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ORIGINAL ARTICLE

The efficacy of silymarin in the treatment of physiological neonatal jaundice: a randomized, double-blind, placebo-controlled, clinical trial

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Abstract:

Background: *Unconjugated hyperbilirubinemia is one of the most common conditions in neonates. Conventional treatment are phototherapy and exchange transfusion. Phototherapy is safe and effective, but it has several disadvantages. That indicates the need to develop an alternative pharmacological treatment strategies. It should be less invasive, and at least, as effective and safe as phototherapy. The present study was designed to investigate the effects of Silybum marianum (silymarin) on the duration of phototherapy, which is known to have antioxidant, anti-inflammatory, hepatic protective, regenerative and enhancing glucoronidation activities.*

Patients and Methods: *A randomized double-blind clinical trial was conducted in170 full term healthy neonates with UCB in two well-matched groups. 85 received oral 3.75mg/kg of Silymarin twice daily plus phototherapy and 85 neonates received placebo and phototherapy. Total serum bilirubin (TSB) was measured every 24h, SGPT and SGOT level were measured before and after therapy for both groups.*

Results: *The mean duration of phototherapy was found to be significantly reduced from 5.3 ± 0.82 days in the control group to 4.2 ± 0.76 days in Silymarin-treated group (p=0.001). SGPT and SGOT levels were significantly normalized (p=.001).*

Conclusion: *Silymarin dose of 3.75mg/kg twice daily along with phototherapy was more effective than phototherapy alone in treating full term healthy neonates with UCB.*

Keyword: *silymarin, neonatal jaundice, phototherapy, bilirubin*

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Introduction

Neonatal hyperbilirubinemia is the most common clinical sign in neonatal medicine; it is usually physiological but only rarely is it associated with bilirubin neurotoxicity or the harbinger of significant underlying disease. It reflects accumulation of yellow-orange pigment of bilirubin in the skin, sclera, and other mucous tissues of the neonate. Serum bilirubin level is increased due to imbalance between bilirubin′ s production and elimination [1]. Where the breakdown of erythrocytes and heme-containing protein is accelerated the and the liver is unable to function adequately metabolize all the extra-load of produced bilirubin [2] it was shown by Tazawa study, that 31% of breast-fed jaundiced infants had at least one item of abnormal liver function that may suggest mild hepatic dysfunction [3], which decreases bilirubin elimination. Newborn appears jaundiced when the serum bilirubin level is >7 mg/dl [4]. Severe elevation of serum bilirubin levels can result in brain damage, known as kernicterus which is a lifelong neurologic sequelae and may lead to death [4]. Treating indirect hyperbilirubinemia at the appropriate time is of high importance in neonates. The intensity and invasiveness of therapy are determined by many factors such as gestational age, relative health of neonate, TSB and etiology of jaundice. Phototherapy and exchange

transfusion are two main interventions that are used to decrease TSB, they have many side effects that may reach mortality.

Pharmacologic agents used in the management of hyperbilirubinemia can accelerate bilirubin clearance via the normal metabolic pathways, inhibit the enterohepatic circulation of bilirubin or interfere with bilirubin formation by either blocking the degradation of heme or inhibiting hemolysis [5, 6]. Metalloporphyrin [7], d-penicillamine [8], phenobarbital and clofibrate [8] are pharmacological agents that can be used in management of hyperbilirubinemia.

Herbal therapy, including Silymarin, has recently received special attention as a mode of complementary therapy. Silymarin is a flavonoid complex, which is extracted from seeds of Milk thistle Family: Asteraceae/Compositae) [9]. That was approved by FDA as herbal medicine indicated as a dietary supplement that is widely used in traditional remedies for almost 2000 years as liver tonic in European medicine [10]. The main component of the silymarin complex is silybin [11]. The extracts are still widely used to protect the liver against toxins and to control chronic liver diseases, hepatic viruses, fibroses and jaundice. Recent experimental and clinical studies suggest that milk thistle extracts also have anticancer, antidiabetic, cardioprotective effects, antihyperchlosteremic and induction of breast milk flow [9, 12]. Milk thistle extracts are known to be safe and well tolerated. Toxic or adverse effects, observed in the reviewed clinical trials, seem to be minimal [9], [13].

Attempts to decrease the risk of hyperbilirubinemia should be directed at the early establishment of effective lactation and adequate caloric intake [14].

No clinical trials have been completed in neonate examining the effect of silymarin in treatment of neonatal jaundice. But it is used safely in treatment of neonatal lupus erythematosus with cholestatic hepatitis [15].

The present study was to investigate the efficacy of silymarin as an adjunct therapy that decreases duration of phototherapy for treatment of neonatal jaundice.

