

The protective role of Deferoxamine in the prevention of hepatic fibrosis in children treated with Doxorubicin: a randomized controlled clinical trial

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Abstract

Background: Hepatic fibrosis is an ominous sign which may follow treatment with Doxorubicin (DOX) chemotherapy. The aim of this study was investigating the protective effect of Deferoxamine (DFO) against hepatic fibrosis in treatment-naïve pediatric cancer patients.

Methods: In this prospective randomized controlled trial, 61 treatment-naïve children (2-18 years) with different types of cancer who referred to a tertiary teaching hospital in South of Iran were enrolled. They were randomly assigned to 3 groups; group 1 (control, n=21), group 2 (DFO 10 times DOX dose, n=20), group 3 (DFO 50mg/kg, n=20). DFO was administered as an 8-hour continuous intravenous infusion during and after DOX infusion in each chemotherapy cycle. Non-invasive serum markers of liver fibrosis including APRI, FIB-4 score and Fibro Test were measured in each individual. Besides, hepatic Fibro Scan was used after the last course of chemotherapy to estimate fibrosis degree.

Results: Fifty-six patients were analyzed. Alanine aminotransferase was mildly increased in the treatment groups compared to pre-treatment. Treatment with DFO 10 times DOX dose was associated with significant decline in post-treatment APRI (adjusted odds ratio 0.17; 95% confidence interval 0.03-0.84). METAVIR fibro scores were in the F0-F1 zone in all participants, and the results were comparable in the study groups. No adverse drug effect was reported in the treatment groups.

Conclusion: DOX may not lead to severe liver fibrosis if the maximum allowed cumulative dose is not exceeded. DFO at the dose of 10 times of DOX dose may have a potential protective role against liver fibrosis. Larger multi-center studies with longer follow up are warranted to further assess this issue.

Introduction

Anthracyclines are a group of chemotherapy agents which play a main role in the treatment of different types of childhood cancer [1, 2]. They include doxorubicin (DOX), daunorubicin, epirubicin, idarubicin, and mitoxantrone. It is speculated that their anti-neoplastic action is taken through the induction of topoisomerase II, failure of DNA stands and interference with DNA formation. Chemotherapy is generally associated with toxicity to healthy organs including liver[3]. Hepatic damage is usually observed 1 to 4 weeks after the treatment has begun. Hepatotoxicity is often induced by the drugs as haptens and prohaptens which lead to immunological injury [4, 5]. Although the exact mechanism of acute DOX toxicity has yet to be known, it is believed that it is mediated through free radical formation, iron-dependent oxidative damage of biological macromolecules, and lipid peroxidation of cell membrane [6]. Induction of apoptosis and alteration of nitric oxide metabolism are other mechanisms that may be associated with DOX side effects [7, 8]. The toxicity to vital organs including heart and liver is a major limiting step in wide-spread use of DOX, and a big concern in those who received high cumulative doses of the drug [9].

On the other hand, Deferoxamine (DFO), as an iron chelator, can bind to ferric ion with its hydroxamic acid group, and enhances the drug excretion in the urine. Chelation of excess iron prevents its deposition and damage to several organs and tissues including liver [10]. It also prevents collagen accumulation and

hepatic satellite cell activity, thus decreases liver injury [11]. In addition, DFO was shown to have anti-proliferative effect which may ultimately lead to cell cycle arrest and apoptosis [12]. Furthermore, DFO maintains the activity of the liver enzyme glucose-6-phosphatase [13]. It can also serve as an antioxidant, and reduces liver fibrosis which is manifested as a decreasing trend in serum markers of liver fibrosis including liver hydroxyproline and liver α -smooth muscle actin (α -SMA). Therefore, DFO through different mechanisms decreases profibrogenic factors, and inhibits inflammatory cascade [9-12].

There is no single antidote with proven efficacy in the human being to protect against DOX-induced hepatotoxicity. Although a number of animal studies have shown some benefit with using DFO, no clinical trial has been conducted in patients with cancer to assess its efficacy. Therefore, this study was designed to assess whether DFO has any beneficial role in the prevention of hepatotoxicity in children treated with DOX. Our primary endpoint was to compare hepatic fibrosis measurements in treated and non-treated groups. Our secondary endpoint was to assess whether different doses of DFO has different efficacy compared to the control subjects.

