

Hepatic Steatosis and Radiological, Clinical and Biological Correlation in Malnourished Children in Two Referral Hospitals in Cotonou

Florence Alihonou^{1,*}, Patricia Yekpe¹, Francis Honnorat Lalya¹, Mohammed Dakin², Marcelline D'Almeida Hounnou², Olivier Biau²

¹Department of Pediatrics and Medical Genetics, National Teaching Hospital Hubert Koutoukou Maga, Faculty of Health Sciences, Cotonou, Benin

²Radiology and Medical Imaging Department, National Teaching Hospital Hubert Koutoukou Maga, Faculty of Health Sciences, Cotonou, Benin

Email address:

florencealikhonou@yahoo.fr (F. Alihonou)

*Corresponding author

To cite this article:

Florence Alihonou, Patricia Yekpe, Francis Honnorat Lalya, Mohammed Dakin, Marcelline D'Almeida Hounnou, Olivier Biau. Hepatic Steatosis and Radiological, Clinical and Biological Correlation in Malnourished Children in Two Referral Hospitals in Cotonou. *International Journal of Nutrition and Food Sciences*. Vol. 10, No. 6, 2021, pp. 126-133. doi: 10.11648/j.ijnfs.20211006.12

Received: October 27, 2021; **Accepted:** November 16, 2021; **Published:** November 23, 2021

Abstract: Malnutrition in children is associated with signs of liver dysfunction such as hepatic steatosis, which increases morbidity and mortality. This research aims to study the epidemiological, clinical and paraclinical aspects of hepatic steatosis in malnourished children at the CNHU-HKM and CHU-MEL of Cotonou. This was a descriptive and analytical cross-sectional study that took place from May 1 to July 31, 2017. Malnourished children aged 06 months to 05 years old who were hospitalized or received outpatient care during the study period were included. An exhaustive sampling of all children meeting the eligibility criteria during the study period was conducted. Ninety-four malnourished children were included in the study. Their mean age was 18.47 months. The hospital prevalence of hepatic steatosis was 60.6%. Hepatic steatosis was diffuse and homogeneous without focal lesions. Clinically, it was significantly associated with hepatomegaly, underweight and severe chronic malnutrition. A statistically significant association was also observed between, respectively, non-supplementation with iron/folic acid, recent non-adherence to EPI vaccines and hepatic steatosis. Biologically, there was a statistically significant association between hepatic steatosis and transaminases, gamma GT, triglycerides, albuminemia. Hepatic steatosis is common in malnourished children. There is a clinico-biological correlation. Liver ultrasound should be systematic in cases of malnutrition.

Keywords: Hepatic Steatosis, Malnutrition, Associated Factors, Benin

1. Introduction

Malnutrition is one of the crucial problems facing humanity. It is defined by the World Health Organization (WHO) as a medical condition resulting from a relative or absolute deficiency or excess of one or more essential nutrients [1]. This definition makes it possible to distinguish two aspects of malnutrition: the first linked to a deficiency and the second linked to an excess. According to the WHO, malnutrition in all its forms affects one in three people and children are a particularly vulnerable group [1]. Malnutrition is responsible for 45% of deaths among children under five

years of age in the world [1]. In its severe form, malnutrition can be associated with signs of liver dysfunction such as hepatic steatosis and hypoalbuminemia as well as metabolic disorders with increased oxidative stress. However, the pathophysiology of these various disorders, particularly hepatic steatosis, is still under discussion.

Non-alcoholic fatty liver disease (NAFLD) is a multifactorial disorder ranging from simple steatosis to non-alcoholic steatohepatitis (NASH), with or without fibrosis [2]. Over the past two decades, the high prevalence of overweight and obesity probably explains its emergence as the leading cause of liver disease in the pediatric

