ORIGINAL RESEARCH ARTICLE

http://www.eternalpublication.com

INTERNATIONAL JOURNAL OF ANATOMY PHYSIOLOGY AND BIOCHEMISTRY

IJAPB: Volume: 2; Issue: 2; February 2015 *ISSN(Online):2394-3440*

Demonstrating glycogen at various stages of developing human liver

Published online on 2nd February 2015©www.eternalpublication.com

DR. HASHMI INTKHAB C.[1]
[1] Assistant Professor
Dept. of Anatomy, ReVera Institute of Medical Sciences and Research, Vikramghad, Thane.

Corresponding Author:

Dr. Intkhab C. Hashmi
703, B wing, Tirupati building,
Laxmi Park, Naya Nagar,
Mira Road (East)
Thane 401107 (Maharashtra, India)
+919004900568
hashmiintkhab@gmail.com

Received: 19th Jan 2015; Accepted: 28th Jan 2015

How to cite this article: Hashmi IC. Demonstrating glycogen at various stages of developing human liver. International Journal of Anatomy Physiology and Biochemistry 2015; 2(2):1-4.

Abstract:

Ninety five percent of all the glycogen stored in the body is found in the liver and muscles. Liver glycogen serves as a major source of blood glucose and is of utmost importance for supply of glucose to other organs, especially the brain. The metabolism of liver glycogen will depend on the nutritional state of the animal/human. Forty human fetuses (19 males and 21 females) of different gestational ages ranging from 12th to 36th gestational weeks were procured for the research work. Slides of liver were stained with periodic acid Schiff (PAS) stain to demonstrate the glycogen. The Diastase PAS stain was used for the demonstration of glycogen in combination with the PAS stain. Glycogen granules appeared first at 15th to 16th weeks of gestation in the form of fine granules; thereafter they increase in size till birth.

Key words: diastase PAS, glycogen granules, hepatocytes, periodic acid Schiff, PAS

Introduction:

The liver, being the principle site of many metabolic activities, has been called the '*custodian of milieu interior*'.[1] Ham and Cormack[2] (1979) said that the liver is unique; there is no division of labour between those cells that produce the exocrine secretion and those that elaborate the endocrine ones. Transport of nutrients across the hepatocytes is the key regulatory step in the fetal growth and development. Mucins and glycogen are the two main entities to be considered in tissue carbohydrate demonstration of which mucins are the largest group, comprising 'mucopolysaccharides', 'mucosubstances' and 'glyco-conjugates'. The original term 'mucin' was mentioned in a book by an American worker named Carpenter as long ago as 1846. Much later the term changed to 'mucins' as the complex nature of these substances began to emerge.[3] The glycogen macromolecules are very large and may contain over a million glucose residues. Ninety five percent of all the glycogen stored in the body is found in the liver and muscles. Glycogen in the muscles serves as an energy reserve solely for the use of the particular cell in which it occurs. Liver glycogen, on the other hand, serves as a major source of blood glucose and is of utmost importance for the supplying of glucose to other organs, especially the brain. The metabolism of liver glycogen will depend on the nutritional state of the animal/human. Rapid glycogen synthesis occurs after a carbohydrate meal, and the liver glycogen is

then later converted back to glucose in the post-absorptive state. The liver contains an enzyme, glucose-6- phosphatase, which is responsible for the converting of sugar phosphates into free glucose. This enzyme is not found in muscle or in most other tissues. (Candy[4], 1980)

The present study was carried out to demonstrate glycogen at various stages of development of liver.

