnective tissue stroma which enclosed thin spaces (sinusoids) and converge into the central venules.

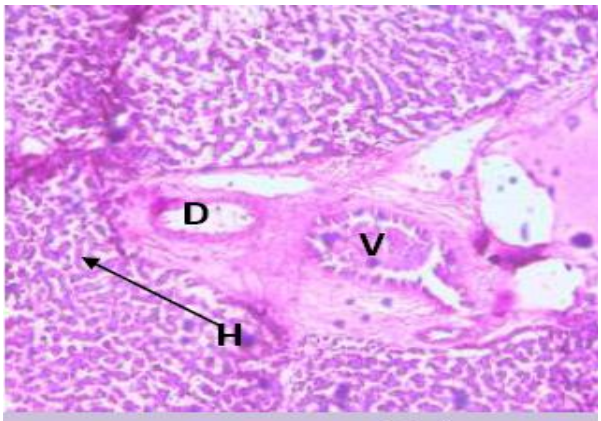

Fig 2.5: PAS tissue section of dog liver. H (hepatocyte), D (bile ductule), V (portal vein). ×100

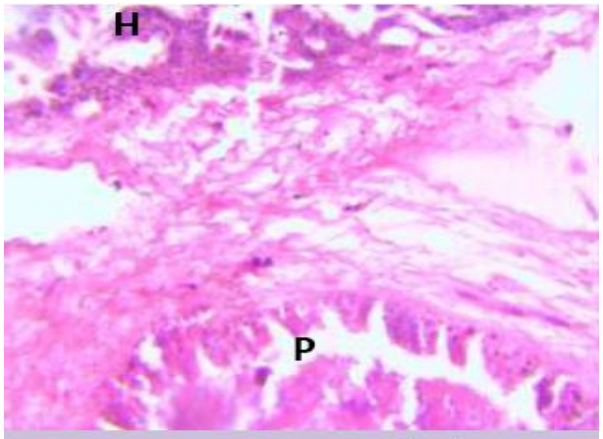

Fig 2.6: PAS tissue section of dog liver. H (hepatocyte), P (portal vein). ×400

The liver tissue sections figure 2.5 and 2.6 shows hepatocytes disposed predominantly in clusters and few tubules in dog liver, separated by a loose connective tissue stroma in which were sinusoids. Individual hepatocytes possessed a distinct cell border with nucleus and granular cytoplasmic intense stain responds to PAS. Nucleolus is prominent in most nuclei.

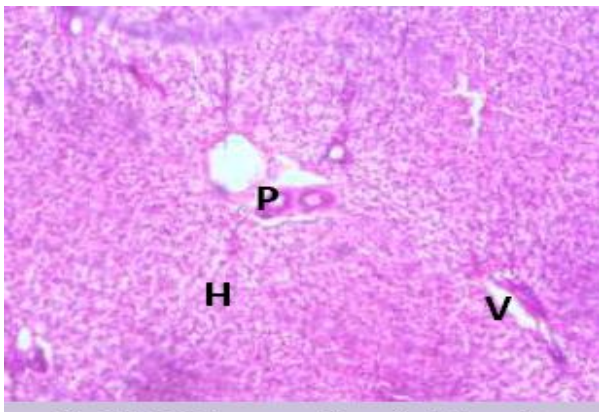

Fig 2.7: PAS tissue section of cat liver. H (hepatocyte), P (portal triad), V (portal vein). ×40

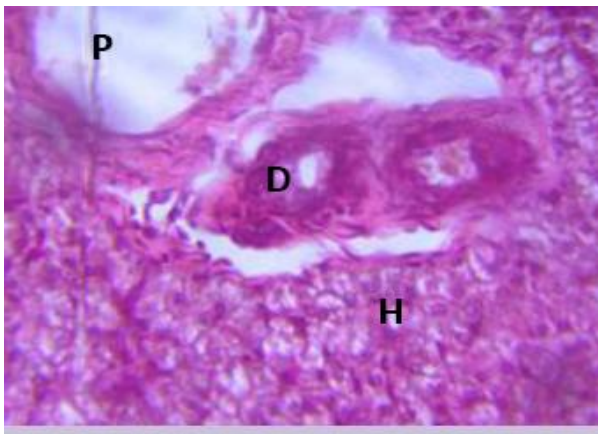

Fig 2.8: PAS tissue section of cat liver. H (hepatocyte), D (bile ductule), P (portal vein). ×400

The micrograph tissue sections of cat liver figure 2.7 and 2.8 shows veins of varying sizes (central veins) with hepatocytes disposed predominantly in sheets, separated by a loose fibro-connective tissue stroma which enclosed scanty thin spaces (sinusoids). Individual hepatocytes possessed a distinct cell borderwith granular cytoplasm and intermittent stain responds to PAS, some cells were barely stained with PAS and a nucleolus is prominent in most nuclei. Also present in the PAS stained cat liver were portal triad in which bile ductules, hepatic arterioles and venules were embedded or enmeshed within a fibromyxoid connective tissue stroma.