population worldwide. Its prevalence varies from 20-30% in developed countries due to the obesity pandemic and is estimated at 10% in children in general and reaches 40-70% in obese children [3]. The simple steatosis encountered in NAFLD is not associated in the short term with a dramatic increase in morbidity or mortality. However, its progression to NASH significantly increases the risk of cirrhosis, liver failure and hepatocellular carcinoma (HCC) [3]. Most studies on liver steatosis in malnourished children have focused on obese children, for whom its management is well codified and includes weight loss, antioxidants, oral antidiabetic drugs, vitamin therapy, and bariatric surgery [3]. Nevertheless, some studies [4-8] have demonstrated the existence of this steatosis in malnourished children due to deficiency. However, its pathophysiological mechanism is not well known [4, 9]. A deeper knowledge of the mechanisms of occurrence of this hepatic steatosis is needed so that its management can be codified. This management could be combined with that of malnutrition to improve the overall outcome of malnutrition treatment. The latter is still unsatisfactory, despite major advances in treatment, with a lethality rate of more than 30% in hospital [9].

In our African context and particularly in Benin, scientific data that could help draw the attention of health professionals to the risk of a malnourished child to have liver steatosis and its outcome are non-existent. It is therefore to help overcome this information deficit that we propose to study the ultrasound aspects of hepatic steatosis in malnourished children and to establish a radiological, clinical and biological correlation in order to have a baseline in Beninese malnourished children. Through this study, the echo structure of the liver will be used by the nursing staff as a monitoring indicator of malnutrition in order to detect steatosis at an early stage and manage it to avoid its evolution towards steatohepatitis, cirrhosis or even hepatocellular carcinoma.

2. Methods

2.1. Study Framework

This was a cross-sectional study with an analytical aim that took place from May 1st to July 31st, 2017 at the Centre National Hospitalier Universitaire Hubert Koutoukou Maga (CNHU-HKM) and the Centre Hospitalier Universitaire de la Mère et de l'Enfant Lagune (CHU-MEL). At the CNHU-HKM, it took place in the Department of Pediatrics and Medical Genetics, in the Department of Medical Imaging and the Department of Clinical Biochemistry. At the CHU-MEL, it took place in the General Pediatrics Department.

2.2. Study Population

It was represented by malnourished children who were hospitalized or receiving outpatient care. Included were all children aged 6 to 59 months who came to the CNHU-HKM or CHU-MEL during the study period in whom anthropometric parameters were measured and who were

diagnosed as malnourished. Children whose parents did not give their consent for a liver ultrasound and a blood sample for the biological assessment were not included. Malnourished children who missed one of the tests performed were excluded. Children with congenital liver malformations or with known acute or chronic liver pathology and those who have been put on drugs that can cause liver steatosis (corticosteroids, tetracyclines, valproic acid, methotrexate, antiretrovirals, ethanol, etc.) have also been excluded.

2.3. Sampling

An exhaustive sampling of all children meeting the eligibility criteria during the study period was conducted.

2.4. Variables

The dependent variable was the presence or absence of hepatic steatosis. The latter was determined by the ultrasound aspect of the liver during a liver ultrasound. The independent variables were the variables related to general characteristics (age, sex); the variables related to medical and nutritional history (type of breastfeeding, age of diversification, mode of diversification, age of ab lactation, up-to-date EPI vaccinations, up-to-date non-EPI vaccinations, iron-folic acid supplementation, recent deworming); variables related to physical examination (mucosal staining, presence of bilateral edema, dyscratic signs, hair dystrophy, behavioral disorders, hepatomegaly); variables related to anthropometric parameters (weight, height, Z-scores, nutritional status); the presence and abundance of ascites and variables related to the biological profile (ALAT, ASAT, total cholesterol, HDL, LDL, triglycerides, uricemia, Gama GT, protidemia, albuminemia and HIV status).

2.5. Data Collection

2.5.1. General Characteristics, Medical and Nutritional History and Physical Examination

After the children were identified, their health and/or hospitalization records were consulted for general information, vaccinations and vitamin A supplementation. Next, the child's weight, height, and age were recorded. The age was determined from the date of birth (day/month/year) recorded in the child's birth record. For weight gain, children were fully clothed. The height of the children was measured to the nearest millimeter using a height gauge. The next step was to determine the weight/height; height/age; weight/age ratios. The anthropometric parameters were accurately determined using the World Health Organization (WHO) Anthro software version 3.2. Subsequently, an interview was conducted with the children's mothers regarding feeding behavior, supplementation and deworming. Thus, we searched for:

- 1) the type of breastfeeding (exclusive, artificial or mixed breastfeeding) and the age of weaning;
- 2) the method of dietary diversification (considered appropriate when it has been progressive, as varied as possible, neither too salty nor too sweet, respecting the

- child's tastes and made of quality foods);
- 3) the age of the ablation,
 - 4) iron/folic acid supplementation (we considered a child to have received good iron/folic acid supplementation when it was given at six months of age for two months and every six months until the age of two years;
 - 5) deworming (it was considered recent when it was less than three months old).

