



Original Research Article

Clinicopathological study of liver diseases in paediatric age group

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ABSTRACT

Context: Liver disorders occupy a noteworthy proportion amongst the aetiologies for childhood morbidities and mortalities. They have characteristic histological features whose timely reporting can abet the further management and overall prognosis of the conditions.

Aims: 1. To study the clinicopathological features of liver diseases in paediatric patients. 2. To analyse the histological presentation of various liver conditions in paediatric patients. 3. To assess the pattern of distribution of the conditions in different age groups of paediatric patients.

Materials and Methods: A retrospective and prospective observational study of liver specimens of children, satisfying the inclusion and exclusion criterias, sent to Pathology department, BVDUMC, Pune over a span of 48 months.

Statistical Analysis used: SPSS (Statistical Package for social sciences) version 24.0 software.

Results: Total 34 cases were studied of which 31 were liver biopsies and 03 were resections. Most common age group was between one month to two years. Male to female ratio was 1.6:1. Most common clinical feature was icterus, LFT was deranged in 79.41% cases. Most common histopathology amongst non neoplastic cases was cholestasis and amongst neoplasms was Hepatoblastoma. Histopathology and radiology impression correlated in 94.12% cases. Bile duct proliferation, presence of intracanalicular bile plugs, portal tract fibrosis, presence of extramedullary hematopoiesis (EMH) and intracellular bile stasis all were statistically significant ($p < 0.05$) in cases of EHBA while presence of bile plugs, intracellular bile stasis, EMH and giant cell transformation was statistically significant ($p < 0.05$) in cases of NH and presence of Intracanalicular as well as intracellular bile stasis were statistically significant ($p < 0.05$) with respect to cases showing cholestasis.

Conclusions: Thus, in children presenting with jaundice, hepatomegaly or other clinical presentations of underlying liver disease, liver biopsy can aid in understanding the underlying pathology in order to reach a definitive diagnosis and thus aid in proper management of patients in liver diseases.

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1. Introduction

Liver disorders occupy a noteworthy proportion amongst the etiologies for childhood morbidities and mortalities. Liver diseases such as cholestasis, cirrhosis, hepatitis, biliary atresia, non-alcoholic fatty liver disease, non-alcoholic steatohepatitis, tumours like hepatoblastoma, hepatocellular carcinoma as well as metabolic disorders such as Wilson's disease, glycogen storage disorder, hemosiderosis etc have

characteristic histological features¹ whose timely reporting can abet the further management and overall prognosis of the condition.

These histological features include the presence of definite features like fibrosis, fatty change, ballooning degeneration, distortion of architecture of liver parenchyma, bile stasis, bile plugs and necroinflammation to name a few.² These features are reviewed along with an evaluation of portal tracts, lobules and the central vein.¹

The etiology of the disease can be assessed using liver biopsy in children presenting with signs and symptoms such

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as jaundice, hepatomegaly, pain in abdomen, ascites which are suggestive of underlying liver conditions. Liver biopsies are of significance in paediatric health care as they serve the dual purpose of being an excellent diagnostic tool while also aiding the assessment of the severity of the histopathological lesions identified.³

2. Materials and Methods

A retrospective and prospective observational study was carried out on all liver biopsies and liver resection specimens sent to the department of pathology for histopathological examination from May 2016 till May 2020. The patient selection was based on inclusion and exclusion criteria. For retrospective evaluation slides and requisition forms was collected from the Pathology department. The required history and clinical details was collected from the medical records department of Bharati Hospital.

Histopathological specimens was processed as per the standard methods. The tissue obtained from the biopsy was kept in 10% neutral buffered formalin. After processing, paraffin blocks were made and the sections were cut at 4 to 5 micron thickness and stained with haematoxylin and eosin and other necessary special stains where indicated.

The sections were studied to form a histopathological report. The data thus generated was correlated with the clinical and investigational lab data collected to form the patient information sheet. The results were studied to reach a conclusion to the extent feasible about the etiopathogenesis and the pattern of distribution of the liver conditions in different age groups

The age distribution was divided into the following sub groups:

- A) Birth to 1 month
- B) 1 month to 1 year
- C) 1 year to 5 years
- D) 5-16 years

Tools for data collection: Proforma consisting of:-

- A) Patients demographic and clinical details.
- B) Histopathological reports.
- C) Laboratory and other relevant investigational reports:

Relevant biochemical and hematological assays: LFT, coagulation profile.