Materials and Method:

Forty human fetuses (19 males and 21 females) of different gestational ages ranging from 12th to 36th gestational weeks were procured for the research work from the department of Obstetrics and Gynecology of tertiary care hospital with prior permission of the head of Department. Consent was taken from respective parents with approval of the Ethical Committee of our Medical College. These fetuses included the spontaneous abortus and stillborn. They were without any gross abnormality. Fetuses were obtained within 4-5 hrs of birth to avoid post-mortem changes. Liver was fixed in 10% formalin then was cut into pieces and fixed in Bouin's medium for 24 hrs. PAS staining method and Diastase Periodic Acid Schiff (PAS) staining was used for staining (Carleton, 1980).[5] Periodic acid Schiff (PAS) stain was used to demonstrate the glycogen. The Diastase PAS stain was used for the demonstration of glycogen in combination with the PAS stain. Glycogen is the only PAS positive component that is removed by diastase, so that for practical purposes diastase digestion converts the PAS reaction into a specific test for glycogen. It shows that if there is presence of PAS positive substances like glycogen, neutral mucosubstances, basement membranes, collagen fibers, fungal cell walls, and glycoprotein such as yolk vesicles and phospholipids; diastase PAS is performed on other sections of similar tissue block. Looked for diastase sensitivity that means there is clearing of glycogen and no other PAS positive substance and thus the presence of glycogen is confirmed. The observations were made at different gestational ages ranging from 12 weeks to 36 weeks.

Results:

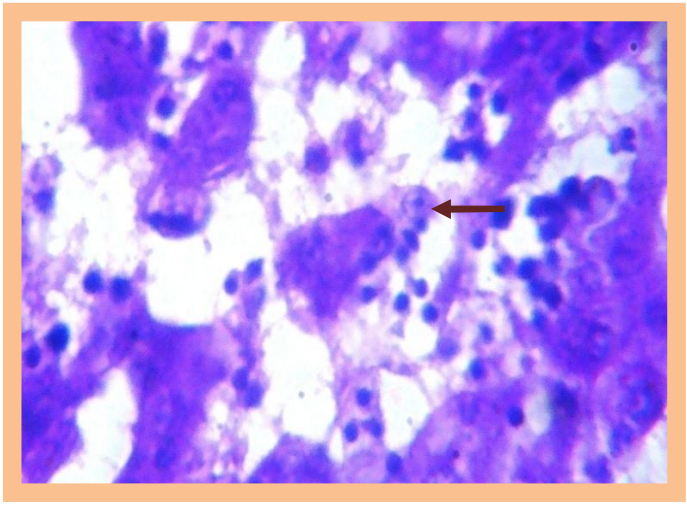
Glycogen at various stages of development of liver is as follows:

i) Liver at 12th to 14th week of gestation:

Glycogen has just appeared at this stage in the form of granules. Granules are very fine and cells containing glycogen granules are irregularly scattered. Therefore different patches of glycogen containing cells are seen.

ii) Liver at 15th to 19th weeks of gestation:

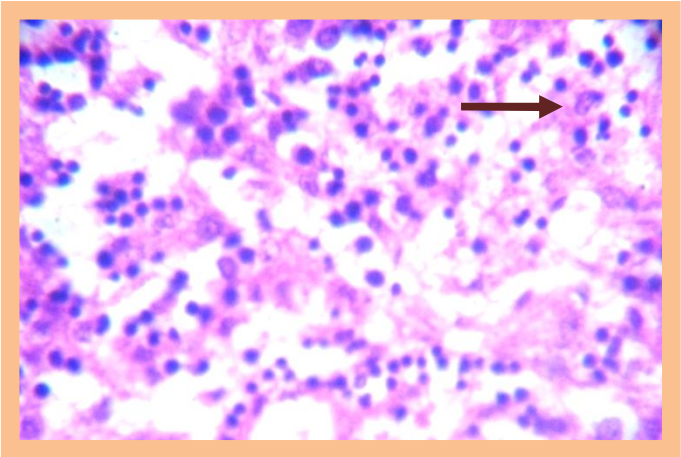
Glycogen granules can be identified very clearly at this stage. The number and size of glycogen granules are increased. Glycogen containing cells still show patchy appearance in the developing hepatic lobule, though their number increase. Concentration of glycogen granules in different cells is not uniform.