Once this information was collected, a physical examination was done in which we assessed the coloring of the mucous membranes and looked for edema of the lower limbs, the presence of hepatomegaly, disomic signs, the presence of hair dystrophy, behavioral disorders and other clinical signs.

2.5.2. Biological Assessment

The children were given a rapid diagnostic test for HIV infection as soon as the blood sample was taken. The samples were collected in tubes containing a separator and immediately sent to the biochemistry laboratory, where they were centrifuged for 10 minutes at 3,500 rpm. The sera were then collected in aliquots. The samples were stored in a cool place and were analyzed as a block at the end of recruitment. Each child was tested for the following: alanine aminotransferase (ALAT), aspartate aminotransferase (ASAT), uricemia, Gamma glutamyl transpeptidase (Gamma GT), lipid profile (total cholesterol, HDL, LDL, triglycerides), protidemia, albuminemia. These tests were performed on a BS200-MINDRAY multiparametric automaton.

2.5.3. Ultrasound Examination

Liver ultrasounds were performed in all children by the same experienced radiologist on the same ultrasound scanner and under the same settings to avoid inter and intra-operative variability. This was a Hitachi Eub 500 type ultrasound scanner, commissioned in 2008, using a sectorial probe operating between 3 and 5 MHz in B and Doppler mode and then a linear probe operating at 7.5 Mhz.

Children aged between 06 and 24 months were kept on their mother's knee in a half-seated position, while those over two years old were laid supine in bed with their pelvic limbs fully extended and their heads resting on a pillow. Then the abdominal area was then stripped and gel-coated in all the children. We first measured the size of the right lobe on a sagittal section of the liver passing through the right medico-clavicular line (MCL) in the left lateral decubitus position, and in deep inspiration. The distance between the two edges (upper and lower) was then measured and constituted the hepatic arrow. This measurement was then compared with the size of the liver for the child's age to infer whether the child had hepatomegaly. Table 1 shows this change in liver size with age [10]. We measured the size of the left lobe on a sagittal section passing through the aorta in the supine position. The contours of the liver were explored and dysmorphism was investigated as well as liver parenchyma on a sagittal section of the liver passing through the right kidney. The echo structure of the liver was then assessed as well as the existence of hepatic steatosis through the

hepato-renal gradient. In mild steatosis, there is a slight diffuse increase in the echo structure of the hepatic parenchyma with normal visualization of the diaphragm and intrahepatic vessel wall. Moderate steatosis is represented by a moderate and diffuse increase in the echo structure of the hepatic parenchyma with a slight decrease in the visualization of the intrahepatic vessel wall and diaphragm. Severe steatosis is characterized by a marked increase in liver echo structure with poor or absent visualization of the intrahepatic vessel walls, diaphragm, and posterior portion of the right lobe of the liver, and no visualization of the liver capsule [11]. To limit bias, we have classified steatosis into two groups: moderate steatosis and severe steatosis. In addition, we have searched for focal lesions, studied the portal trunk caliber on recurrent subcostal cuts and verified their Doppler permeability, studied the caliber and permeability of the suprahepatic veins on recurrent, oblique and subcostal cuts, and searched for ascites (effusion in Morrison's space, in Douglas' cul-de-sac or in the parietocolic gutters).