Microbiological investigations: Relevant viral markers (eg: Hepatitis, TORCH infection).

Relevant radiological reports.

The data was analyzed using SPSS (Statistical Package for social sciences) version 24.0 software. For Qualitative data various rates, ratios and percentage (%) were calculated. A two tailed test with P – value <0.05 was considered as significant.

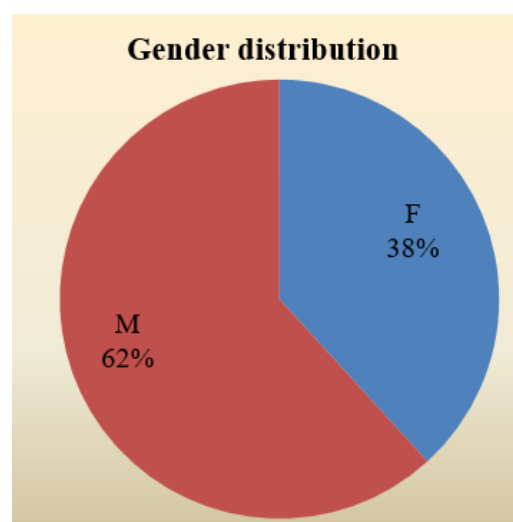
3. Results

It included the study of 34 pediatric patients with liver disease.

Table 1: Age group wise incidence

Age	No. of cases	Percentage (%)
Birth-1 month	4	11.76
1 month-1 year	26	76.47
1 year-5 years	1	2.94
5-16 years	3	8.82
Grand Total	34	100.00

Majority of cases belonged to age group of 1 month-1 years (76.47%) followed by Birth-1 month age group (11.76%).



Graph 1: Gender distribution of the cases

Majority of the patients were males (61.76%) when compared to females (38.24%). Male to female ratio was 1.6:1.

Table 2: Clinical presentations of cases

Clinical presentations	No. of cases	Percentage
Icterus	29	85.29
Clay colored stools	13	38.24
Hypoglycemia	3	8.82
Variceal bleed	3	8.82

Most common clinical presentation was icterus seen in 29 cases (85.29%), followed by clay colored stools in 13 cases (38.24%).

Hepatomegaly was present in 16 cases (47.06%) and absent in 18 cases (52.94%).

Deranged LFT were noted in majority of the patients (79.41%).

Viral markers were reactive in only 03 cases (8.82%), all 03 cases were reactive for anti CMV IgG.

Table 3: Laboratory profile of the cases

Laboratory investigations		No. of case	Percentage
LFT	Deranged	27	79.41
	WNL	7	20.59
Viral Markers	Reactive (anti CMV Ig G+)	3	8.82
	NR	31	91.18
Coagulation Profile	Deranged	8	23.53
	WNL	26	76.47

Coagulation profile were within normal limits in majority of the patients (76.47%) and were deranged in 08 cases (23.53%).

Table 4: Radiological spectrum of diseases

Radiological impressions	No of Cases	Percent (%)
Non Neoplastic		
Biliary atresia	18	52.94
Liver parenchymal disease	7	20.59
Biliary atresia + polysplenia	1	2.94
Biliary atresia + cystic kidney	1	2.94
A-V Malformation	1	2.94
Neoplastic		
Hepatoblastoma	3	8.82
Metastasis	2	5.88
Embryonal sarcoma	1	2.94
Total	34	100.00

Radiologically out of 34 cases, 28 (82.35%) were diagnosed as non neoplastic and 06 (17.65%) were diagnosed as neoplastic.

Biliary atresia was the commonest impression seen in 18 (52.94%) followed by Liver parenchymal diseases in 7 (20.59%) cases.

Table 5: Histopathological division of cases

HPE Diagnosis	No of Cases	Percent
Non neoplastic	30	88.24
Neoplastic	4	11.76
Total	34	100.00

Histopathologically out of 34, 30 (88.24%) cases were diagnosed as non neoplastic and 04 (11.76%) were diagnosed as neoplastic.