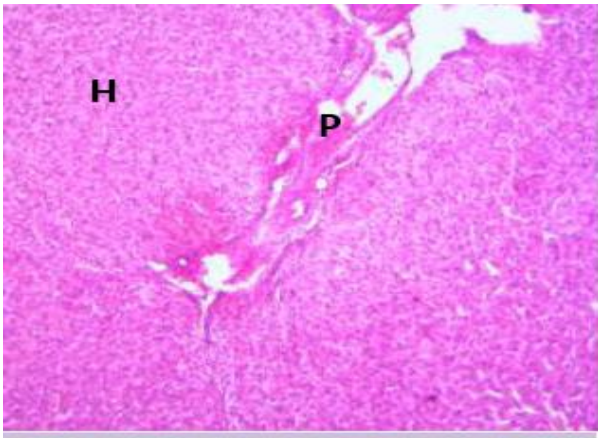

Fig 2.9: PAS tissue section of pig liver. H (hepatocyte), P (portal triad). ×100

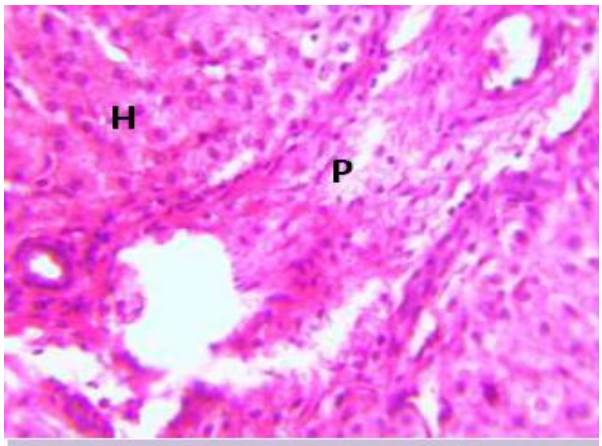

Fig 2.10: PAS tissue section of pig liver. H (hepatocyte), P (portal triad). ×400

The histological tissue sections of pig liver figure 2.9 and 2.10 shows distinct arrangement of hepatocytes in predominantly tubules, separated by a loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids). Individual hepatocytes possessed a distinct cell borderwith an intense even stain responds to PAS in the granular cytoplasm and the centrally placed round to oval vesicular nucleus; a nucleolus is prominent in most nuclei. Also present were portal triad in which bile ductules, hepatic artery and portal vein were embedded or enmeshed within a fibromyxoid connective tissue stroma.

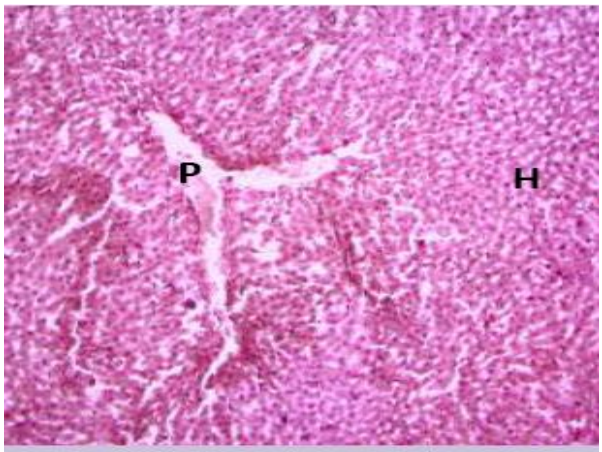

Fig 2.11: PAS tissue section of rat liver. H (hepatocyte), P (portal triad). ×100

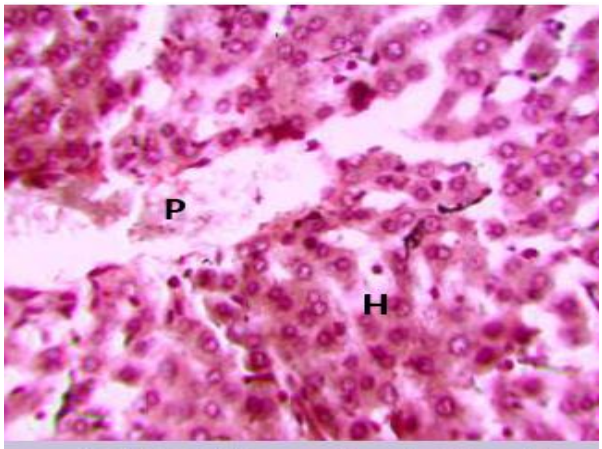

Fig 2.12: PAS tissue section of rat liver. H (hepatocyte), P (portal triad). ×400

The liver tissue sections of rat figure 2.11 and 2.12 shows the liver enclosed by fibro-connective tissue stroma and the fibromyxoid connective tissue in the portal triad, enmeshed by the portal triad content; portal vein, bile ductule, and hepatic

artery. Individual hepatocytes possessed a distinct cell border, granular cytoplasm and a centrally placed round to oval vesicular nucleus with one or two prominent nucleolus in the nuclei which intensely responds to PAS stain. The hepatocytes were disposed in predominantly sheets and tubules separated by a loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids).

Periodic acid-Schiff reagent with diastase tissue stain sections

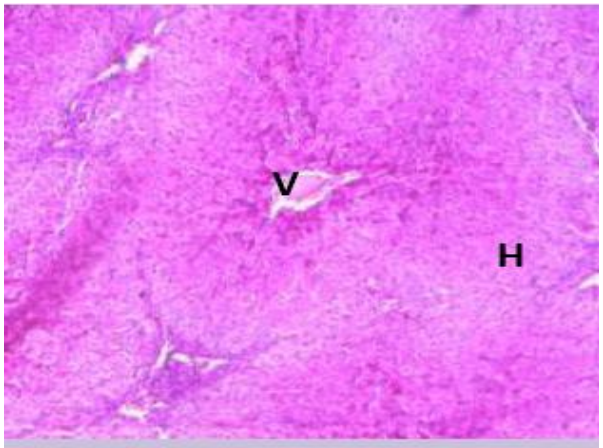

Fig 3.1: PAS-D tissue section of cow liver.
H (hepatocyte), V (central vien). ×100

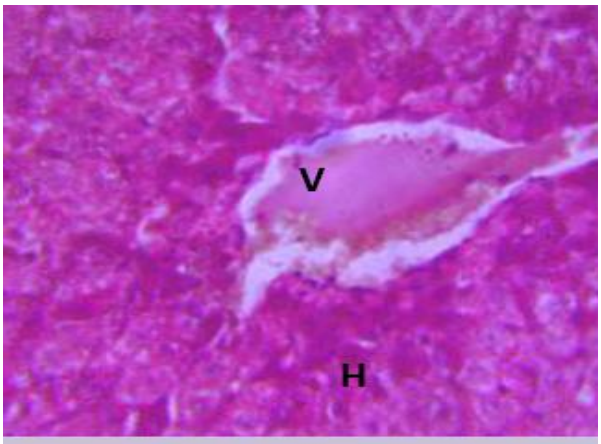

Fig 3.2: PAS-D tissue section of cow liver.
H (hepatocyte), V (central vien). ×400