Methods

This single-center, prospective, parallel-group, and randomized-controlled trial was conducted in a referral oncology hospital in south of Iran from July 2016 to April 2017. The study population included treatment-naïve children with the age range of 2–18 years with different types of cancer who were going to be treated with DOX as part of their chemotherapy regimen. Patients with primary or metastatic liver tumors, and those with underlying liver disease or active hepatitis B or C infection were excluded. The study was approved by the Ethics Committee of Shiraz University of Medical Sciences with the code number IR.SUMS.REC.1394.193. The study protocol was registered in Iranian Registry of Clinical Trials at www.irct.ir with registry number IRCT2016080315666N4. Informed written consent was obtained from all participants or their parents.

Given that there was no previous experience with DFO with regards to its hepatoprotective effect in cancer patients, we assumed that patients treated with DFO would have lower probability of DOX-associated liver fibrosis compared to the non-treated patients. Therefore, we used aspartate aminotransferase [AST]-to-Platelet Ratio Index (APRI) as an indirect marker of hepatic fibrosis, and compared APRI in the treatment groups with their control counterparts. A value of 0.5 was considered as the cut-off point to discriminate mild vs no fibrosis [14]. A five percent decrease in the baseline APRI was considered as an acceptable antifibrotic effect [15].

Accordingly, assuming a type I error of 0.05, and a statistical power of 80%, minimum sample size was estimated at 16 patients in each study group. We decided to enroll at least 20 patients in each group to compensate for a possible 20% drop out due to lost to follow up.

During the study period, 123 new cases were admitted in our center that 85 patients were eligible to be enrolled. Among these eligible patients, 61 persons accepted to participate in the study. They were randomly allocated to three groups using a computer-generated block randomization sequence which

was done by a statistician who was blind to the study protocol. Group 1 (n = 21) consisted of patients who served as control group, and no intervention was done. Patients allocated to group 2 (n = 20) and group 3 (n = 20) were pre-treated with DFO (Desferal®, Novartis, Switzerland) 10 times the DOX dose and 50 mg/kg, respectively. Intravenous infusion of DFO was started 2 hours prior to starting chemotherapy, continued during DOX infusion (at least 4 hours), and for another 2 hours after termination of the infusion, making up a total of 8 hours. This regimen was repeated in each chemotherapy course alongside with the infusion of DOX.

Liver function tests (LFT) and complete blood count (CBC) were measured before and after treatment.

After the last course of chemotherapy, γ -glutamyltransferase (γ GT), Haptoglobin, α 2 -macroglobulin (α 2 MG) and apolipoprotein A1 (apoA1) were measured by Enzyme linked immunosorbent assay (ELISA) (Bioassay, China). FIB-4 score and APRI were calculated using the following formulas: FIB-4 score = [age (Y) \times AST (U/L)] / [platelet count (10^9 /L) \times square root of alanine aminotransferase (ALT) (U/L)].

$$\text{APRI} = [\text{AST (U/L)} / 45] \times 100 / \text{platelet (} 10^9/\text{L)}$$

FIB-4 score \leq 1.45 and APRI \leq 0.5 were considered as no fibrosis while the values more than 3.25 and 1.5 were regarded as significant fibrosis, respectively [14, 16, 17]. FIB-4 Scores were dichotomized as zero for scores equal or less than 1.45 and one for scores more than 1.45 [18].

FibroTest combines five standard biomarkers including γ GT, total bilirubin, α 2 MG, apoA1, and Haptoglobin. These markers are weighted depending on the patient's age and sex. A cut-off value of 0.58 was assigned to delineate severe fibrosis [19].