Cholestasis was seen in 14 (41.18%) cases followed by equal frequency of EHBA and Neonatal hepatitis seen in 10 cases (29.41%) each.

Histopathology and radiology impression correlated in 32 cases (94.12%) and were non correlated in 02 cases (5.88%).

Histopathology and radiology impression correlated in 32 cases (94.12%) and were non correlated in 02 cases (5.88%).

Table 6: Histopathological spectrum of diseases

HPE Diagnosis	No of Cases	Percentage
Non Neoplastic		
Cholestasis	14	41.18
EHBA	10	29.41
Neonatal hepatitis	10	29.41
Cirrhosis	4	11.76
Glycogen storage disease	2	5.88
Portal fibrosis	2	5.88
Near normal	2	5.88
Near Normal +No metastasis	1	2.94
A-V Malformation	1	2.94
Neoplastic		
Hepatoblastoma	2	5.88
Embryonal sarcoma	1	2.94
Metastasis from neuroendocrine tumor	1	2.94

Table 7: Correlation between radiology and histopathology

Histopathology vs radiology	No of cases	Percentage
Correlated	32	94.12
Non correlated	2	5.88
Total	34	100.00

Table 8: Correlation between radiology and histopathology

Radiological Impression	HPE Diagnosis		Total
	Neoplastic	Non neoplastic	
Neoplastic	4	2	6
Non neoplastic	0	28	28
Total	4	30	34

By application of Kappa test: Kappa coefficient (k)= 0.767 (0.463-1: Good agreement).

Thus, good agreement was noted between radiological and histopathological reporting upto the level of classifying the condition as neoplastic or non-neoplastic.

Microscopic study showed that bile duct proliferation, presence of intracanalicular bile plugs, portal tract fibrosis, presence of extramedullary hematopoiesis (EMH) and intracellular bile stasis all were statistically significant in cases of EHBA.

Effaced architecture, necrosis, degeneration of hepatocytes were present in majority of cases of EHBA, it was not statistically significant with respect to EHBA cases.

Microscopic study showed presence of bile plugs, intracellular bile stasis, EMH and giant cell transformation was statistically significant in cases of NH.

Effaced architecture, necrosis, degeneration of hepatocytes were present in majority of cases, it was not statistically significant with respect to NH cases.

Out of 34 cases, cirrhosis was seen in 04 cases. None of the microscopic features were statistically significant with

Table 9: Histological features in cases of EHBA

Microscopy	EHBA		Total	p value
	Present	Absent		
Effaced architecture	8	2	10	0.052
Bile duct proliferation	10	0	10	<0.001*
Bile plugs (Intracanalicular)	10	0	10	0.038*
Fibrosis	9	1	10	0.013*
Necrosis				
Spotty	4	2	10	0.21
Interface hepatitis	4			
Cell plate				
Degeneration	9	1	10	0.522
Steatosis	0	10	10	0.209
GCT	3	7	10	0.853
EMH	7	3	10	0.014*
Bile stasis (Intracellular)	10	0	10	0.038*

Table 10: Histological features in cases of Neonatal hepatitis

Neonatal Hepatitis				
Microscopy	Present	Absent	Total	p value
Effaced architecture	7	3	10	0.242
Bile duct proliferation	3	7	10	0.853
Bile plugs (Intracanalicular)	10	0	10	0.038*
Fibrosis	5	5	10	0.53
Necrosis				
Spotty	2	3	10	0.103
Interface hepatitis	5			
Cell plate				
Degeneration	10	0	10	0.092
Steatosis	0	10	10	0.209
GCT	10	0	10	<0.001*
EMH	7	3	10	0.014*
Bile stasis (Intracellular)	10	0	10	0.038*

Table 11: Histological features in cases showing cirrhosis

Microscopy	Cirrhosis		Total	p value
	Present	Absent		
Effaced architecture	4	0	4	0.052
Bile duct proliferation	3	1	4	0.050
Bile plugs (Intracanalicular)	3	1	4	0.901
Fibrosis	4	0	4	0.069
Necrosis				
Spotty	3	0	4	0.104
Interface hepatitis	1			
Cell plate				
Degeneration	4	0	4	0.347
Steatosis	1	3	4	0.267
GCT	0	4	4	0.139
EMH	0	4	4	0.089
Bile stasis	3	1	4	0.901

