The efficacy of *Silybum marianum* (L.) Gaertn. (Silymarin) in the treatment of physiological neonatal jaundice: a randomized, double-blind, placebo-controlled, clinical trial

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Back ground and aim of work

Unconjugated hyperbilirubinemia (UCB) is one of the most common conditions in neonates. Conventional treatments are phototherapy and exchange transfusion. Phototherapy is safe and effective, but it has several disadvantages, which indicates the need to develop alternative pharmacological treatment strategies. These alternative treatment strategies 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, and regenerative properties, including enhancing glucuronidation activities.

Patients and methods

A randomized double-blind clinical trial was conducted on 170 full-term healthy neonates with UCB divided into two well-matched groups. Of the 170 neonates, 85 received 3.75 mg/kg of silymarin orally, twice daily, in addition to phototherapy, and 85 received placebo and phototherapy. Total serum bilirubin was measured every 24 h, and alanine aminotransferase (SGPT) and alanine transaminase (SGOT) levels were measured before and after therapy in 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 the silymarin-treated group ($P = 0.001$). SGPT and SGOT levels were significantly normalized ($P = 0.001$).

Conclusion

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

Keywords:
bilirubin, neonatal jaundice, phototherapy, silymarin

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Introduction

Neonatal hyperbilirubinemia is the most common clinical symptom in neonatal medicine; it is usually physiological but only rarely is it associated with bilirubin neurotoxicity or with significant underlying disease. It reflects accumulation of a yellow–orange pigment of bilirubin in the skin, sclera, and other mucous tissues of the neonate. The serum bilirubin level is increased because of imbalance between the production and elimination of bilirubin [1]. When the breakdown of erythrocytes and heme-containing protein is accelerated, the liver is unable to function adequately to metabolize the extra load of bilirubin produced [2]. It was shown in the study by Tazawa *et al.* [3] that 31% of breast-fed infants with jaundice had at least one item of abnormal liver function that may suggest mild hepatic dysfunction, decreasing bilirubin elimination. Newborns appear jaundiced when the serum bilirubin level is greater than 7 mg/dl [4]. Significant elevation of serum bilirubin levels can result in brain damage, known as kernicterus, which is a life-long 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 the neonate, total serum bilirubin (TSB), and etiology of jaundice. Phototherapy and exchange transfusion are two main interventions that are used to decrease TSB. Phototherapy has several disadvantages. Most notably, short-term phototherapy does not always decrease plasma UCB to nontoxic levels in neonates, whereas long-term phototherapy, such as that needed for patients with Crigler–Najjar disease, becomes less effective with age and has a profound impact on social life. Under conditions of very severe unconjugated hyperbilirubinemia (UCB) or hyperbilirubinemia with an insufficient response to phototherapy, a 'rescue' treatment consists of exchange transfusion in which the hyperbilirubinemic blood is removed and is replaced with nonjaundiced blood. Exchange transfusion,

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however, has considerable morbidity, especially in sick preterm newborns; mortality has also been reported [5,6]. The potential neurotoxicity of UCB and the disadvantages of the present treatments have prompted the investigation into and development of alternative pharmacological treatment strategies for UCB. These alternative treatment strategies should be less invasive and at least as effective and safe as phototherapy.

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

Herbal therapy, including silymarin, has recently received special attention as a mode of complementary therapy. Silymarin is a flavonoid complex that is extracted from seeds of milk thistle (family: *Asteraceae*/*Compositae*) [9]. This has been approved by FDA as a herbal medicine and has been indicated as a dietary supplement. It has been widely used in traditional European medicine as a liver tonic for almost 2000 years[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 have suggested that milk thistle extracts also have anticancer, antidiabetic, cardioprotective, and antihypercholesterolemic effects and induce the flow of breast milk [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 at adequate caloric intake [14].