Patients and Methods

A blind randomized, placebo-controlled clinical trial was conducted in newborn services at Neonatal Intensive Care Clinical Center of Doctor Abdu Al-Naser Badawy in Sohag, Egypt. Local ethical approval was obtained for the study protocol and all patients were subjected to through history and clinical examination before enrollment in the study. 170 (73 Females) healthy, full term neonates were enrolled in this study, and then randomly assigned to one of the two study groups. All infants were consecutive studied by one blind single investigator, after informed parental consents were obtained. Study group received phototherapy and silymarin [n=85 (40 Females)], and control group received phototherapy and placebo [n= 85 (33 Females)].

Inclusion Criteria

1. Included patients met the criteria of the 2004 AAP (American Academy of Pediatric) treatment guidelines of hyperbilirubinemia using phototherapy [16].
2. Only healthy neonates with unconjugated hyperbilirubinemia, non-hemolytic jaundice, and with no need for urgent exchange transfusion.
3. Healthy near-term and full-term newborns infants' gestational age (38-42 weeks), have jaundice in the age of 1-10 days.
4. Laboratory tests with negative direct combs test.

Exclusion criteria

1. Birth weight less than 2500gm.
2. Prior or current use of phenobarbitone by the mother or child [17].
3. Initial indication of double or triple phototherapy.
4. Newborn submitted to blood transfusion.
5. Newborn with congenital defect; hereditary disease of erythrocytes or autoimmune disease with intense haemolysis.
6. Newborn with conjugated hyperbilirubinemia or any other disease rather than jaundice were excluded (sever sepsis, pneumonia, respiratory distress, anemia, etc.).
7. Newborn with ABO or Rh incompatibility.
8. Newborn with decreased glucose-6-phosphate dehydrogenase (G6PD) determination.

Included patients could have to be excluded from the research due to the following exclusion criteria:

1. Registering spectral irradiance below 4.0μW/cm²/nm in any of the measurements for phototherapy calibrations. [18]
2. Changing modality of phototherapy to double or triple.
3. Technical or clinical impossibility to determine TSB.
4. Death during the period of phototherapy.

The inclusion criteria for starting phototherapy according to AAP and those to stop phototherapy according to an internal guidelines depending on TSB, age, gestational age of the neonate. All the criteria of inclusion and discharge are the same for

both control and Silymarin treated group. There were no infants excluded from the study during the study from both groups.

Laboratory tests: The laboratory technician was blinded; he didn′t know the patient group. TSB was measured on admission, after 12 hours of admission and every 24 hours until discharge. Aspartate aminotransferase (SGOT) and Alanine aminotransferase (SGPT) were measured in all neonates both before and after the study.

A dose of 3.75 mg/kg of silymarin syrup dosage form was received twice daily orally by infants in the silymarin-treated group within 12 hours of admission. Laboratory tests including complete blood count, total and direct serum bilirubin, reticulocyte count, direct Coombs′ agglutination test, maternal and neonatal blood group, G6PD determination and peripheral blood smear were performed and recorded routinely before the beginning of therapy for all jaundiced infants in both groups. Total and direct serum bilirubin levels were measured daily until phototherapy was discontinued.

Phototherapy was started immediately on admission for all studied patients case and control until TSB decreased to a safe level according to the internal guidelines which depended on the infant′ s gestational and postnatal age. A nurse who was not involved in drug administration recorded duration of phototherapy. Each phototherapy unit contained 8 special white fluorescent tubes labeled TL 52/20w (Philips, Eindhoven, Netherlands) adjusted at a 20 cm distance above the infant. During study all neonates had a careful physical observation of any symptoms appeared on neonate as vomiting, loose stools, skin rashes and hyperthermia. Laboratory tests were followed 48 hours and 1week after discharge included complete blood count, TSB, for detection of rebound hyperbilirubinemia. Lamps of phototherapy units were changed regularly after 1500 hours of usage to keep irradiance in the photo effective range. TSB measurement was performed on the basis of spectrophotometric principles by using Bilimeter3 (Pfaff Medical GmbH, Germany). Direct bilirubin measurement was performed by using Autoanalyser Random Access (Selectra E, Vital Scientific, Netherlands). The equipments were standardized periodically.

All data were analyzed using statistical package of social sciences SPSS V15.0 (SPSS Inc., Chicago, IL). Statistical analysis of data was performed by Student t- test to compare between the two groups and Paired t- test to compare within each group and p values less than 0.05 were considered significant for all checked results.

Results

170 neonates studied, (73 Females) completed the study. Table 1 show the characteristics and clinical data of control and Silymarin-treated groups collected prior to therapy. Table 2 shows the mean±S.D. data and statistical significance of both control and Silymarin-treated groups, from which there were no significant differences in age, gestational age, mean total serum bilirubin, SGPT and SGOT at the time of admission of neonates between both groups.