Table 12: Histological features in cases showing Cholestasis

Microscopy	Cholestasis			p value
	Present	Absent	Total	
Effaced architecture	5	9	14	0.052
Bile duct proliferation	4	19	14	0.690
Bile plugs (Intracanalicular)	14	0	14	0.006*
Fibrosis	8	6	14	0.925
Necrosis				
Spotty	7			
Interface hepatitis	2	5	14	0.133
Cell plate				
Degeneration	13	1	14	0.217
Steatosis	1	13	14	0.665
GCT	3	11	14	0.242
EMH	5	9	14	0.756
Bile stasis (Intracellular)	14	0	14	0.006*

respect to cirrhosis.

Out of 34 cases, cholestasis was seen in 14 cases. Microscopic study showed that presence of Intracanalicular as well as intracellular bile stasis were statistically significant in with respect to cases showing cholestasis

In age group of Birth-1 month, Cholestasis related to EHBA and NH was the commonest finding.

In age group of 1 month-1 year, Cholestasis was the commonest finding seen in 12 cases, followed by equal incidence of EHBA and NH.

In age group of 1 year-5 year, 01 case of portal fibrosis was studied.

In age group of 5-16 years, 01 case each of embryonal sarcoma and 01 case of near normal biopsy was studied.

3.1. Neoplastic cases

Total 04 cases of neoplasm were received, 02 of which were Hepatoblastoma, 01 was Embryonal sarcoma and 01 was from neuroendocrine tumor.

3.2. Resections

03 resections were received 01 was that of Hepatoblastoma, 01 was an A-V malformation and 01 was embryonal sarcoma.

4. Discussion

These observations were made in 34 liver specimens (Biopsies & Lobectomy) received at Department of Pathology, Bharati Vidyapeeth Deemed University Medical College and Bharati Hospital, Pune over a course of 48 months.

Incidence of variceal bleed in studies by both Dhole et al. (2015) and Hanif et al. (2004) were 28% and 42% respectively which was much higher than result of 8.82% in the present study. This can be explained by the low incidence of cirrhosis and associated portal hypertension in

the present study.

In present study deranged LFT was the commonest finding amongst laboratory investigations which was in seen in 79.41% of the cases, this was similar to other studies all of which showed deranged LFT in a vast proportion of their cases.

Anti-CMV IgG antibody was the only positive viral marker and was seen in 8.82% of cases.

A study done by Shibata et al. (2005)¹¹ showed 23% of cases of infantile hepatitis were positive for CMV DNA, thus indicating that CMV could be major pathogen responsible for hepatitis in pediatric patients.

In the present study no cases of Hepatotropic viruses were seen, including Hepatitis B virus which was common in older times. A possible explanation for this is the increasing awareness of universal precautions and the introduction of vaccination against hepatitis B in the national programme.

In the present study coagulation profile was deranged in 23.53% of cases, this was lower in comparison to other studies due to the protocol at our centre to normalise coagulation profile prior to any procedure to reduce risk of post-operative complications for the patients.

In present study an equal incidence of EHBA and neonatal cholestasis was seen (29.41%) which was also seen in study done by Dhole et al. (2015).⁶ All the above studies showed similar trend with respect to frequency of EHBA and Neonatal hepatitis.

In study by Ahmad et al. (2005)⁵ and study by Monajemzadeh et al. (2009)¹ incidence of secondary hemochromatosis in Thalassemia Major patients was high in comparison to present study where we found no cases for the same. A possible explanation for this is the difference in prevalence of consanguineous marriages in different geographical regions. In present study we found a low incidence of metabolic disorders as compared to study by Hashmi et al. (2017)¹² that found metabolic disorders to

Table 13: Age wise distribution of spectrum of cases

HPE Diagnosis	Frequency	Age wise distribution of HPE			
		Birth-1 month	1 month-1 year	1 year- 5 years	5-16 years
Cholestatic Disease	14	2	12	0	0
EHBA	10	1	9	0	0
Neonatal hepatitis	10	1	9	0	0
Cirrhosis	4	0	4	0	0
Hepatoblastoma	2	0	2	0	0
Glycogen storage disorder	2	0	2	0	0
Portal fibrosis	2	1	0	1	0
Near normal biopsy	2	0	1	0	1
A-v malformation	1	1	0	0	0
Metastasis from neuroendocrine tumor	1	0	0	0	1
Embryonal sarcoma	1	0	0	0	1