Table 1. Liver's size changes on a CML slice in 0 to 5 years children.

| Age (month) | 0 | 1 | 4 | 11 | 19 | 30 | 42 | 60 |
|-------------|----|----|----|----|----|----|----|----|
| Size (mm) | 57 | 61 | 65 | 71 | 75 | 79 | 83 | 85 |

2.6. Data Processing and Analysis

The data were entered into Excel 2010. Once the data were cleared, the analysis of the data was performed using SPSS 21 software. Quantitative variables were expressed as a mean of plus or minus standard deviations and qualitative variables as a percentage. Comparisons of proportions by the Chi² test and analyses of variance by the Fisher F-test and Welch t-test were used to look for significant associations between the dependent variable and each of the independent variables. The significance level was 5%.

2.7. Ethical Aspects

Informed consent was obtained from the parents of the children included in this study. To this end, a pre-established consent form was signed by the parents. Similarly, the authorization of the various heads of pediatrics, medical imaging and biochemistry departments of the targeted centers as well as that of the hospital directors was obtained. The results of the examinations were explained to the parents and in some cases, the children were presented to the pediatricians for care and follow-up.

3. Results

3.1. General Characteristics of Malnourished Children

A total of 52 patients had been recruited at CNHU-HKM and 42 at CHU-MEL. The age group most affected by malnutrition was 6-12 months. The mean age of the children retained was 18.47 ± 14.23 months with extremes of 6 and 59 months. The sex ratio of boys to girls was 0.95.

3.2. Medical and Nutritional History of Malnourished Children

Breastfeeding was the most common method of breastfeeding (67%). Among weaned children, more than half (56.4%) were weaned before the age of six months and the weaning method was most often unsuitable (64.9%). Also, ab lactation was done in more than half the cases (51%) before the children's second birthday. Children had received vitamin A supplementation in 70.2% of cases, while only 43% had received iron/folic acid supplementation. Three out of four children (74.4%) had up-to-date immunization status according to the Expanded Programme on Immunization (EPI). But 9 out of 10 children had not yet initiated non-EPI vaccines.

3.3. Anthropometric Parameters

Severe underweight was the most common type of malnutrition with two out of three children, while moderate chronic malnutrition was the least common. It affected one child in four.

3.4. Biological Profile

Approximately 14% of the children were HIV-positive. Table 2 presents the biological parameters of the malnourished children in the study.

Table 2. Biological parameters of malnourished children.

| Biological parameters | Mean | Standard deviation | Min | Max |
|-----------------------|--------|--------------------|-------|--------|
| ALAT | 89.76 | 77.83 | 18.00 | 679.00 |
| ASAT | 102.41 | 92.85 | 20.00 | 787.00 |
| Total Cholesterol | 1.43 | 0.56 | 0.53 | 4.19 |
| HDL Cholesterol | 0.36 | 0.16 | 0.10 | 0.73 |
| Triglyceride | 1.81 | 1.45 | 0.47 | 7.02 |
| LDL Cholesterol | 0.71 | 0.45 | 0.04 | 2.88 |
| Gamma GT | 97.74 | 140.57 | 7.00 | 724.00 |
| Uric acid | 54.50 | 34.20 | 20.00 | 212.00 |
| Protidemia | 66.38 | 13.61 | 28.00 | 110.00 |
| Albuminemia | 37.83 | 9.88 | 18.00 | 74.00 |

3.5. Liver Biometrics and Prevalence of Hepatic Steatosis in Malnourished Children

The prevalence of hepatic steatosis in malnourished children was 60.6%. It was severe in 12.3% of cases. About 3 out of 4 malnourished children had hepatomegaly. Moreover, there was no dysmorphic disorder, the liver contours were regular. The portal trunk and the suprahepatic veins were of normal size, permeable without dilation of the bile ducts. Steatosis was diffuse in all the children in our series without associated focal lesions. We had two cases of ascites of great abundance. The liver biometrics are summarized in the table 3.