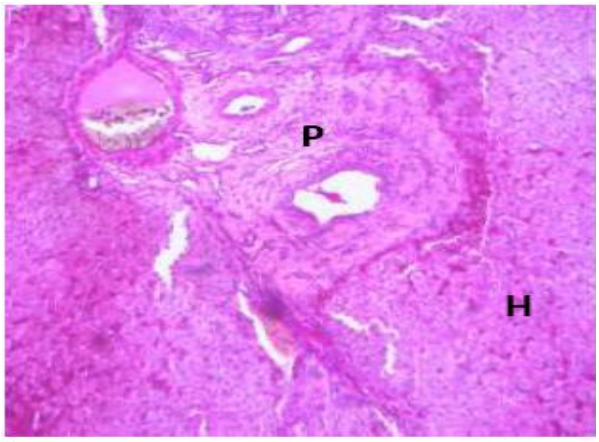

Fig 3.3: PAS-D tissue section of cow liver.
H (hepatocyte), P (portal triad). ×100

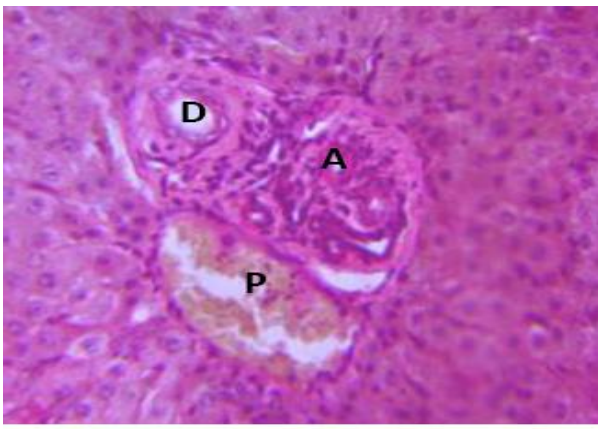

Fig 3.4: PAS-D tissue section of cow liver.
H (hepatocyte), P (portal vien), D (bile ductule),
A (artery). ×400

The micrograph sections, figure 3.1- 3.4 shows the hepatocytes of cow liver disposed predominantly in sheets, tubules and clusters separated by a loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids) with the portal triad content; bile ductule, hepatic artery and portal vein embedded or enmeshed within a fibromyxoid connective tissue stroma. The perivenular hepatocytes (surrounding the central vein tributaries) were lading with dark pigmented intracytoplasmic vesicles, although which were small in size but demonstrated round to oval plomorphism. These vesicles also demonstrated some degree of birefugence. It is noteworthy that the intensity and quantity of this vesicles increase as the hepatocytes approach the lumen of the veins. Importantly, most of these cells display moderate to max cytoplasmic vacoulation.

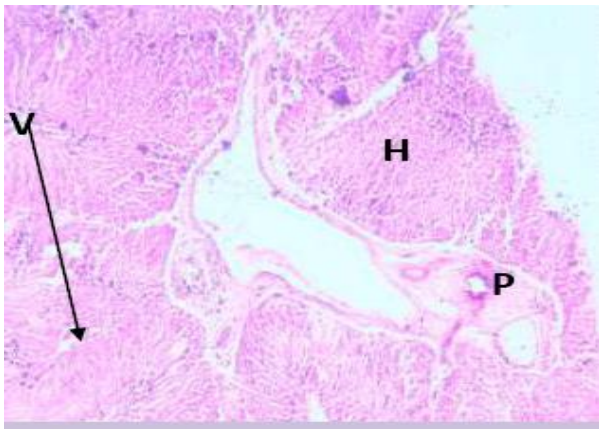

Fig 3.5: PAS-D tissue section of goat liver. H (hepatocyte), V (central vein), P (portal triad). ×40

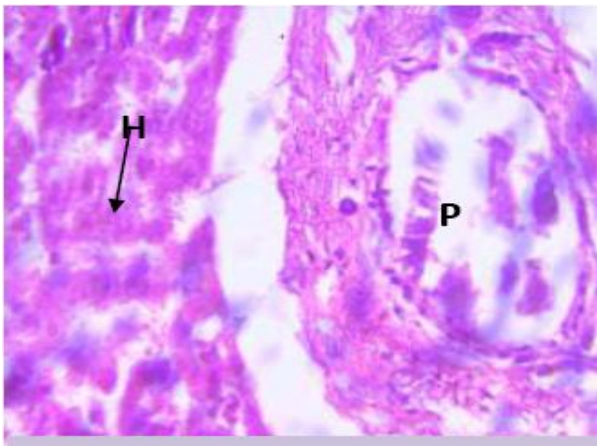

Fig 3.6: PAS-D tissue section of goat liver. H (hepatocyte), P (portal vein). ×400

The histological sections figure 3.5 and 3.6 shows a fibromyxoid connective tissue stroma in goat liver with hepatic artery, portal vein and bile ductule enmeshed in it. The hepatocytes were arranged majorly in tubules and cords separated by loose fibroconnective tissue stroma which enclosed thin spaces (sinusoids) and converge into the central venules. The hepatocytes were lading with dark pigmented intracytoplasmic vesicles,

although which were small in size but demonstrated round to oval plomorphism. These vesicles also demonstrated some degree of birefugence. The fibro-connective tissue, epithelial cells lining the bile ductules and hepatocytes responds intensely to PAS-D stain.