Table 14: Age incidence

Studies	Most frequent age range	%
Present study	1 month-1 year	76.47
Sabir et al (2010) ⁴	1 month-1 year	42
Ahmad et al (2004) ⁵	1 month-1 year	34
Monajemzah et al (2009) ¹	1 month-1 year	41

Table 15: Gender incidence

Studies	Male: Female ratio
Present study	1.6:1
Sabir et al (2010) ⁴	1.4:1
Ahmad et al (2004) ⁵	1.2:1
Monajemzah et al(2009) ¹	1.4:1

Table 16: Clinical manifestations

Clinical manifestation	Present study % (n=34)	Dhole et al % (2015) (n=55) ⁶	Hanif et al % (2005) (n=55) ⁷	Ashraf et al % (2019) (n=151) ⁸	Onyiriuka AN et al % (2016) (n=40) ⁹
Icterus	85.29	73	49	26	-
Hepatomegaly	47.06	63	38	40	-
Acholic stools	38.24	-	-	21	-
Variceal bleed	8.82	28	42	-	-
Hypoglycemia	8.82	-	-	-	22.5
Cholelithiasis	5.88	-	-	-	-

Table 17: Laboratory investigations findings

Studies	Deranged LFT %	Deranged coagulation profile %	CMV- Positive %
Present study (n=34)	79.41	23.53	8.82
Hanif et al (2005) (n=55) ⁷	90	90	-
Dhole et al (2015) (n=55) ⁶	73	-	5.4
Ashraf et al (2019) (n=151) ⁸	82	-	2.6
Rajeshwari et al (2008) (n=174) ¹⁰	62.1	46.4	2.29

Table 18: Histological Spectrum of paediatric liver diseases

Diagnosis	Present study	Dhole et al (2015) (n=55) ⁶	Monajemzadeh et al (2009) (n=425) ¹	Ahmad et al (2005) (n=100) ⁵	Sabir et al(2010) (n=450) ⁴
Cholestasis	41.18	23.0	-	-	-
EHBA	29.41	7.27	9.7	20.0	10
Neonatal hepatitis	29.41	7.27	8.7	10.0	20
Cirrhosis	11.76	41.8	6.5	10.0	26
Hepatoblastoma	5.88	-	1.24	1.0	1.3
Metastasis	5.88	-	-	1.0	-
Glycogen storage disorder	5.88	3.6	3.74	12.0	6
Portal fibrosis	5.88	3.6	0.24	1.0	-
A-V malformation	2.94	-	-	-	-
Near Normal	5.88	-	8.7	-	4.4

be the commonest etiology for liver diseases in children of which maximum cases were due to glycogen storage disorder.

Muthupei et al. (2000)¹³ also reported a reduced incidence in neoplastic and metabolic diseases in children which was similar to the present study as well as the other studies that have been tabulated.

4.1. Statically significant histological features

In present study we found certain histological features which were statistically significant ($p < 0.05$) in certain conditions and could stand out as strong indicators for the same.

In cases of EHBA we found that bile duct proliferation, presence of intracanalicular bile plugs, portal tract fibrosis, presence of extramedullary hematopoiesis (EMH) and intracellular bile stasis all were statistically significant in cases of EHBA.

In cases of Neonatal hepatitis(NH) we found that presence of bile plugs, intracellular bile stasis and EMH was common for both EHBA and neonatal hepatitis, however the presence of giant cell transformation was statistically significant in cases of NH an important distinguishing feature between the two.

Thus, liver biopsy can aid in distinguishing between EHBA and neonatal hepatitis quite accurately, which is important for planning further management of the patient EHBA is treated surgically while NH has primarily medical management.

Features such as effaced liver parenchymal architecture, hepatocyte degeneration and necrosis were seen in many of the conditions and weren't statistically significant to any particular pathologies.