Additionally, liver stiffness was measured in all patients by FibroScan or transient elastography (TE) (Echosens, Paris, France). Liver stiffness was evaluated while the patient was lying on dorsal decubitus position, with the arms in maximal abduction. The measurements were taken in the right and left intercostal spaces. Liver stiffness was expressed in kilopascals (kPa) and was computed for each subject as the median of 10 validated measurements in accordance with the manufacturer's instructions. Measurements with an interquartile range of $<$ 30% of the median value and a success rate of $>$ 60% were considered reliable. Two-dimensional shear wave elastography (SWE) studies were performed using the Aixplorer ultrasound system (SuperSonic Imagine SA, Aix-en-Provence, France) with a convex broadband probe (SC6-1, Super-Sonic Imagine). This technique has an advantage that the probe can be installed on ultrasound machines. At the time of SWE examination, the patients were asked to hold their breath for 3 to 4 s. Liver stiffness was recorded in the right lobe, while the patient was lying on dorsal decubitus position, in accordance with the protocol used for FibroScan. An SWE box was placed 1.5 to 2 cm away from the Glisson capsule and on liver parenchyma to avoid measurements of large vessels. For quantitative measurements, a round region of interest was placed inside the SWE box, and minimum and maximum values of stiffness expressed in kPa were recorded. Four measurements were made, and the median value was recorded. METAVIR fibrosis score, which is graded on a 5-point scale from 0 to 4, was

used to delineate the degree of fibrosis (Supplement Table 1). The scores were compared between the three groups.

Normality of data was checked by Shapiro-Wilk test. Bivariate comparison of quantitative variables among three groups was done by ANOVA and Kruskal Wallis tests. Chi-square test was used to compare qualitative variables between the three groups. Paired t-test, Wilcoxon signed-rank test, and McNemar's test were used to compare data before and after treatment in each group. Null hypothesis was assumed to be a decrease of less than 5% of APRI scores compared with their respective baselines. Then, a binary variable was generated considering change value in the patient APRI score (i.e. APRI score after the intervention minus APRI score at baseline), in which patients with more than five percent decrease in their APRI score considered as observations with good outcome and others were defined as poor outcome observations. Binary Logistic regression was applied to estimate independent (Adjusted for age, sex and baseline APRI score) effect of the treatment in terms of adjusted odds ratio (OR) and its robust 95% confidence interval (95% CI). A two-sided p-value of less than 0.05 or a one-sided p less than 0.025 were considered statistically significant. Data were analyzed using SPSS software (version 21).

Results

During the study period, one patient who was assigned to group 2 and 2 patients in group 3 died of cancer. Another 2 patients in group 3 withdrew their consents and were excluded. Therefore, the study ended with 56 patients. No adverse drug reaction was reported with using DFO in the treatment groups. The study flowchart is available in the supplement file (Fig. 1).

The patients in three groups were statistically comparable in terms of age, sex and duration of treatment (Table 1).

Table 1
Demographic data of the study population

Variable	Group 1	Group 2	Group 3	P-value
Number (Male/Female)	21(16/ 5)	19(15/ 4)	16(11/5)	0.62
Age (y) (Mean ± SD)	6.8 ± 4.8	7.9 ± 4.7	8.6 ± 4.8	0.7
Body surface area (/m ²) (Mean ± SD)	0.93 ± 0.40	0.92 ± 0.35	1 ± 0.33	0.55
Malignancy (%) Leukemia Lymphoma Other tumors	11(52.4%) 7(33.3%) 3(14.3%)	9(47.4%) 8(42.1%) 2(10.5%)	8(50%) 6(37.5%) 2(12.5%)	0.57
Duration of treatment(Mo) (Mean ± SD)	14 ± 4	10 ± 3	12 ± 4	0.61
Doxorubicin cumulative dose (mg/m ²) (Mean ± SD)	225 ± 132	224 ± 116	218 ± 130	0.68
Group 1: control group; group 2: treated with Deferoxamine 10 times the Doxorubicin dose; group 3: treated with Deferoxamine 50 mg/kg				

Mean parameters of CBC and LFT, pre- and post-treatment with DFO were compared between the 3 groups, and the results were shown in Table 2.