Fig 3.7: PAS-D tissue section of dog liver. H (hepatocyte), V (central vein), P (portal triad). ×100

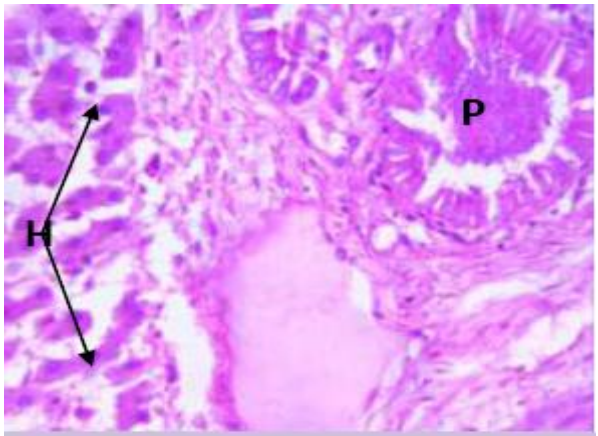

Fig 3.8: PAS-D tissue section of dog liver. H (hepatocyte), P (portal vein). ×400

The liver tissue sections figure 3.7 and 3.8 shows hepatocytes disposed predominantly in clusters and few tubules in dog liver, separated by loose connective tissue stroma in which were sinusoids. Individual hepatocytes were observed with an intense responds to PAS-D, lading with dark pigmented intracytoplasmic vesicles, although which weresmall in size but demonstrated round to oval plomorphism. These vesicles also demonstrated some degree of birefugence.

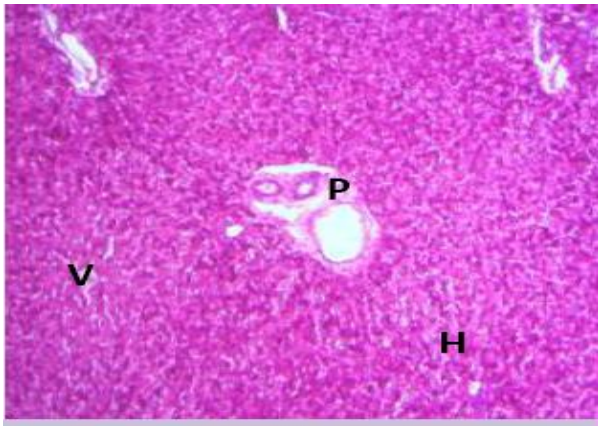

Fig 3.9: PAS-D tissue section of cat liver. H (hepatocyte), V (central vein), P (portal triad). ×100

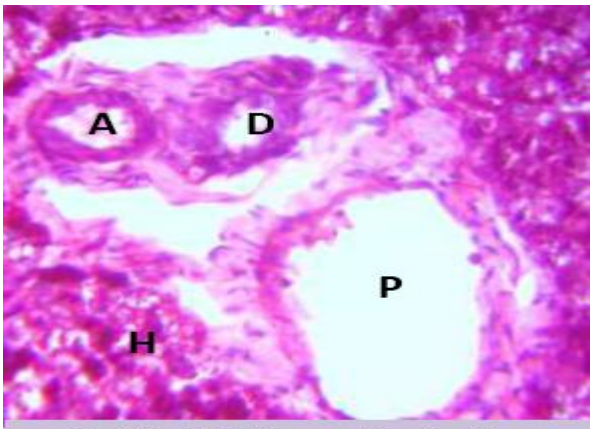

Fig 3.10: PAS-D tissue section of cat liver. H (hepatocyte), P (portal vein), D (bile ductule), A (hepatic artery). ×400

The micrograph tissue sections of cat liver figure 3.9 and 3.10 shows veins of varying sizes (central

veins) with hepatocytes disposed predominantly in sheets, separated by a loose fibro-connective tissue stroma which enclosed scanty thin spaces (sinusoids). Individual hepatocytes possessed a distinct cell border with granular cytoplasm and an intense stain responds to PAS-D. The hepatocytes were lading with dark pigmented intracytoplasmic vesicles, although which were small in size but demonstrated round to oval plomorphism. These vesicles also demonstrated some degree of birefugence.Also present were portal triad in which bile ductules, hepatic arterioles and venules were embedded or enmeshed within a fibromyxoid connective tissue stroma and their cells also responds to PAS-D stain.

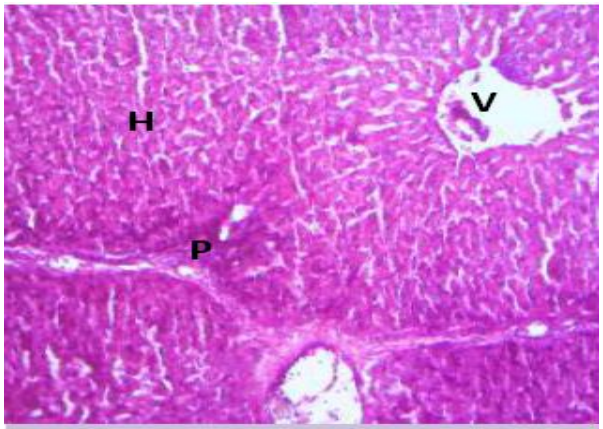

Fig 3.11: PAS-D tissue section of pig liver. H (hepatocyte), V (central vein), P (portal triad). ×100

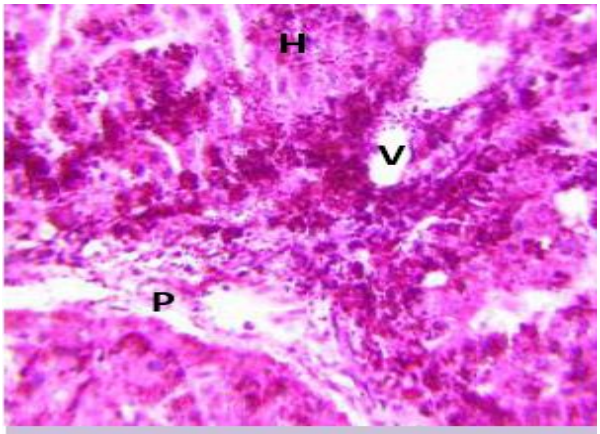

Fig 3.12: PAS-D tissue section of pig liver. H (hepatocyte), V (central vein), P (portal triad). ×400

The histological tissue sections of pig liver figure 3.11 and 3.12 shows distinct arrangement of hepatocytes predominantly in tubules, separated by loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids). Individual hepatocytes possessed distinct cell border with an intense even stain responds to PAS-D in the granular cytoplasm lading with dark pigmented intracytoplasmic vesicles, although which were small in size but demonstrated round to oval plomorphism. These vesicles also demonstrated some degree of birefugence. Also present were portal triad in which bile ductules, hepatic artery and portal vein were embedded or enmeshed within a fibromyxoid connective tissue stroma.