5. Conclusion

- 34 histopathological specimens of pediatric patients with underlying liver disease were studied

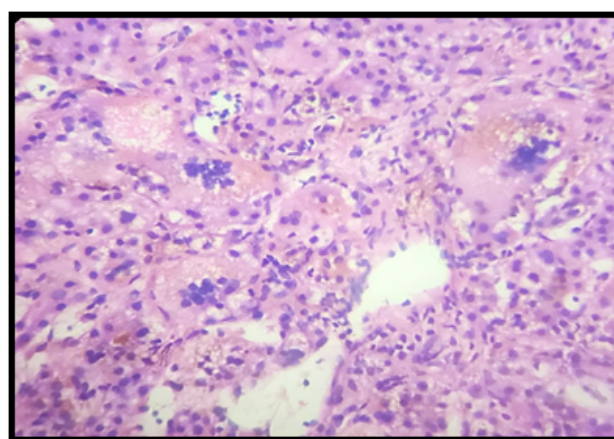


Fig. 1: Neonatal Hepatitis (H &E 400x): Giant cell transformation seen

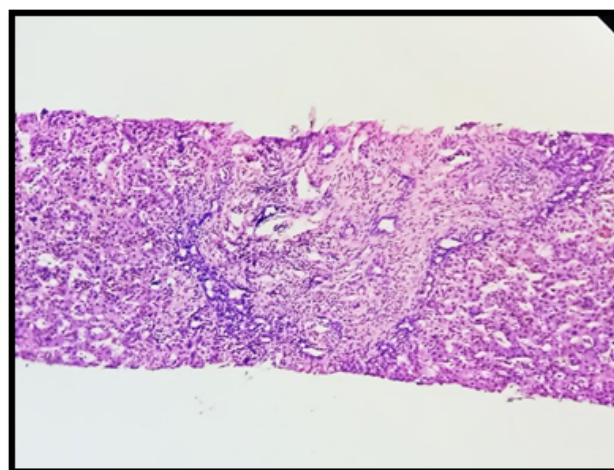


Fig. 2: Interface hepatitis (H&E, 100x)

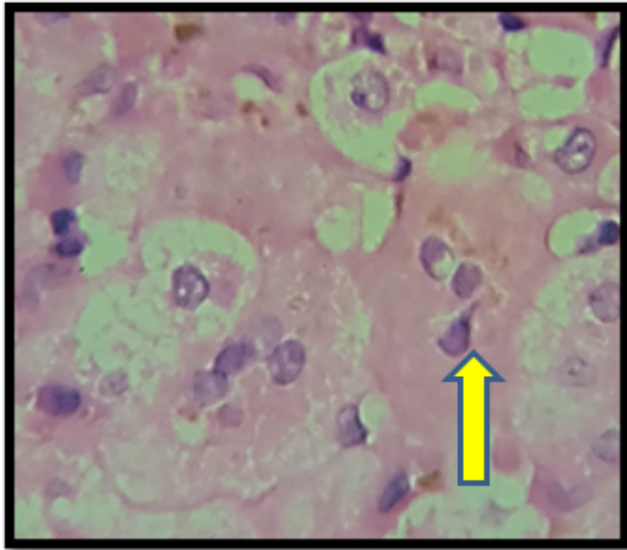


Fig. 3: Degeneration of hepatocytes (H& E 100x and 400x): Hepatocytes showing ballooning and feathery degeneration

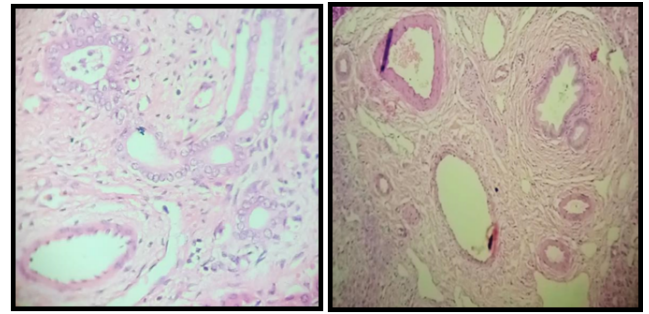


Fig. 6: EHBA with Bile duct proliferation (H& E 100x and 400x)

