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Physiochemical Effect of Neonatal Jaundice on Some Liver Enzymes and Cholecystokin in Hormone.

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Abstract

This study was conducted with the aim of determining the relationship between some physiological and biochemical variables and the incidence of neonatal jaundice in newborns and comparing them with healthy group, the study include 120 blood samples were collected for children attending Fallujah Hospital for Women and Children for the period from October / 2020 to March / 2021, and it was divided into two groups of unequal number, namely the control group G1 and included 30 blood samples for healthy children and the second included (90) blood samples For children with congenital jaundice, the group of patients was divided into three groups according to the concentration of total bilirubin in the blood, each group included 30 samples, and they are the second group G2 with a concentration (10.6 - 13.9) mg / dl, and the third group G3 (14.0 – 17.6) mg / dl and the fourth group G4 (17,7-31,0) mg / dl, the result of this study recorded a significant decrease in the number of white blood cells in the three groups compared with the G1 control group. The results also showed that there was a significant increase in the activity of the ALT enzyme in groups of newborns patients with congenital jaundice compared to the control group. While there was a significant decrease in the activity of the AST enzyme with the same groups above. The results also showed that there was a significant increase in the activity of the ALP enzyme in the three groups compared to the control group. While the results showed that the G3 group with a concentration of (14-17.6) mg / dl showed a significant increase compared with the control group G1 and other affected groups. The results also showed that there was a significant increase in the effectiveness of CCK hormone in the three groups compared to the control group. **We conclude** from this study that there was a significant increase between ALT, ALP and CCK with jaundice, while there was a significant decrease between AST and WBC with jaundice.

Keywords: Neonatal jaundice,Cholecystokinin, Gamma-Glutamyl transferase (GGT).

1. Introduction

The World Health Organization stated that four million newborns who are born after or before the end of pregnancy are exposed to life-threatening ailments and health problems (**World Health Organization, 2014**). Congenital jaundice is the most common case and affecting more than 50% of newborns in the first week after birth**(Knupfer et al., 2005; Bhutani et al., 2013)**. Itssyndrome clear by yellowish discoloration of the skin and is caused by the accumulation of bilirubin pigment that is unconjugated, non-polar and soluble in skin fatand requires medical care in newborns**(Kliegman et al., 2016)**.Minor intake of calories or dehydration from poor breastfeeding, subsequent weight loss, delayed passage of stools, and bruising at birth make the infant more susceptible to jaundice **(Seidman et al. 2008; Newman et al. 2005).**It is considered a benign postpartum transitional phenomenon that demonstrates the dynamic balance between the process of bilirubin formation and elimination. As the process of increasing bilirubin production and impairment of its elimination puts the infant at great risk due to the development of hyperbilirubinemia with subsequent jaundice. **(Stevenson et al., 2002).**Bilirubin is a well-known antioxidant when it is in low concentrations, but it becomes effective neurotoxicity when its concentration increases. It may cause permanent damage in the brain due to the toxic effect on brain cells when it reaches a higher level, and that delaying treatment leads to behavioral and

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neurological weakness that triggers neurotoxicity or kernicterus, which leads to permanent encephalopathy**.(Boulanger et al., 2013; Wisnowski et al.2016).**The syndrome of neurological dysfunction induced by bilirubin represents a group of neurological manifestations in impaired newborns following exposure to high levels of bilirubin. So early detection of this condition can protect them from long-term and irreversible brain damage.**(Johnson and Bhutani, 2011).**There are a number of factors that may play an important role in the effect of hyperbilirubinemia in newborns such as physical factors such as birth weight, gestational age as well as the mother's infection with infectious diseases during pregnancy **(Mesic et al., 2014)**.The production and assimilation of bilirubin is the final product of the degradation of heme. About 80% of bilirubin arises from the degradation of hemoglobin of red blood corpuscles in the reticuloendothelial system, and the remaining 20% comes from inactive erythrocytes formed in the bone marrow and the dissolution of other proteins **(Cheifetz and Adam, 2010)**.Haemoxygenase. The production of biliverdin is the first major step in the pathway This process begins with the production of biliverdin in hem by the enzyme, then followed by the second step, where the enzyme biliverdinreductase works by converting biliverdin to produce bilirubin in an unconjugated form, via the enzyme glucuronyltransferase, which bilirubin becomes conjugated in the liver, making it soluble in water. Most of which goes into the gallbladder and then exits into the small intestine. Although most of the bile acid is reabsorbed at the end of the ileum to participate in the enterohepatic circulation, the conjugated bilirubin is not absorbed but rather goes into the colon. The coliform bacteria break down and process it or metabolize bilirubin into colorless Urobilinogen, which can be oxidized to form urobilin and stercobilin. Where urobilin is excreted through the kidneys with urine, which gives it the distinctive yellow color, while stercobilin is excreted with the stool, where it gives the stool its distinctive brown color. A very small amount (˜1%) of Urobilinogen is reabsorbed into the gastro-hepatic circulation to be excreted in the bile (**Sticova and Jirsa, 2013; Mustafa et al.,2020**).