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

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

Patients and methods

A blind, randomized, placebo-controlled clinical trial was conducted at Doctor Abdu Al-Naser Badawy's Neonatal Intensive Care Clinical Center in Sohag, Egypt. Approval from the local ethical committee was obtained for the study protocol and all patients were subjected to through history taking and clinical examination before enrollment. A total of 170 (73 girls) healthy, full-term neonates were enrolled into this study and randomly assigned to one of two study groups. All infants were consecutively studied by one blinded investigator after informed parental consent had been obtained. The study group received phototherapy and silymarin [$n = 85$ (40 girls)], and the control group received phototherapy and placebo [$n = 85$ (33 girls)].

Inclusion criteria:

(1) Patients who fulfilled the criteria of the 2004 American Academy of Pediatrics guidelines for the treatment of hyperbilirubinemia using phototherapy [16].
(2) Healthy neonates with UCB, nonhemolytic jaundice, and who did not require an urgent exchange transfusion.
(3) Healthy near-term and full-term newborns with a gestational age of 38–42 weeks, having jaundice at the age of 1–10 days.
(4) Those with negative results for the direct coombs test.

Exclusion criteria:

(1) Newborns with birth weight less than 2500 g.
(2) Prior or current use of phenobarbitone by the mother or the child [17].
(3) Initial indication of double or triple phototherapy.
(4) Newborns subjected to blood transfusions.
(5) Newborns with congenital defects, hereditary disease of erythrocytes, or autoimmune diseases with intense hemolysis.
(6) Newborns with conjugated hyperbilirubinemia or with any disease other than jaundice (severe sepsis, pneumonia, respiratory distress, anemia, etc.).
(7) Newborns with ABO or Rh incompatibility.
(8) Newborns with decreased levels of glucose-6-phosphate dehydrogenase.

After the initial selection, the neonates were excluded from the research if any of the following criteria were met:

(1) Spectral irradiance below 4.0 μW/cm/nm was registered for any of the measurements for phototherapy calibrations. [18]
(2) The modality of phototherapy was changed to double or triple.
(3) Determination of TSB was technically or clinically impossible.
(4) Death occurred during the period of phototherapy.

The inclusion criteria for starting phototherapy according to American Academy of Pediatrics and to stop phototherapy according to internal guidelines depended on TSB, age, and gestational age of the neonate. All the criteria of inclusion and discharge were the same for both control and silymarin-treated groups. No infant was excluded from either group during this study.

Laboratory tests

The laboratory technician was blinded to the patient group.

TSB was measured on admission, after 12 h of admission, and then every 24 h 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 was administered orally, twice daily, to the infants in the silymarin-treated group within 12 h of admission. Laboratory tests including determination of complete blood count, total and direct serum bilirubin levels, reticulocyte count, maternal and neonatal blood groups, and glucose-6-phosphate dehydrogenase level; direct Coombs agglutination test and peripheral blood smear were also performed and recorded routinely before initiating therapy in all jaundiced infants in both groups. Total and direct serum bilirubin levels were measured daily until phototherapy was discontinued.

Phototherapy was initiated immediately on admission for both patients and controls 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 the duration of phototherapy. Each phototherapy unit consisted of eight special white fluorescent tubes labeled TL 52/20w (Philips, Eindhoven, the Netherlands) adjusted 20 cm above the infant. During the study, all neonates underwent careful physical observation for any symptoms such as vomiting, loose stools, skin rashes, and hyperthermia. Laboratory tests were conducted 48 h and 1 week after discharge and included complete blood count and TSB for detection of rebound hyperbilirubinemia. Lamps of phototherapy units were changed regularly after 15:00 h of usage to maintain irradiance in the photoeffective range. TSB measurements were taken on the basis of spectrophotometric principles using Bilimeter3 (Pfaff Medical GmbH, Germany). Direct bilirubin measurement was taken using Autoanalyser Random Access (Selectra E; Vital Scientific, the Netherlands). The equipments were standardized periodically.

All data were analyzed using SPSS V15.0 (SPSS Inc., Chicago, Illinois, USA). Statistical analysis of the data was carried out using Student's *t*-test for between group comparisons and the paired *t*-test for within group comparisons. *P*-values less than 0.05 were considered significant for all checked results.