Fig 3.13: PAS-D tissue section of rat liver. H (hepatocyte), V (central vein), P (portal triad). ×100

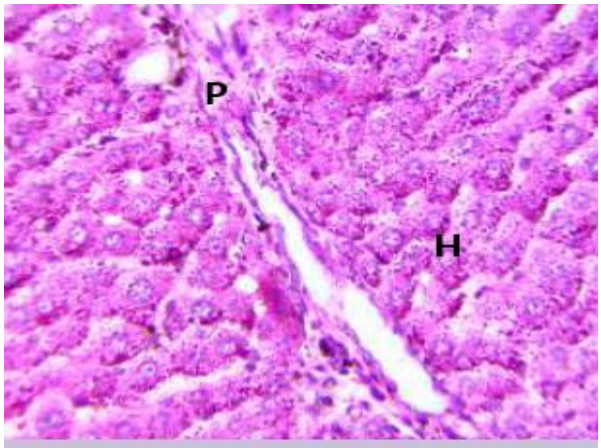

Fig 3.14: PAS-D tissue section of rat liver. H (hepatocyte), P (portal triad). ×400

The liver tissue sections of rat figure 3.13 and 3.14 shows the liver enclosed by fibro-connective tissue stroma and the fibromyxoid connective tissue in the portal triad, enmeshed by the portal triad content; portal vein, bile ductule, and hepatic artery. Individual hepatocytes possessed distinct cell border, granular cytoplasm and a centrally placed round to oval vesicular nucleus with one or two prominent nucleolus in the nuclei which intensely responds to PAS-D stain. The hepatocytes were disposed predominantly in sheets and tubules separated by a loose fibro-connective tissue stroma which enclosed thin spaces (sinusoids). The hepatocytes were lading with dark pigmented intracytoplasmic vesicles, although which were small in size but demonstrated round to oval plomorphism. These vesicles also demonstrated some degree of birefugence.

Gomori's method for demonstration of reticulin tissue stain sections

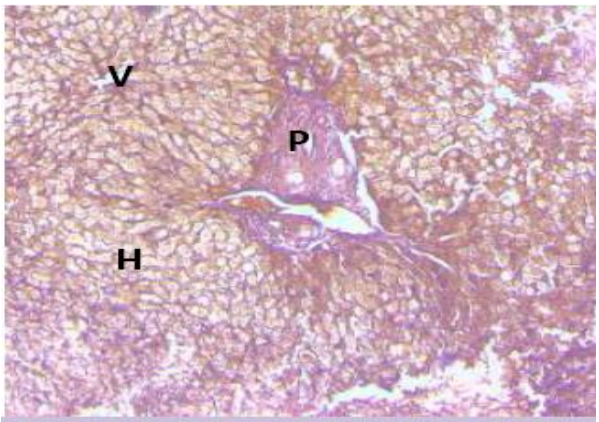

Fig 4.1: Reticulin tissue section of cow liver. H (hepatocyte), V (central vein), P (portal triad). ×100

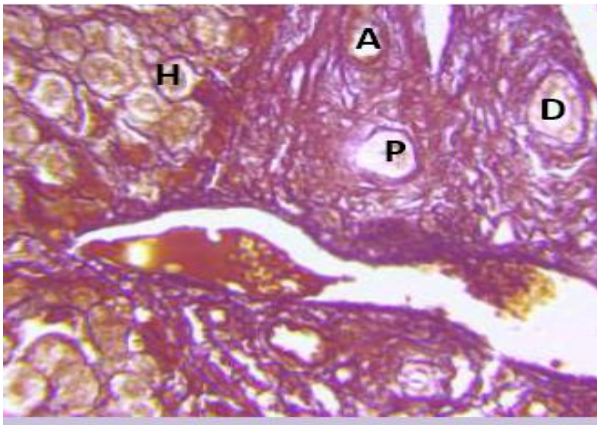

Fig 4.2: Reticulin tissue section of cow liver. H (hepatocyte), P (portal vein), D (bile ductule), A (artery). ×400

The micrograph sections, figure 4.1 and 4.2 shows the hepatocytes of cow liver disposed predominantly in sheets, tubules and clusters separated by an occasional potential and sometimes distinct sinusoidal space by their stain responds to type III collagen (reticulin). The cell membraneof individual hepatocytes responds mildly to the reticulin stain unlike the nuclear memebrane which stain more prominent. The fibromyxoid connective tissue stroma in which the portal structures are enmeshed are also stained intensely with reticulin stain.