Table 3. Liver biometrics of malnourished children.

| | Mean | Standard deviation | Min | Max |
|--------------|-------|--------------------|-------|--------|
| Aorta | 63.50 | 13.04 | 10.00 | 99.00 |
| MCL | 86.38 | 16.00 | 57.00 | 133.00 |
| Portal trunk | 5.77 | 1.27 | 3.00 | 12.00 |
| Liver veins | 3.06 | 0.87 | 1.60 | 6.30 |

3.6. Factors Associated with Hepatic Steatosis

Tables 4, 5 and 6 show the results of statistical tests between independent variables and hepatic steatosis. A statistically significant association was observed between hepatic steatosis and hepatomegaly. A statistically significant association was also observed between respectively non-supplementation of iron/folic acid, recent non-adherence to EPI vaccines and hepatic steatosis. Moderate or severe underweight and severe chronic malnutrition were also associated with hepatic steatosis. The mean ALAT value was statistically different for malnourished children with (or without) fatty liver. The same was true for mean values for AST, Gamma GT, triglycerides and albuminemia. A statistically significant association was found between hepatic steatosis and elevated triglycerides.

Table 4. Association between the history of malnourished children, nutritional status, health status and liver steatosis.

| Variables | Hepatic steatosis | | Chi ² p-value |
|---------------------------------|-------------------|------------|-----------------------------|
| | Yes | No | |
| Sex | | | |
| Male | 26 (56.5%) | 20 (43.5%) | 0.42 |
| Female | 31 (64.6%) | 17 (37.4%) | |
| Breastfeeding | | | |
| Exclusive | 42 (66.7%) | 21 (33.3%) | 0.098 |
| Artificial | 06 (66.7%) | 03 (33.3%) | |
| Mixed | 09 (40.9%) | 13 (59.1%) | |
| Age of diversification | | | |
| Less than 6 months | 31 (58.6%) | 22 (41.5%) | 0.39 |
| More than 6 months | 26 (63.4%) | 15 (36.6%) | |
| Diversification mode | | | |
| Adapted | 17 (51.5%) | 16 (48.5%) | 0.18 |
| Not adapted | 40 (65.6%) | 21 (34.4%) | |
| Ab lactation | | | |
| No | 33 (67.3%) | 16 (32.7%) | 0.11 |
| Yes | 24 (53.3%) | 21 (46.7%) | |
| Age of the ab lactation | | | |
| < 6 months | 05 (83.3%) | 01 (16.7%) | 0.4 |
| 6-12 months | 14 (73.7%) | 05 (26.3%) | |
| 13-24 months | 14 (60.9%) | 09 (39.1%) | |
| >24 months | 00 (0.00%) | 01 (100%) | |
| Not classified | 25 (55.6%) | 20 (44.4%) | |
| Iron/Folic Acid Supplementation | | | |
| Yes | 19 (46.3%) | 22 (53.7%) | 0.010 |
| No | 38 (71.7%) | 15 (28.3%) | |
| Supplementation in Vitamin A | | | |
| Yes | 36 (54.5%) | 30 (45.5%) | 0.060 |
| No | 21 (75%) | 7 (25%) | |
| Recent de-worming | | | |
| Yes | 21 (48.8%) | 22 (51.2%) | 0.026 |
| No | 36 (70.6%) | 15 (29.4%) | |
| EPI vaccination | | | |
| Up to date | 40 (58%) | 29 (42%) | 0.26 |
| Not Up to Date | 17 (68%) | 08 (32%) | |
| Non-EPI vaccination | | | |
| Up to date | 2 (22.2%) | 7 (77.8%) | 0.018 |
| Not up to date | 55 (64.7%) | 30 (35.3%) | |

Table 5. Association between hepatomegaly, nutritional status of malnourished children and hepatic steatosis.