Materials and working methods:

Study design:

The study targeted newborns lying in the halls of premature babies with hyperbilirubinemia and diagnosed by specialized pediatricians, and the study included 120 blood samples for newborns and the experiment was conducted during the period between October / 2020 until March / 2021 and the samples were divided into four groups each group contains 30 blood samples for newborn babies. Depending on the intensity of the Total Serum Bilirubin concentrationas shown:

1. Group I: Control Group G1, all of them healthy (20 males and 10 females) with total bilirubin percentage less than (8.0) mg/dl.
2. The second group: G2, include (14 males and 16 females), and the percentage of total serum bilirubin ranges between (10.6 - 13.9) mg/dl.
3. The third group: G3 include (18 males and 12 females), and the percentage of total serum bilirubin ranges between (14.0-17.6) mg/dl.
4. The fourth group: G4, include (22 males and 8 females), and the percentage of total serum bilirubin ranges between (17.7-31.0) mg/dl.

Samples: of bloodCollection

Blood samples were collected from newborns attending the Fallujah Hospital for Women and Children in Fallujah District, Anbar province, Iraq. sleeper in the premature unit diagnosed by specialized pediatricians and other children from Fallujah without hyperbilirubinemia (Control group) through medical syringes with a capacity of 5 ml. and 1 ml of blood was placed in An EDTA tube (Ethylene Damien Tetra Acetic acid) after mix well until the anticoagulant is mixed with the blood for the purpose of calculating the total number of white blood cells (WBCs).

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As for the remaining 4 ml of blood, it was placed in a gel tube free of anti-coagulants for the purpose of separating the serum through a centrifuge and kept at a temperature of -20 ° C until the tests were carried out that included estimating the activity of the enzyme Alanine aminotransferase ALT, estimating the activity of the enzyme Aspartate Aminotransferase AST, Alkaline phosphatase ALP, determination of the activity of the Gamma-Glutamyl transferase GGT and determination of the activity of Cholecystokinin CCK in the blood by ELISA technique.

Examine the number of white blood cells

White blood cell counts were performed by complete blood count CBC test.

Physiological and biochemical examinations:

Determination of the efficacy of liver enzymes (ALT, AST, ALP and GGT) by using the FUJI DRI-CHEM kit and by using the FUJIFILM device produced by Fuji, Japan. Also

Determination of the efficacy of CCK hormone in the blood by using the CCK measurement kit produced by CUSABIO, China and by using the ELISA device at a wavelength of 450 nm.

Statistical analysis:

The results were analyzed statistically using (SAS V. 2001), using the ANOVA test to extract the standard rate and error, and the arithmetic means of the coefficients were tested using the Duncan multiple range test with a significant level of 0.05 to determine the significant differences between the groups (**Duncan, 1955**).

Results and discussion:

The effect of neonatal jaundice on white blood cells (WBCs):

The results in Table 1 showed that there was a significant decrease (P <0.05) in the number of white blood cells in the three groups of newborns patients with neonataljaundice within the groups G2, G3 and G4, which included concentrations (10.6-13.9). ((14,0-17,6) (17,7-31,0)) mg / dl, respectively, compared to control group G1. While the groups of children with jaundice did not show any significant differences between them. Our results agreed with (**Rashid, 1998**) who reported no increase in the number of leukocytes in patients and disagreed with (**Al-Muhammadi et al., 2014**) who studied some physiological changes in children with jaundice and reported that there was a slight increase (P <0.05) in the aggregates. Pathogenesis compared to control(AL-Samarraie,et al.,2021).

Table (1) the effect of neonatal jaundice on WBC

Groups / Standard	G1	G2	G3	G4
L WBC Cell/mm^3mg/dl	11.72 ± 0.40 a	10.50 ± 0.31 b	10.16 ± 0.36 b	10.41 ± 0.30 b

* The values shown in the table represent Mean (± SE).

* Letters that differ horizontally indicate the presence of a significant difference at P ≤ 0.05.

The effect of jaundice in newborns on some biochemical and physiological variables

Effect of neonatal jaundice on ALT and AST enzyme activity:

The results in Table 2 showed that there was a significant increase (P <0.05) in the activity of the ALT enzyme in groups of newborns patients with neonataljaundice within the groups G2, G3 and G4, which included concentrations ((10.6-13.9)). 14.0-17.6) (17.7-31.0)) mg/dl respectively, compared with control group G1. While there was a significant decrease (P <0.05) in the activity of the AST enzyme with the same groups above and as shown in Table (2). This is in agreement with (**Edwards, 2002**) who found that ALT levels increased slightly in all patients compared to the control group. The reason for the increase in the ALT enzyme compared to the AST enzyme may be attributed to the fact that the first is mainly present in the liver, which increased its concentration (**Robert et al., 2000**).+

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Also, G3 children with jaundice with a concentration of (14.0-17.6) mg/dl showed a significant increase compared to the other groups with the enzyme ALT. This may indicate a disorder or breakdown in the liver cells.