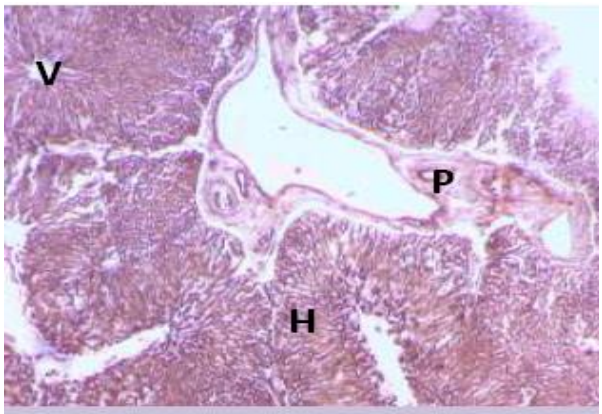

Fig 4.3: Reticulin tissue section of goat liver. H (hepatocyte), V (central vein), P (portal triad). ×40

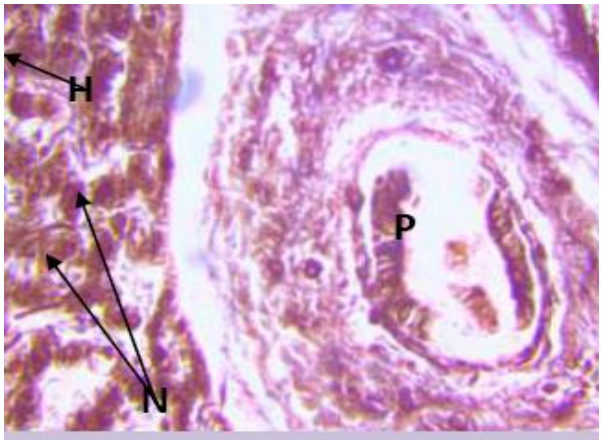

Fig 4.4: Reticulin tissue section of goat liver. H (hepatocyte), N (nucleus), P (portal vein). ×400

The histological sections figure 4.3 and 4.4 shows a fibromyxoid connective tissue stroma in goat liver with hepatic artery, portal vein and bile ductule enmeshed in it. Individual hepatocytes cell and nuclear membrane are stained but the nuclear membrane responds more intensely to reticulin stain than the cell membrane. Also seen are reticulin positively stained epithelial cells of

bile ductule and blood vessel wall. The distinct stain responds of reticulin in the sinusoidal walls, distinct the arrangements of hepatocytes which exist majorly in tubules and cords.

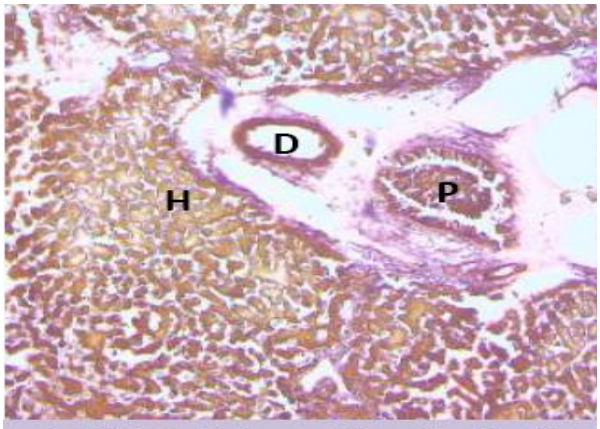

Fig 4.5: Reticulin tissue section of dog liver. H (hepatocyte), P (portal vein), D (bile ductule). ×100

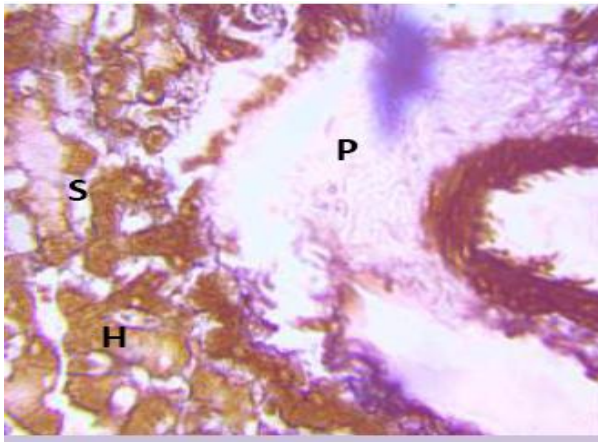

Fig 4.6: Reticulin tissue section of dog liver. H (hepatocyte), S (sinusoid), P (portal triad). ×400

The liver tissue sections of dog figure 4.5 and 4.6 shows the prominent clusters and few tubular arrangements of hepatocytes which are separated by the prominent stain responds of the sinusoidal wall type III collagen (reticulin). Individual hepatocytes cell and nuclear membrane are stained but the nuclear membrane responds more intensely to reticulin stain than the cell membrane. Also seen are reticulin positively stained epithelial cells of bile ductule and blood vessel.

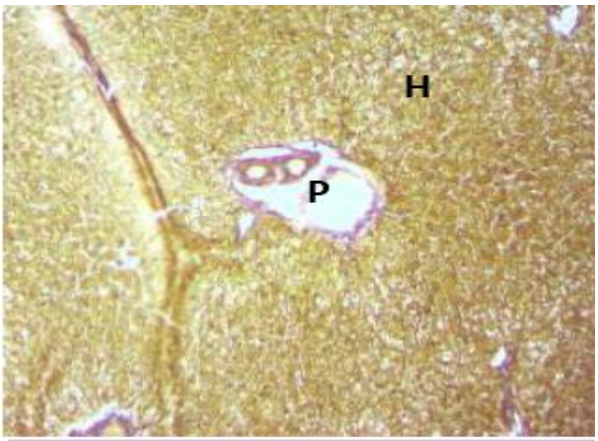

Fig 4.7: Reticulin tissue section of cat liver. H (hepatocyte), P (portal triad). ×40

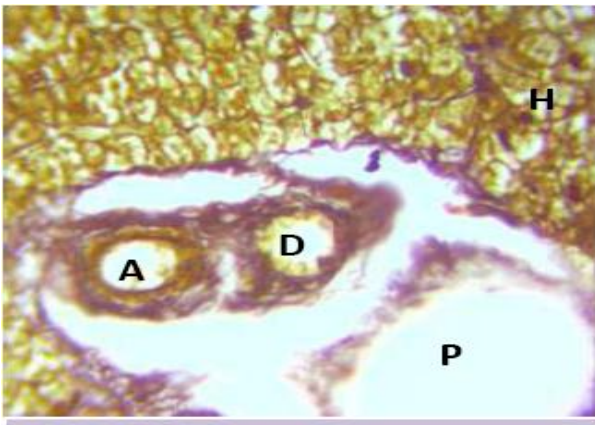

Fig 4.8: Reticulin tissue section of cat liver. H (hepatocyte), P (portal vein), D (bile ductule), A (hepatic artery). ×400