| Variables | Hepatic steatosis | | Chi ² p-value |
|-------------------------------|-------------------|------------|-----------------------------|
| | Yes | No | |
| Hepatomegaly | | | |
| Yes | 24 (92.3%) | 2 (7.7%) | 0.000 |
| No | 33 (48.5%) | 35 (51.5%) | |
| Type of malnutrition | | | |
| No oedema | 51 (61.4%) | 32 (38.6%) | 0.74 |
| With oedema | 06 (54.5%) | 05 (45.5%) | |
| Moderate wasting | | | |
| Yes | 15 (53.6%) | 13 (46.4%) | 0.36 |
| No | 42 (63.6%) | 24 (36.4%) | |
| Severe wasting | | | |
| Yes | 31 (70.5%) | 13 (29.5%) | 0.068 |
| No | 26 (52%) | 24 (48%) | |
| Moderate chronic malnutrition | | | |
| Yes | 14 (60.9%) | 9 (39.1%) | 0.97 |
| No | 43 (60.6%) | 28 (39.4%) | |
| Severe chronic malnutrition | | | |
| Yes | 33 (70.2%) | 14 (29.8%) | 0.045 |
| No | 24 (51.1%) | 23 (48.9%) | |
| Moderate underweight | | | |
| Yes | 10 (35.7%) | 18 (64.3%) | 0.001 |
| No | 47 (71.2%) | 19 (28.8%) | |
| Severe underweight | | | |
| Yes | 44 (71.0%) | 18 (29.0%) | 0.004 |
| No | 13 (40.6%) | 19 (59.4%) | |

Table 6. Association between biological parameters of malnourished children and hepatic steatosis.

| Biological parameters | Hepatic steatosis | | p-value | |
|-----------------------|-------------------|-------|-----------|----------|
| | Yes | No | | |
| | Mean | Mean | F. Fisher | T. Welch |
| ALT | 107.84 | 61.89 | 0.005 | 0.001 |
| AST | 123.14 | 70.49 | 0.007 | 0.001 |
| Total Cholesterol | 1.43 | 1.42 | 0.925 | 0.918 |
| HDL Cholesterol | 0.34 | 0.39 | 0.173 | 0.182 |
| Triglycerides | 2.00 | 1.52 | 0.117 | 0.092 |
| LDL Cholesterol | 0.69 | 0.73 | 0.725 | 0.705 |
| Gamma GT | 141.86 | 28.78 | 0.000 | 0.000 |
| Uric acid | 56.40 | 51.57 | 0.506 | 0.499 |
| Protidemia | 64.19 | 69.76 | 0.052 | 0.42 |
| Albuminemia | 34.77 | 42.54 | 0.000 | 0.000 |

4. Discussion

4.1. Prevalence of Hepatic Steatosis in Malnourished Children

The prevalence of steatosis was 60.6%. This value is significantly lower than that found by Doherty *et al* [5] in Jamaica (94%), Lalwani *et al.* [8] in India (92%). These two authors evaluated liver steatosis on ultrasound upon admission of malnourished children to the hospital. This high frequency could be explained by two reasons. In our study, we used a 3.5 MHz probe whereas these authors used respectively 7.5 MHz and 5 MHz probes which could give them better visibility of steatosis. Besides, their sample consisted only of severely malnourished children while we included both moderate and severe malnourished children. However, our frequency is comparable to that observed in obese children, where the prevalence of hepatic steatosis

varies between 40 and 70% [3]. Over the last two decades, the increase in overweight and obesity has led to the emergence of fatty liver as the major liver disease in the global pediatric population [2, 3, 12, 13].

Steatosis was severe in 12.3% of children with hepatic steatosis and moderate in the remainder. These results are comparable to those of Lalwani *et al.* who found 14% severe steatosis and 42% moderate steatosis in India [8]. In contrast, Doherty *et al.* found 35% severe steatosis and 24% moderate steatosis in severely malnourished children in Jamaica [5]. Shannon *et al.* in a study of 208 children with histologically proven steatosis, found moderate steatosis in 42% and 37% and severe steatosis in 27% and 22% on biopsy and ultrasound, respectively [14]. These variations in the results observed in the literature are thought to be related to three factors. The first factor would be the method used (type of device and its performance, device settings). The second factor would be related to the competence of the operator. The latter explains the inter and intra-operator variations. The third factor would be related to the echogenicity of the patients which was variable according to the type of malnutrition concerned by the study.

All the children in our series had diffuse, homogeneous steatosis. The liver was evenly contoured without dysmorphism or focal lesions. The portal trunk and the suprahepatic veins were of normal size, permeable without dilation of the bile ducts. In the literature, diffuse hepatic steatosis is the most frequently encountered form [15].