Microphotograph 1: 40X ; 18 weeks.
Hepatocytes with less glycogen granules (arrow).

iii) Liver at 20th to 23rd weeks of gestation:

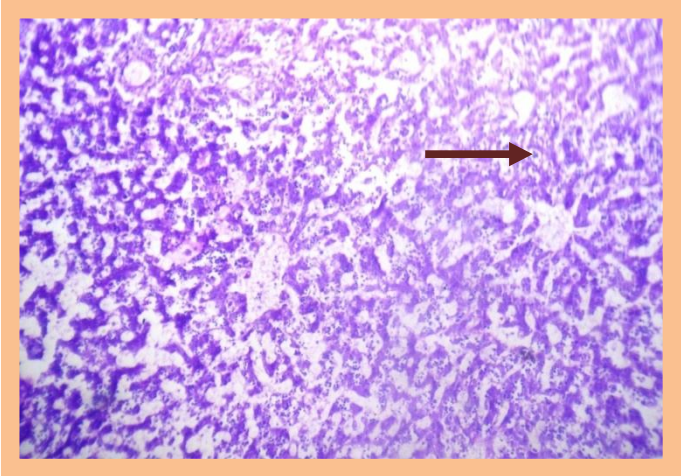
Cells containing glycogen granules still show patchy distribution in the hepatic lobule. Total concentration of glycogen granules in different cells is increased. Glycogen granules are heavily loaded in some cells. Number and size of glycogen granules are increased further.



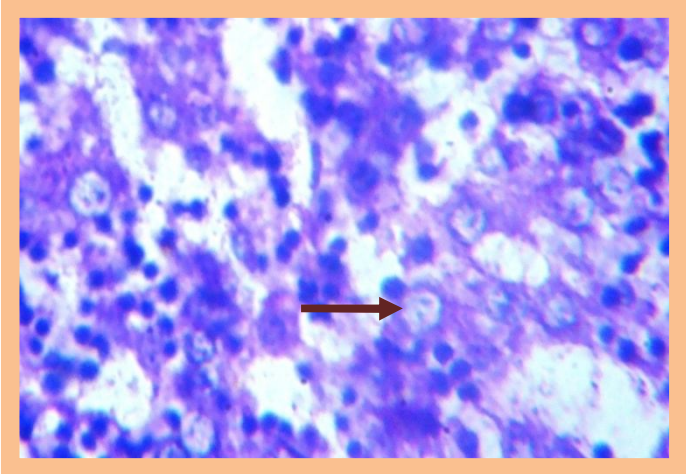
Microphotograph 2: At 40X ; 22 weeks. Hepatocytes with increased glycogen granules (arrow).

iv) Liver at 24th to 28th week of gestation:

At this stage it is clearly seen that the glycogen granules are more concentrated in the cells.



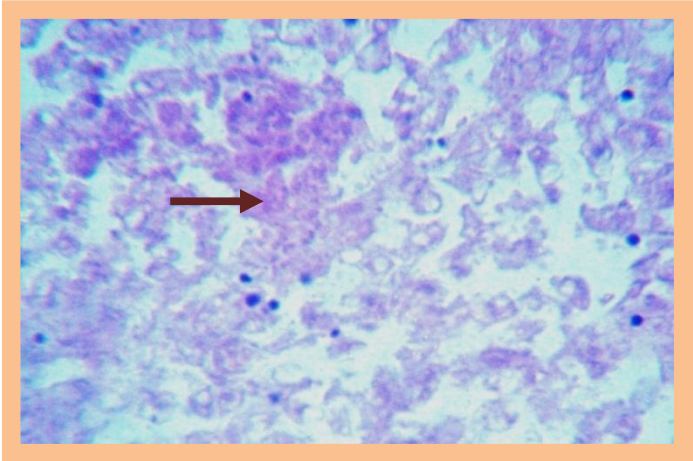
Microphotograph 3: At 10X ; 28 weeks. Glycogen accumulation is more around central vein (arrow).