Table 2

Complete blood count and liver function tests in the study population pre- and post-treatment with Deferoxamine*

Pre-treatment values	Group 1 (n = 20)	Group 2 (n = 19)	Group 3 (n = 16)	P ₁ value
WBC (10 ⁹ /L)	11.15 ± 15.98	8.09 ± 7.65	18.47 ± 23.60	0.17
Hemoglobin (g/dl)	10.12 ± 2.71	9.55 ± 1.34	9.61 ± 2.61	0.69
Platelet (10 ⁹ /L)	181.19 ± 128.28	203.79 ± 120.39	160.750 ± 152.78	0.63
Total protein (g/dl)	6.03 (5.12–8.01)	5.90 (4.25–7.30)	6.10 (5.05–8.90)	0.57
Albumin (g/dl)	4.0 (3.10–5.40)	3.90 (2.70-5.00)	3.75 (3.00-4.50)	0.41
Total bilirubin (mg/dl)	0.50 (0.40–1.50)	0.53 (0.25–2.20)	0.52 (0.23–2.40)	0.53
Direct bilirubin (mg/dl)	0.10 (0.01–0.40)	0.10 (0.10–0.30)	0.10 (0.10–0.90)	0.42
Aspartate aminotransferase (U/dl)	33.0 (15.0–78.0)	31.0 (11.0-286.0)	32.50 (11.0–73.0)	0.35
Alanine aminotransferase (U/dl)	27.0 (8.0-136.0)	19.0 (11.0-317.0)	20.50 (5.0-115.0)	0.47
Alkaline phosphatase (U/dl)	367.0 (230.0-760.0)	370.0 (210.0-2850.0)	335.0 (246.0-952.0)	0.62
Post-treatment values				
WBC (10 ⁹ /L)	4.48.±1.44	7.0 ± 6.78	8.73 ± 9.15	0.13
P ₂ value	0.07	0.68	0.10	
Hemoglobin (g/dl)	10.23 ± 1.62	9.84 ± 0.83	10.11 ± 1.12	0.60

Group 1: control group; group 2: treated with Deferoxamine 10 times the Doxorubicin dose; group 3: treated with Deferoxamine 50 mg/kg

*Data related to parameters with normal distribution are summarized as mean and standard deviation, and those related to non-normal distribution are shown as median and interquartile range. P₁ is related to the comparison of values between the 3 groups; P₂ is related to the comparison of pre- and post-treatment values in each group.

Pre-treatment values	Group 1 (n = 20)	Group 2 (n = 19)	Group 3 (n = 16)	P₁ value
P ₂ value	0.82	0.40	0.44	
Platelet (10 ⁹ /L)	161.08 ± 93.93	199.59 ± 85.08	198.12 ± 114.66	0.37
P ₂ value	0.28	0.88	0.31	
Total protein (g/dl)	6.0 (5.0-7.45)	5.80 (4.25–6.45)	5.87 (4.75-7.0)	0.52
P ₂ value	0.20	0.10	0.046	
Albumin (g/dl)	3.95 (3.05–4.50)	3.90 (2.90–4.35)	3.85 (2.75–4.25)	0.40
P ₂ value	0.56	0.80	0.85	
Total bilirubin (mg/dl)	0.5.0 (0.25–2.20)	0.55 (0.25–1.50)	0.57 (0.30–1.85)	0.35
P ₂ value	0.52	0.22	0.79	
Direct bilirubin (mg/dl)	0.10 (0.05–0.50)	0.11 (0.01–0.31)	0.15 (0.10–0.75)	0.65
P ₂ value	0.41	0.48	1.00	
Aspartate aminotransferase (U/dl)	32.50 (9.50–84.0)	29.50 (18.0-355.50)	26.25 (16.0-111.0)	0.47
P ₂ value	0.09	0.43	0.34	
Alanine aminotransferase (U/dl)	28.50 (14.0-106.50)	36.0 (23.0-698.50)	31.50 (18.0-225.0)	0.43
P ₂ value	0.42	0.03	0.03	
Alkaline phosphatase (U/dl)	324.0 (215.0-545.0)	380.0 (171.0-525.0)	331.25 (221.50–540.0)	0.57
P ₂ value	0.21	0.64	0.28	
Group 1: control group; group 2: treated with Deferoxamine 10 times the Doxorubicin dose; group 3: treated with Deferoxamine 50 mg/kg				

*Data related to parameters with normal distribution are summarized as mean and standard deviation, and those related to non-normal distribution are shown as median and interquartile range. P₁ is related to the comparison of values between the 3 groups; P₂ is related to the comparison of pre- and post-treatment values in each group.