We have not had any cases of cirrhosis in our population. It has been described that hepatic steatosis in obese children, although benign, can progress to cirrhosis and hepatocellular carcinoma when the disease progresses to steatohepatitis [2, 3, 12-14, 17]. Since our sample is composed of children under 5 years of age, the absence of cirrhosis may be related to the short time to progression of hepatic steatosis. Nevertheless, the elevated liver enzymes presented by the children could be strongly suggestive of steatohepatitis.

The main ultrasound abnormality found was hepatomegaly in 73.40% of cases. In children with hepatic steatosis, hepatomegaly was found in 92.3% of cases. This proportion greatly exceeds that found by Hourigan *et al.* who found hepatomegaly in 48% of cases in children with histologically proven hepatic steatosis [18]. This difference in proportion can be explained by the fact that our study was carried out in a malaria-endemic zone, which could be a secondary factor associated with hepatomegaly. The study by Hourigan *et al.* was conducted in the United States. Also, the thresholds used to define hepatomegaly could be different.

4.2. Factors Associated with Hepatic Steatosis

A statistically significant association was observed between iron/folic acid non-supplementation and hepatic steatosis. The liver is an important site for iron and lipid metabolism and the main site for interactions between the two pathways. Iron is an integral part of certain enzymes and transporters involved in lipid metabolism; as such, it can exert a direct effect on hepatic lipid load, intrahepatic metabolic pathways and hepatic lipid secretion [19].

Sherman et al. [20, 21] have shown that iron deficiency in rats is associated with increased lipogenesis, leading to the accumulation of liver triglycerides and steatosis. Since metabolic absorption pathways are downwardly regulated, an increase in lipogenesis could be due to a decrease in fatty acid utilization. Bartholmey et al. [22] have shown that in iron-deficient rats, there is a decrease in the oxidation of fatty acids, which are then diverted to the formation of triglycerides that cause hepatic steatosis. β -oxidation of fatty acids that are then diverted to the formation of triglycerides that cause hepatic steatosis. Supporting this hypothesis, other authors such as Hulse et al. [23], have found a lower level of carnitine in iron-deficient rats, since she uses it as a co-factor. However, others argue that β -oxidation remains unchanged while de novo lipogenesis increases [20, 21]. This increase in lipogenesis is supported by genomic studies that show a simultaneous decrease in the expression of genes involved in β -oxidation and an increase in those involved in lipogenesis [24].

In our study, recent non-de-worming and failure to complete non-EPV vaccines were associated with hepatic steatosis. However, this could be purely theoretical because the link is difficult to establish from a pathophysiological point of view. Moreover, our observations show that hepatic steatosis is a chronic condition. Thus, this association would be paradoxical, especially concerning the recent non-de-worming. Moreover, the latter could be linked to an information bias, given that the deworming was not always marked in the children's care record and we sometimes relied on the parents' memory. Nevertheless, we could try to explain these associations by the fact that the non-de-worming and the non-adherence of non-EPI vaccines expose the child to certain infectious diseases. These could be manifested by diarrhea-vomiting, which degrades the child's nutritional status and exposes him/her to hepatic steatosis.

There was a statistically significant association between hepatic steatosis and severe chronic malnutrition as well as moderate and severe underweight. These statistical links had not been sought in the literature and show that hepatic steatosis is not an acute settling phenomenon.

We found a statistically significant association between hepatic steatosis and alanine aminotransferase and aspartate aminotransferase. Thus, hepatic steatosis in our series was associated with elevated transaminases. This association was highlighted in the literature by Oh et al. who identified non-alcoholic and alcoholic liver steatosis as the most frequent causes of transaminase elevation [4]. van Zutphen et al. [9] in the Netherlands in 2016 in an experimental study observed that by subjecting malnourished rats to a diet similar to that seen in malnourished people in developing countries where staple foods such as maize and cassava are rich in carbohydrates and low in protein (about 5% protein), the latter then developed liver steatosis after four weeks with an increase in alanine aminotransferase compared to controls on a normoprotein diet. It is deduced that hepatic steatosis is accompanied by hepatic cytolysis. This is therefore the concept of steatohepatitis, the definition of which is however histological. In 2015, Cruz et al. in Brazil found a statistically

significant association between the importance of hepatic steatosis and an increase in ALT ($p=0.001$) and AST ($p=0.0001$) in adults (non-alcoholic, without liver pathology) who had presented hepatic steatosis on ultrasound [25]. Our study shows that transaminases can be used to suspect hepatic steatosis in malnourished children or specially to predict its severity (steatohepatitis). However, Lalwani et al. found no correlation between the importance of steatosis and an increase in transaminases [8]. Hourigan et al. found a significant association between hepatic steatosis and AST but not ALT [18].