The mean duration of phototherapy was significantly lower in control group in comparison with silymarin-treated group ($p<0.01$). Both SGPT and SGOT were significantly increased ($p<0.01$) within normal SGPT and SGOT serum level in silymarin-treated group at the end of therapy where it was increased insignificantly at the end of therapy in control group.

As shown in Figure 1, the reduction rate, amount removed per unit time, of total and indirect plasma bilirubin levels were significantly higher in the silymarin-treated group compared to the control group. Asterisks at 60h and 84h of life on figure 1 are signs of significance. The difference became significant ($p<0.05$) between mean TSB of the two groups in day three of therapy. Table 3 demonstrates a comparison between Number and percent of symptoms; those were recorded during duration of therapy in both groups. During the duration of study, two cases of rebound hyperbilirubinemia were recorded from the control group.

Discussion

Jaundice is the most common condition that requires medical attention in newborns. Conventional treatment of jaundice includes phototherapy and exchange transfusion in severe cases. They have various and serious adverse effects. Development of intensified Phototherapy units and the use of drugs have contributed significantly to decrease the need for exchange transfusion due to itś high risk of morbidity and mortality. Efficacy of phototherapy needs a lot of precautions to justify the required minimal effective dose. Hence, numerous newborns still submitted to subtherapeutic doses of phototherapy, which may lead to neurological sequelae that may not be detected in childhood [18, 19]. Several pharmacological drugs are used to treat neonatal jaundice [8]. The belief, that the natural medicines are much more safe than synthetic drugs, has gained popularity in recent years and led to tremendous growth of phytopharmaceutical usage [20].

Table 1. Basic clinical data of and jaundice risk factor in the two study groups.

Variable	Silymarin-treated group	Control group
Age, mean (SD), day	(2-7), 3.69 (2.488)	(2-8), 3.54 (2.58)
Gestational age, mean (SD), week	(38-42), 38.94 (4.2)	(38-42), 39.01, (2.8)
Gender, NO. (%)		
-Male	45, (53)	52, (61)
-Female	40, (47)	33, (39)
Mode of delivery, NO. (%)		
-Normal	55, (65)	56, (66)
-Cesarean section	30, (35)	29, (44)
Consanguinity, NO. (%)		
-Yes	62, (73)	60, (71)
-No	23, (27)	25, (29)
Feeding, NO. (%)		
-Formula:	37, (44)	33, (39)
-Mixed:	48, (56)	52, (41)
Hyperbilirubinemia in previous siblings: NO. (%)		
-No previous sibling:	10, (12)	7 , (8)
-Present	47, (52)	56, (66)
-Absent	28, (36)	29, (26)

p-value= 0.695 (insignificant), p-value=0.904

Table 2. Main result of the study in both control and Silymarin-treated groups.

Variables	Minimum	Maximum	Mean±S.D.	p-value
TSB				
control	8.79	24.38	15.38±3.61	**0.837**
Silymarin-treated	9.43	24.35	15.26±3.80	
Duration of phototherapy				
Control	84	168	127.47±19.61	**<0.001*****
Silymarin-treated	64	132	100.66±18.30	
SGOT before				
Control	23	91	50.67±14.55	**0.342**
Silymarin-treated	17	89	48.35±17.04	
SGOT after				
Control	34	89	58.88±14.68	**<0.001*****
Silymarin-treated	63	123	94.84±14.26	
SGPT before				
Control	10	31	19.51±4.84	**0.721**
Silymarin-treated	10	35	19.80±5.84	
SGPT after				
Control	15	35	24.80±4.53	**<0.001*****
Silymarin-treated	12	43	35.15±5.23	

*** Significant at 0.05 level, ** Significant at 0.01 level, *** Significant at 0.001 level**

Table 3. Number and percent of symptoms appeared during duration of therapy in each of control and silymarin-treated group.

Other symptoms	Silymarin-treated group Number (%)	Control group Number (%)	p-value
Number of neonates showed no other symptoms during study	24.0 (28.2)	16.0 (18.8)	0.205
Skin rash from (mild to severe)	7.0 (8.2)	36.0 (42.4)	0.001**
Vomiting	21.0 (24.7)	46.0 (54.1)	0.001**
Hyperthermia	8.0 (9.4)	12.0 (14.1)	0.475

*** Significant at 0.05 level, ** Significant at 0.01 level, *** Significant at 0.001 level**

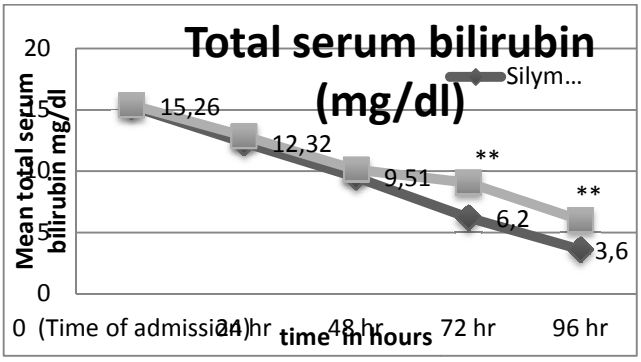

Figure 1. Mean total serum bilirubin (mg/dl) measured every 24hours along the duration of therapy in the two groups.
**** Highly significant p-value**