Results

A total of 170 neonates (73 girls) completed the study. Table 1 shows the characteristics and clinical data of the control and silymarin-treated groups collected before therapy. Table 2 shows the mean ± SD and statistical significance of both the control and silymarin-treated groups; no significant differences in age, gestational age, mean TSB, SGPT, and SGOT at the time of admission of neonates were noted between the groups.

Table 1 Basic clinical data and risk factors for jaundice in the two study groups

Variables	Silymarin-treated group	Control group
Age [mean (SD), days]	(2–7), 3.69 (2.488)*	(2–8), 3.54 (2.58)*
Gestational age [mean (SD), weeks]	(38–42), 38.94 (4.2)**	(38–42), 39.01, (2.8)**
Sex [*N* (%)]		
Male	45 (53)	52, (61)
Female	40 (47)	33, (39)
Mode of delivery [*N* (%)]		
Normal	55 (65)	56, (66)
Cesarean section	30 (35)	29, (44)
Consanguinity [*N* (%)]		
Yes	62 (73)	60 (71)
No	23 (27)	25 (29)
Feeding [*N* (%)]		
Formula	37 (44)	33 (39)
Mixed	48 (56)	52 (41)
Hyperbilirubinemia in previous siblings [*N* (%)]		
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 ± SD	*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	

SGOT, alanine transaminase; SGPT, alanine aminotransferase; TSB, total serum bilirubin.
*Significant at 0.001 level.

The mean duration of phototherapy was significantly lower in the control group compared with the silymarin-treated group ($P<0.01$). Both SGPT and SGOT were significantly increased ($P<0.01$) in the silymarin-treated group at the end of therapy, where as it was increased insignificantly at the end of therapy in the control group.

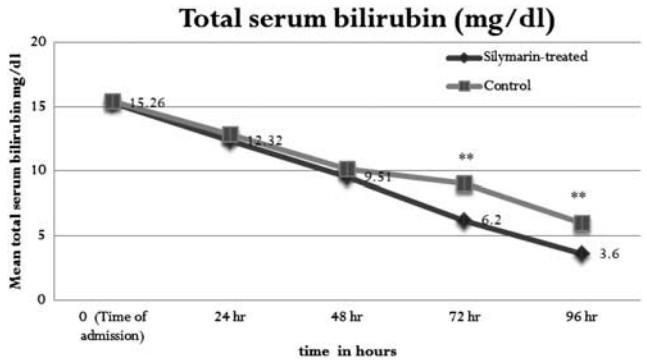
As shown in Fig. 1, the reduction rate (the amount removed per unit time) of total and indirect plasma bilirubin levels was significantly higher in the silymarin-treated group compared with the control group. Asterisks at 60 and 84 h of life in Fig. 1 are signs of significance. The difference in mean TSB between the two groups became significant ($P<0.05$) on day 3 of therapy. Table 3 shows a comparison between the number and percentage

of symptoms recorded during therapy in both groups. During the study, two cases of rebound hyperbilirubinemia were recorded in the control group.

Discussion

Jaundice is the most common condition that requires medical attention in newborns. Conventional treatment for jaundice includes phototherapy and exchange transfusion in severe cases. These therapeutic modalities have various and serious adverse effects. Development of intensified phototherapy units and the use of drugs have contributed significantly to a reduction in the need for exchange transfusion, which is associated with a high risk of morbidity and mortality. Efficacy of phototherapy needs a lot of precautions to justify the required minimal effective dose. Hence, numerous newborns continue to be subjected 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 natural medicines are safer compared with synthetic drugs has gained popularity in recent years and has led to tremendous growth in phytopharmaceutical usage [20]. Physiological jaundice in neonates may be because the liver is unable to function adequately, and hence there is a need to support liver function [2]. Silymarin is a natural herbal supplement that supports liver activity with evidence of a wide margin of safety. It has several mechanisms of action that may contribute to reduction of serum bilirubin. No study has been published previously on the use of silymarin in the treatment of neonatal jaundice. Silymarin was used in the treatment of a neonatal lupus erythmatosus with cholestatic hepatitis [15]. It has several mechanisms of action, one or more of which can reduce the TSB level. It can enhance glucuronidation [21–23], inhibit reabsorption of bilirubin by enterohepatic circulation through its mild laxative effect [12,24,25], and stimulate ribosomal RNA polymerase and subsequent protein synthesis and thus enhance hepatocyte regeneration, which may drive 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 also has the ability to regulate membrane permeability [21], thus increasing membrane stability and decreasing excess heme metabolism by stabilizing RBCs.