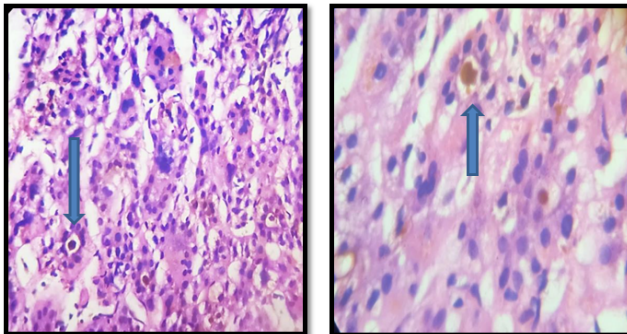


Fig. 4: Bile plugs (Intra canalicular) (H& E 100x and 400x)

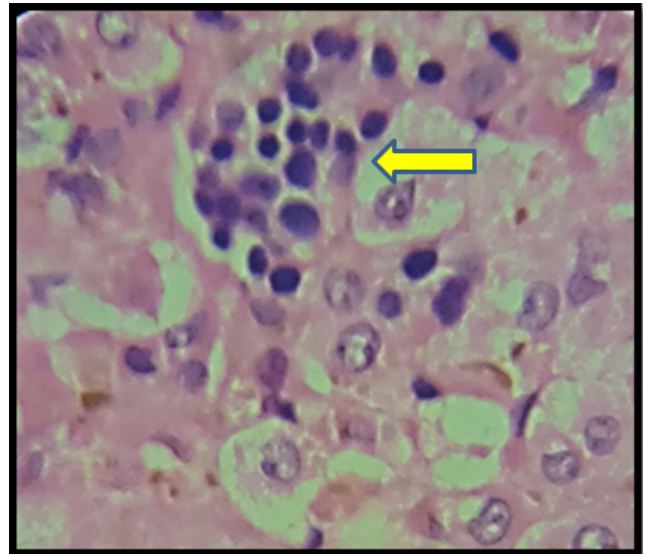


Fig. 7: Extramedullary hematopoiesis (H& E 400x)



Fig. 5: Cholestasis (H&E 400x)

2. Most common age group was between 1 month-1 year (76.47%) in which Cholestasis related to Extra hepatic biliary atresia and Neonatal hepatitis was the commonest etiology, male to female ratio was 1.6:1. Most common clinical presentation was icterus (85.29%), followed by hepatomegaly in 47.06% cases and clay colored stools (38.24%)
3. Deranged Liver function test was seen in 79.41%, Anti CMV Ig G was reactive in 8.82% cases, Coagulation profile was deranged in 08 cases 23.53%.
4. Histopathology and radiology impression correlated in 94.12% cases and good agreement (Kappa coefficient (k)= 0.767) was noted between radiological and histopathological reporting upto the level of classifying the condition as neoplastic or non neoplastic
5. Neonatal cholestasis due to extra hepatic biliary atresia or neonatal hepatitis was seen in 41.18% cases and was the commonest non neoplastic condition, Hepatoblastoma, embryonal sarcoma and metastasis from neuroendocrine carcinomas were neoplastic conditions and glycogen storage disorder was seen in

5.88% cases and was only metabolic disorder in this study

6. Microscopic features such as bile duct proliferation, intracanalicular bile plugs, intracellular bile stasis, portal tract fibrosis, extramedullary hematopoiesis and giant cell transformation of hepatocytes are few important histologic clues that can aid in diagnosing the underlying liver pathology
7. Bile duct proliferation, presence of intracanalicular bile plugs, portal tract fibrosis, presence of extramedullary hematopoiesis (EMH) and intracellular bile stasis all were statistically significant ($p < 0.05$) in cases of EHBA while presence of bile plugs, intracellular bile stasis, EMH and giant cell transformation was statistically significant ($p < 0.05$) in cases of NH and presence of Intracanalicular as well as intracellular bile stasis were statistically significant ($p < 0.05$) with respect to cases showing cholestasis
8. Thus, in children presenting with jaundice, hepatomegaly or other clinical presentations of underlying liver disease, liver biopsy can aid in understanding the underlying pathology in order to reach a definitive diagnosis and thus aid in proper management of patients in liver diseases.

6. Source of Funding

None.

7. Conflict of Interest

The authors declare that there is no conflict of interest.

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