Physiological jaundice in neonates may be contributed to the fact that liver is unable to function adequately so we need to support liver function [2]. Silymarin is a natural herbal supplement that supports liver activity with an evidence of wide margin of safety. It has several mechanism of action that may contribute to reduction of serum bilirubin. There was no previous study had been published using Silymarin in treatment of neonatal jaundice. Silymarin was used in treatment of a neonatal lupus erythmatosus with cholestatic hepatitis [15]. It has several mechanism of action one or more of them can reduce total serum bilirubin level. It can enhance Glucuronidation [21-23]. It inhibits reabsorption of bilirubin by enterohepatic circulation through Its mild laxative effect [12, 24, 25]. It stimulates ribosomal RNA polymerase and subsequent protein synthesis, and hence enhances hepatocyte regeneration which may promote the liver to function adequately to metabolize bilirubin. It has an antioxidant effect that may resemble the adaptive role of physiological neonatal jaundice in scavenging reactive oxygen species. It has the ability to regulate membrane permeability [21]sand so increasing membrane stability and decreasing excess heam metabolism by stabilizing RBCs.

Oral syrup dosage form with enhanced bioavailability preparation was used in the study so we avoid contamination of herbal medicines by any heavy metal, microbial toxins or any other contaminants. Silymarin increased the incidence of loose stools with phototherapy, which may have a beneficial effect in lowering hyperbilirubinemia.

In the present study, there were increased numbers of appearances of jaundice and duration of therapy in breastfed infants and hyperbilirubinemia in previous sibling. Hence, breastfeeding and hyperbilirubinemia in previous sibling might be considered as risk factors in neonatal jaundice. There was no correlation between sex, blood group of neonate and appearance of jaundice or duration of therapy. The duration of phototherapy and hospitalization was significantly shorter in infants that were treated with silymarin in addition to phototherapy in comparison with those treated with only phototherapy. As shown in figure 1, total serum bilirubin was significantly decreased in day three of silymarin therapy. No important side effect was determined during the short-term follow-up of infants. Data statistics demonstrated that duration of phototherapy was significantly reduced from 5.3 ± 0.82 days in the control group to 4.2 ± 0.76 days in silymarin-treated group (p=0.001). SGPT and SGOT liver function tests were used in previous studies to indicate safety of some drugs on the liver of the neonate as in measuring the safety of paracetamol on neonatal liver [26], also it was used to follow up cholestasis in neonate [27], [28] and it was used to measure the efficiency and health of the liver [29].

In silymarin-treated group, first values of SGPT and SGOT before therapy was either lower than the normal

range or at the lower limit. At the end of therapy mean SGPT and SGOT values were found to be increased significantly to higher values within the normal range. In control group there was no significant increase in mean SGPT and SGOT values. This may indicate better activity of the liver which means that silymarin can normalize SGPT and SGOT [30]. After the statistical analysis and due to the significant increase of SGPT and SGOT values in Silymarin-treated group. Also there was a little significant pearson correlation SGPT (r=0.23, p-value=.032), and week highly significant Pearson correlation between the duration of therapy and SGOT (r=0.43, p-value=0.001) in silymarin-treated group. This correlation was not found in control group. There was no serious side effect observed during duration of therapy with silymarin. Similar to phenobarbital, silymarin has the same action of also enhance bilirubin conjugation and excretion [21-23] and is a better herbal drug with a wide margin of safety. Phenobarbital has a long half life [31] and many factors can affect the clearance of phenobarbital during the neonatal period [32]. In Heiman & Gladlk study, phenobarbital half life was significantly longer in neonates (118.6±16.1h) [33]. That means its half life may reach more than two days, while clearance half life of silymarin is six to eight hours [21, 34]. Phenobarbital also causes drowsiness in neonates and may slow down the oxidation of bilirubin in the brain leading to worse bilirubin toxicity [8]. Silymarin reduced and restored the phenobarbitone induced sleeping time [35].

From Table 4 it could be noted that silymarin significantly reduced the incidence of skin rash as a side effect of phototherapy. Silymarin significantly decreased the incidence of vomiting of the neonates.

Conclusion

Silymarin dose of 3.75mg/kg twice daily along with phototherapy is more effective than phototherapy alone in treating full term healthy neonates with UCB.

Further studies are required to fully understand Silymarin′s role in treatment of neonatal jaundice and possibility to be used as a prophylactic therapy or to be used in managing pathological neonatal jaundice; also to determine the most effective dose.

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