Figure 1



Mean total serum bilirubin (mg/dl) measured every 24 h throughout the duration of therapy in the two groups. **Highly significant *P*-value.

Orally administered syrups with enhanced bioavailability was used in the study, and thus we were able to avoid contamination of herbal medicines by heavy metals, microbial toxins, and 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 was increased incidence of jaundice and increased duration of therapy in breast-fed infants and increased incidence of hyperbilirubinemia in previous siblings. Hence, breastfeeding and hyperbilirubinemia in previous siblings might be considered risk factors for neonatal jaundice. There was no correlation between sex or blood group of the neonate and appearance of jaundice or duration of therapy. The duration of phototherapy and hospitalization was significantly shorter in infants treated with silymarin in addition to phototherapy in comparison with that in those treated with only phototherapy. As shown in Fig. 1, TSB was significantly decreased on day 3 of silymarin therapy. No important side effects were identified during the short-term follow-up. Statistics demonstrated that the duration of phototherapy was significantly reduced from 5.3 ± 0.82 days in the control group to 4.2 ± 0.76 days in the silymarin-treated group ($P = 0.001$). SGPT and SGOT liver function tests were used in previous studies to indicate the safety of certain drugs with regard to the

Table 3 Number and percentage of symptoms that appeared during the duration of therapy in the control and silymarin-treated groups

	N (%)		
Other symptoms	Silymarin-treated group	Control group	*P*-value
Number of neonates who 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 0.01 level.

liver of the neonate, such as the safety of paracetamol on neonatal liver [26]; in addition, these tests were used in the follow-up of cholestasis in neonates [27,28] and to evaluate the efficiency and health of the liver [29].

In the silymarin-treated group, the initial values of SGPT and SGOT before therapy were either lower than the normal range or at the lower limit. At the end of therapy, the mean SGPT and SGOT values were found to have increased significantly to higher values within the normal range. In the 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], which was concluded after the statistical analysis and on the basis of the significant increase in SGPT and SGOT values in the silymarin-treated group. In addition, there was little significant Pearson's correlation between SGPT and duration of therapy ($r = 0.23$, P-value $= 0.032$) and weak highly significant Pearson's correlation between SGOT and duration of therapy ($r = 0.43$, P-value $= 0.001$) in the silymarin-treated group. This correlation was not found in the control group. No serious side effect was observed during the duration of therapy with silymarin. Similar to phenobarbital, silymarin also enhances 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 the study by Heiman and Gladlk, phenobarbital half-life was significantly longer in neonates (118.6 ± 16.1 h) [33]. This means that its half-life may reach more than 2 days, whereas the clearance half-life of silymarin is 6–8 h [21,34]. Phenobarbital also causes drowsiness in neonates and may slow down the oxidation of bilirubin in the brain, leading to more severe bilirubin toxicity [8]. Silymarin reduced and restored the phenobarbitone-induced sleeping time [35].

Table 3 shows that silymarin significantly reduced the incidence of skin rash as a side effect of phototherapy and also significantly decreased the incidence of vomiting in neonates.

Conclusion

Silymarin at a dose of 3.75 mg/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 the treatment of neonatal jaundice, its most effective dose, and the possibility of it being used as a mode of prophylactic therapy and for managing pathological neonatal jaundice.

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Conflicts of interest

There are no conflicts of interest.

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