The micrograph tissue sections of cat liver figure 4.7 and 4.8 shows veins of varying sizes (central veins) with the prominent arrangement of hepatocytes in sheets which are separated by the

prominent stain responds of the sinusoidal wall type III collagen (reticulin). Individual hepatocytes cell and nuclear membrane are stained but the nuclear membrane responds more intensely to reticulin stain than the cell membrane. Also seen are reticulin positively stained epithelial cells of bile ductule and blood vessel

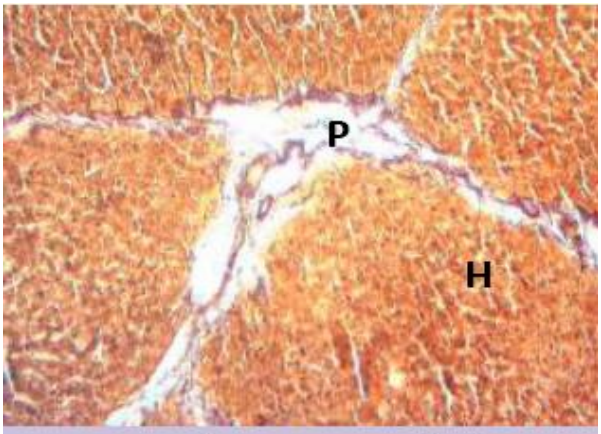

Fig 4.9: Reticulin tissue section of pig liver. H (hepatocyte), P (portal triad). ×100

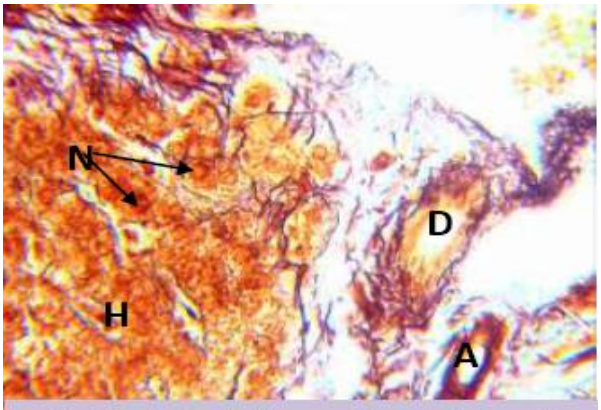

Fig 4.10: Reticulin tissue section of pig liver. H (hepatocyte), N (nucleus), D (bile ductule), A (hepatic artery). ×400

The histological tissue sections of pig liver figure 4.9 and 4.10 shows distinct arrangement of hepatocytes in predominantly tubuleswhich are separated by the prominent stain responds of the sinusoidal wall type III collagen (reticulin). Individual hepatocytes cell and nuclear membrane are stained but the nuclear membrane responds more intensely to reticulin stain than the cell membrane. Also seen are reticulin positively stained epithelial cells of bile ductule and blood vessel.

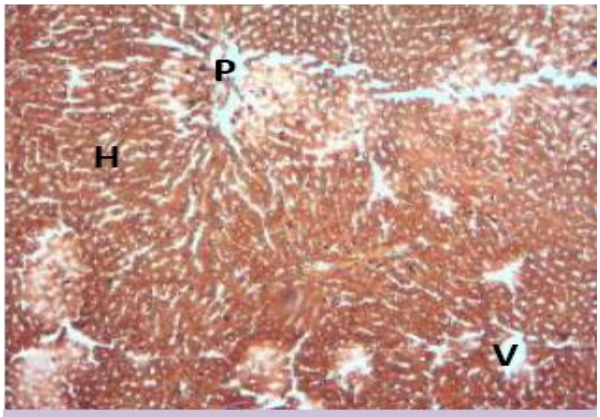

Fig 4.11: Reticulin tissue section of rat liver. H (hepatocyte), V (central vein), P (portal triad). ×100

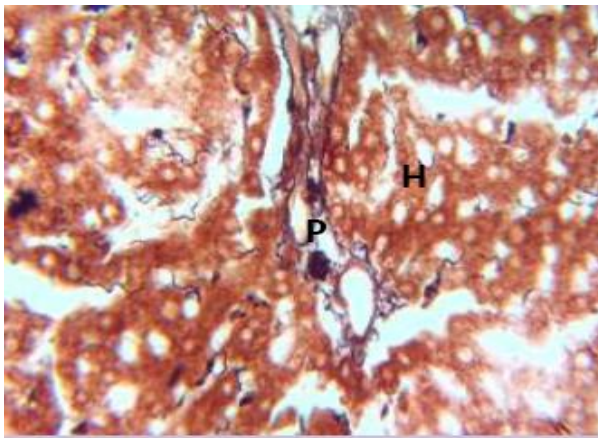

Fig 4.12: Reticulin tissue section of rat liver. H (hepatocyte), P (portal triad). ×400

The liver tissue sections of rat figure 4.11 and 4.12 shows the liver enclosed by fibro-connective tissue stroma and the fibromyxoid connective tissue in the portal triad, enmeshed by the portal triad content; portal vein, bile ductule, and hepatic artery. Individual hepatocytes cell and nuclear membrane are stained but the cell membrane responds more intensely to reticulin stain than the nuclear membrane. Also seen are reticulin positively stained epithelial cells of bile ductule and blood vessel. The hepatocytes were disposed in predominantly sheets and tubules separated by the prominent stain responds of the sinusoidal wall type III collagen (reticulin).

DISCUSSION

The histological architecture of mammalian liver was similar with little variations that explained its evolutionary trend, adaptational changes and metabolic activities peculiar to the index class of mammals. The study corroborated previous studies conducted by Madhan, Raju and Valentina which proved that all the mammalian livers showed evidence of lobular formations; central vein, portal triad, hepatocytes, sinusoids and connective tissues. Some of the liver depicted more of either three types of liver lobule; hepatic lobule, portal lobule and portal acinus.[10, 11] However the mild architectural variation established phylogenic, evolutional and developmental changes that occurred in each animal.