Table (2) the effect of neonatal jaundice on some liver enzymes (ALT, AST and ALP)

Standard / Groups	ALT U/I	AST U/I	ALP U/I
G1	14.86 ± 0.14 c	36.93 ± 1.42 a	169.83 ± 3.35 c
G2	16.30 ± 0.16 b	30.36 ± 1.22 b	191.0 ± 2.98 b
G3	17.30 ± 0.18 a	31.86 ± 1.46 b	202.7 ± 2.22 a
G4	16.6 ± 0.25 b	29.76 ± 1.50 b	182.9 ± 3.23 b

* The values shown in the table represent Mean (± SE).

* Vertically different letters indicate a significant difference at $P \leq 0.05$

The effect of neonatal jaundice on the activity of the enzyme ALP:

The results of the statistical analysis indicated in the table 2 showed that there was a significant increase ($P < 0.05$) in the activity of the enzyme ALP in the three groups of newborns patients with neonatal jaundice within the groups G2, G3and G4, which included concentrations (10.6-13.9). (14.0-17.6) (17.7-31.0)) mg / dl, respectively, compared to control group G1. This is in agreement with findings (**AbdElmonem et al., 2019; Ahmadpour et al., 2015; Mustafa, & AL-Samarraie,2020**) who studied a total of 102 healthy mature babies and reported that there was a significant difference in levels of alkaline cord blood phosphatase between neonates without jaundice and those with clinically jaundice. It also agrees with findings (**Kattwinkel et al. 1973; Salz et al. 1973**) who stated that at the age of one month, alkaline phosphatase values may be 5-6 times higher than normal values for adults. Concentrations of alkaline phosphate slowly decreased until adulthood, when they may be 3-4 times higher than the adult values. Also, the group of sick children with G3 jaundice within the concentration of (14.0-17.6) mg/dl showed a significant difference compared with the other groups. The reason for this increase may be that there is a significant positive relationship between ALP in cord blood and bilirubin concentration. This is in agreement with (**Ahmadpour et al., 2015**) who found that the level of ALP increased in parallel with the total bilirubin levels, reaching its highest values along with the maximum values for bilirubin.

Effect of neonatal jaundice on GGT enzyme activity:

The table 3 showed that the G3 group with a concentration of (14-17.6) mg/dl showed a significant increase ($P < 0.05$) compared with the control group G1 and other affected groups. The results are close to what he found (**Beath, 2003**) during his study of newborns with jaundice and who breastfeeding were, although it is well known that its value is significantly higher in newborns than in older children: levels up to 120 are considered. -130 units / liter is normal in the pediatric population and this is consistent with our results in groups of children G1, G2, and G4. However, GGT activity in breast milk is very high during the first weeks of lactation.

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Table (3) Effect of neonatal jaundice on GGT enzyme and CCK hormone

Groups / Standard	G1	G2	G3	G4
GGT U/I	108.53 ± 1.82 b	109.30 ± 6.33 b	152.33 ± 9.76 a	121.10 ± 8.02 b
CCK Pg/ml	101.13 ± 3.46 d	177.82 ± 8.46 c	220.22 ± 5.51 a	201.45 ± 4.95 b

* The values shown in the table represent Mean (± SE).
* Characters that differ horizontally indicate the presence of a significant difference at $P \leq 0.05$.

The effect of neonatal jaundice on the effectiveness of cholecystokinin (CCK):

Thetable 3 indicates that there was a significant increase ($P < 0.05$) in the effectiveness of the CCK hormone in the three groups of newborns patients with neonatal jaundice within the groups G2, G3 and G4 which included concentrations (10.6-13.9). (14.0-17.6) (17.7-31.0)) mg / dl, respectively, compared to control group G1. The group of G3 children with jaundice showed a significant difference (14.0-17.6) mg / dl compared with the other groups. The reason for the high blood concentration may be due to the fatty acids present in breast milk, which stimulate the I-cells in the duodenal lining, which works to contract the gallbladder (**Otsuki,2000**) and opens the Audi valve, which leads to the excretion of bilirubin collected in the gallbladder. To the intestine and get rid of it through the stool, so doctors advise to increase the number of times of breastfeeding in order to get rid of the largest amount of bilirubin through the stool.

Conclusions

We can be concluded from the present study that neonatal jaundice may cause significant decrease in the number of white blood cells and the activity of the enzyme AST in the blood serum, while there was a significant increase in the activity of the enzyme ALT, ALP, and GGT, as well as a significant increase in the activity of the CCK hormone in the blood serum of newborns with neonatal jaundice.

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