Microphotograph 4: At 40X ; 28 weeks. Hepatocytes with increased glycogen (arrow).

v) Liver at 29th to 36th weeks of gestation:

In these stages the histological structure is similar to that of the previous stage but the number and size of glycogen granules show gradual increase.



Microphotograph : At 40X ; 36 weeks. Increased concentration of glycogen granules (arrow).

Discussion:

Slides of liver were stained with periodic acid Schiff (PAS) stain to demonstrate the glycogen. For confirmation of the presence of glycogen, diastase PAS was performed. The observations were made at different gestational ages ranging from 12 weeks to 36 weeks and compared with other workers.

Potter[6] (1961) mentioned that the glycogen appears at about 30th week of intrauterine life and gradually increases in amount. Glycogen is first formed in the central portion of the lobule.

Martiynyuk and Kirov Milit[7] (1966) had studied the specimens of liver of human embryo between from 7th to 8.5th months and found the cells of the centre of the hepatic lobules were more active with respect to accumulation of glycogen and bile production.

Hamilton and Mossman[8] (1975) stated that the time of hepatic glycogenesis varies between the end of 3rd month or the beginning of 4th month of intrauterine life. At term the glycogen concentrations of the fetal liver reaches 2-3 times higher than those of adult liver. Within 2-3 hours

after birth, this level falls to as low as 1/10th of full term fetus. This decrease in glycogen immediately after birth suggests that glycogenolysis ensures an adequate blood glucose level and supplies a major source of energy to the neonate.

Kovanov, Korolev, Razumnaia, Zymaleva[9] (1975) studied the structure of hepatic cells and reported that the glycogen granules were found in liver at 7-8 weeks of intrauterine life with increase in their amount as the term advanced.

Moussa[10] (1996) had studied the early histogenesis in human embryonic liver and found intensive reaction for glycogen in the hepatocytes.

In the present study also, glycogen granules were first seen in the 15th week of gestation. Glycogen containing hepatocytes in younger fetuses about 23 weeks show a patchy presence. There are some places where the glycogen granules are heavily loaded in the cells while others are devoid of any. From 26th to 36th week glycogen granules concentration increases in the hepatocytes.

Conclusion:

Glycogen granules appeared first at 15th to 16th weeks of gestation in the form of fine granules; thereafter they increase in size till birth.

References:

1. Rezek R, Philipp, Millard M. In autopsy pathology, Springerfield, Thomas 1963:464-467.
2. Ham AW, Cormack DH. Histology: The pancreas, liver and gallbladder. 8th edn. J.B.Lippincott Company. Philadelphia and Toronto 1979:694-724.
3. Bancroft JD, Gamble M. Theory and Practice of Histological Techniques: mucins. 5th Edn. Churchill Livingstone, London 2002:163-200.
4. Candy DJ. Biological functions of carbohydrates. Blackie and Son Limited, Glasgow and London 1980:3-40.
5. Drury RAB, Wallington EA. Carleton's Histological Technique. 5th edn. Oxford University press. Oxford, New York, Toronto 1980:125-150, 232-259.
6. Potter EL. Rate of antenatal growth. In pathology of fetus and infant. 2nd edn. Year Book Medical Publishers, Chicago 1961:10-14.
7. Martiynyuk VA, Kirov Milit SM. Cytogenesis of the secretary epithelium of the liver of man and other mammals. Arkh Anat Gistol Embryol. 1966;50(2):39-47.
8. Hamilton WJ, Mossman HW. Hamilton, Boyd and Mossman's Human Embryology. 4th edn. MacMillan Press Ltd. and The William & Wilkins Company, Landon 1975:339-348.
9. Kovanov VV, Korolev VV, Razumnaia TA, Zymaleva OG. Ultrastructure of human hepatocytes in early prenatal ontogenesis. Arkh Ant Gistol Embryo 1975;69(8):5-10.
10. Moussa L. Early histogenesis in human embryonic liver. Folia Med (Plovdiv) 1996;38(3-4):21-7.