The liver of the six mammalian species studied were enclosed by a dense fibro connective tissue stroma and a fibromyxoid connective tissue enmeshed or embedded by the portal triad content: portal vein, bile ductule and hepatic artery. Granular cytoplasmic hepatocytes were observed with centrally placed vesicular nuclei. These findings corroborated with previous studies.[10-12,14] The hepatocytes of cow, cat and rat were observed arranged predominantly in sheets. While some hepatocytes in rat were seen arranged in tubules [14], some hepatocytes in cow were also seen arranged in tubules and clusters.[10] The few hepatocytes noticed in cow arranged in clusters and tubules were observed replicated

predominantly in dog. The hepatocytes of pig were arranged in tubules [13] similar to that of goat which was predominantly arranged in tubules with few cords.[12] These established the phylogenic categorization of the animals sharing common ancestral origin.

The developments of the different hepatocyte arrangement were sequel to the phylogenic advancement. As the phylogenic advancement is graded from low to high; dog, cow, goat, cat, rat and pig. The hepatocyte arrangement progressed from predominantly, clusters, cords, sheet to tubular form.

Dietary source is an important factor, which determines the adaptation of animals to their environment and their persistence through procreation.[15-19] In metabolism, the liver had been reported to play a vital role.

Omnivorous animals intense responds to PAS is an evidence of it unique evolutionary trend. Glucose availability for direct absorption is determined by the dietary habit of animals. [20, 21] Omnivorous animals (rat and pig) evolve to consume food high in glucose, because of its specific digestive and metabolic adaptational features; presence of glucokinase (an enzyme in the synthesis of glycogen and also glycolysis in the liver) and insulin which in turn causes accumulation of glycogen in the liver. [22]

These findings unlike carnivorous animal known for their dietary habit of consuming prey low in glucose, and their metabolic adaptational features unique in metabolizing food low in glucose is similar with that of dog liver tissue section which showed intense responds to PAS, that presaged high presence of glycogen in dog liver. These findings were explained in the studies by (20-22) which reported that dog sometimes consumes diet high in carbohydrate, as a result of the evolutionary event that led to the domestication of dog.[23-25]

Unlike dog cat liver presents mild positive reaction to PAS in the entire tissue architecture. These findings were explained by the evolutionary event that led to unique dietary

habitpeculiar to the cat species as carnivorous animals, thus, cat consumes food low in carbohydrate.[26] Cat evolved to corrode low carbohydrate prey (Small prey; Rodents and Birds) and poorly digests and metabolizes carbohydrate. Excess carbohydrate in cat could lead to obesity and diabetes mellitus.[27-31]

These findings established a relational trend between the cat and cow. The patchy stain responds to PAS in cow portends poor presence of glycogen. This could be explained by the diet of cow (fibre carbohydrate) which are less metabolized into glucose digestible by mammalian enzymes, but are stored in for estomach for microbial fermentation which is even less fermentable by microbes.[32] Glucose availability for cow in some dietary settings is low.[20] This can also be expounded by the high rate of gluconeogenesis in ruminant after eating and in period of high energy intake which caused a significant decrease in glycogen and the low activity of enzymes involve in glycogenesis.[33]The absence of an inhibitory effect of insulin on hepatic gluconeogenesis also explains the intermittent distribution of glycogen in the section.[34, 35] The increased glucose demand in dairy cow which leads to an increase in gluconeogenesis, also confirmed the reason for the patchy responds to the PAS stain. [33]

In contrast to cow, goat; mammalian specie of herbivorous animals show intense stain responds to PAS which indicates high presence of glycogen in goat liver. These findings opposed the study by [12] which reported mild PAS responds of the hepatocytes and the epithelia tissue lining the bile ductules. The intense responds of goat liver to glycogen can be expounded by the study of [20, 21] which reported that glucose availability for direct absorption is determined by the dietary habit of the animals. Unlike cow, goat feeds mostly on soluble carbohydrates and starch which are even more fermentable by microbes than fibers mostly consumed by cow.[32]

The high presence of glycogen in goat and dog liver explained the adaptational changes in their dietary habits on oppose to their dietary

classification; herbivores and carnivores respectively. The effect of environmental factors in the course of evolution that led to the domestication of these animals was the rationale for the dietary changes. [23-25]

The intense responds of PAS-D in all mammalian liver studied, denoted large amount of glycoprotein and glycolipid presence in their liver. This described the adaptational changes that occurred at the course of evolution which led to the unique digestive and metabolic features peculiar to mammals. Feed proteins of herbivores are degraded by microorganisms in the rumen via amino acid into ammonia and branching chain fatty acids. [36, 37] Phylogeneticaly, carnivorous animals fascinate prey high in protein and moderate in lipids. [26] The high presence of glycoprotein and glycolipid in omnivorous animals confirmed the dietary habit of omnivores and the adaptational changes that occurred at the course of evolution.[38]

Collagen type III (reticulin) were found in all the mammalian species studied. This portends the fact that mammalian class shares common ancestral origin. The occurrence of type III collagen (Reticulin) was restricted to the parenchyma of cow liver.[39-42] The collagen type III (Reticulin) stain which demonstrated the histoarchitecture of the lobular structure, showed, distinct stain responds of the sinusoidal wall, the epithelial cells lining the bile ductules, the nucleus and cytoplasm of individual hepatocytes.[11, 12, 43, 44]

In conclusion, the histological architecture of mammalian liver studied was similar with little variations that explained its evolutionary trend, adaptational changes and metabolic activities peculiar to the index class of mammals. The index comparative study of mammalian classification of animals indicated that mammals share near ancestral origin with differences in their adaptive features developed at the course of evolution. This buttress the fact that the variations in the digestive and metabolic activities of the species studied were accord on the dietary adaptational changes that occurred during evolution.

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