There was also a statistically significant association between Gamma GT and hepatic steatosis. The same observation was made by Cruz et al. [25] and Preidis et al. [26] in Texas. The latter worked on experimental models of malnourished rats in which he found an increase in Gamma glutamyl conjugation peptides reflecting an increase in Gamma GT that would be linked to significant oxidative stress, inflammation and hepatocyte damage related to steatosis [26]. However, Hourigan et al. found no link between hepatic steatosis and increased Gamma GT [18].

We also found a statistically significant association between hepatic steatosis and albuminemia; this is not the case with protidemia. Given that steatosis has been studied in malnourished children, it would have been surprising if we did not find an association between it and albuminemia, which is a biological indicator of nutritional status. An inverse correlation between the importance of hepatic steatosis and albuminemia has been documented in the literature [27, 28]. The lack of association between protidemia and hepatic steatosis could be justified by the fact that protidemia does not reflect the nutritional status that is correlated with hepatic steatosis. Indeed, the children in our series presented sepsis in 31% of cases; this could lead to an increase in alpha-globulin synthesis with erroneous normalization of protidemia despite their hypoalbuminemia.

This hypoalbuminemia in malnourished children would be the cause of the two cases of ascites that we found in our sample.

About lipid profile, only the increase in triglycerides was found to correlate with hepatic steatosis. Opinions differ in lipid assessment in malnourished children with liver steatosis. Indeed, four main hypotheses have been put forward to explain hepatic steatosis in malnourished children by default: either an increase in fatty acid metabolism from adipose tissue, or an increase in fatty acid synthesis in the liver (de novo lipogenesis), or a decrease in fatty acid oxidation, or a decrease in their secretion in the form of lipoproteins [24, 25]. Considering these hypotheses, particularly the decrease in the secretion of triglycerides in the form of lipoproteins, there should be a decrease in plasma triglycerides in children with hepatic steatosis. This is the case, for example, in Zutphen et al. [9] in 2016 in models of malnourished rats. Truswell et al. [27, 28] showed that triglycerides initially correlated with the importance of steatosis in a reverse manner increased with renutrition. However, other authors [4, 6, 29] find contradictory results by evaluating hepatic steatosis on an

uninjected abdominal CT scan in severely malnourished children and by directly measuring apolipoprotein-B-100 (which is a major constituent of VLDL and LDL, the forms in which triglycerides are secreted). They have shown that this apolipoprotein increases with the amount of hepatic steatosis (i.e. a major flow of triglycerides from the liver to the plasma); this results in an increase in triglycerides in children with steatosis regardless of the presence or absence of edema. And this is what we observed in our study.

5. Conclusion

This study determined the frequency and factors associated with hepatic steatosis. It was found that hepatic steatosis is common in malnourished children and that there is a radiological, clinical and biological correlation. The results of this study suggest that liver echo structure should be used by health care personnel as a monitoring indicator of malnutrition to detect steatosis early and to manage it to avoid its evolution towards steatohepatitis, cirrhosis, or even hepatocellular carcinoma. Furthermore, this study could serve as a basis for further research. From a biological point of view, further explorations such as prealbuminemia, martial assessment, and the determination of serum markers of inflammation could be of great interest. Also, the generalization of our work is necessary to determine the prevalence of hepatic steatosis in malnourished children in Benin. It will also make it possible to determine the impact of its management on the vital prognosis of severely malnourished children during the stabilization phase. All this will allow a better understanding of malnutrition to improve its management and reverse the disastrous trends that this pathology is experiencing in children in developing countries.

Acknowledgements

The authors would like to thank the mothers of the children for their participation and the health authorities and hospital officials involved in the study for their collaboration.

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