The measured background parameters were initially comparable in the study groups. Similarly, no difference was observed following treatment between the study groups regarding these background characteristics (Table 3). When pre- and post-treatment values of background variables were compared in each group, only total protein in group 3 (6 mg/dl vs 5.87 mg/dl, $p = 0.047$) and ALT in group 2 (19 mg/dl vs 36 mg/dl), and group 3 (20.5 mg/dl vs 31.5 mg/dl) were significantly different ($p = 0.03$).

Table 3
Comparison of indirect serum markers of liver fibrosis in the study groups*

	Group 1 (n = 21)	Group 2 (n = 19)	Group 3 (n = 16)	P value
APRI	0.62(0.31–1.53)	0.52(0.27–1.09)	0.57(0.24–3.59)	0.87
Pre-treatment	0.66(0.33–0.89)	0.43(0.36–0.64)	0.38(0.25–0.66)	0.20
Post-treatment				
P_2 value**	0.06 [§]	0.22 [§]	0.015 [§]	
FIB-4 score [#]	0.27(0.09–0.71)	0.27(0.19–0.44)	0.38(0.21–1.68)	0.23
Pre-treatment	0.17(0.11–0.52)	0.20(0.13–0.34)	0.21(0.11–0.40)	0.99
Post-treatment				
P_2 value	0.725 [§]	0.191 [§]	0.020 [§]	
FibroTest	0.006 (0.003–0.02)	0.008 (0.004–0.02)	0.01 (0.005–0.02)	0.33
Group 1: control group; group 2: treated with Deferoxamine 10 times the Doxorubicin dose; group 3: treated with Deferoxamine 50 mg/kg; GT: gamma glutamyl transferase; α_2 MG: alpha2 macroglobulin; APOA1: apolipoprotein A1; APRI: AST-to-platelet ratio index				
*Data are shown as median and interquartile range				
**Alternative hypothesis: Pre-treatment APRI score -post-treatment APRI score > 0.5*pre-treatment APRI score				
[#] FIB-4 Scores were dichotomized as zero for scores equal or less than 1.45 and one for scores more than 1.45. [§] Exact one-sided p-value provided by McNemar's test				

Regarding indirect serum biomarkers of liver fibrosis, percentage of at-risk patients according to the FIB-4 score was significantly decreased in group 3 following treatment with DFO (McNemar's test $p = 0.02$). There was no statistically significance association between group and FIB-4 score after the study period ($p = 0.99$). In terms of change in the APRI score, although treatment in group 3 had a significant antifibrotic effect compared with patients' baseline APRI score (one-sided p value 0.015) (Table 3), no statistically independent effect was observed compared with control group (adjusted OR, 0.327; 95% CI: 0.028–3.78). Comparison of odds of an acceptable ant fibrotic effect of treatment in group 2 compared

with the control group was at 0.17 with a 95% CI of 0.03 to 0.84, adjusted for age, sex and patients' baseline APRI score.

The scores were not significantly associated with duration of treatment and DOX cumulative dose of the patients (data not shown). With regards to FibroTest, except one patient in group 3 who exceeded the cut-off value of 0.58, other patients were within the safe zone of mild or no fibrosis.

Figure 2 shows the comparison of Fibroscore among the 3 study groups following treatment with DFO. All patients had METAVIR scores in the F0-F1 zones with no significant difference between groups.

Discussion

This study was the first human clinical trial designed to investigate the protective role of DFO against DOX-induced liver injury in pediatric patients with cancer. We could show that DOX didn't have significant toxicity to the liver tissue at least in the short-term, though transient transaminases were seen in a subgroup of patients. There was no intergroup difference in the LFT parameters of patients treated with DFO and the group. Within each group, all parameters remained comparable except a small rise in ALT level in groups 2 & 3 and a minor drop in total protein in group 3, though within the normal range (Table 3). Due to lack of similar clinical trials in the past, we couldn't compare our results with them. However, in a few animal studies like Saad's study [20], they reported that DFO with the dosage of 15–20 times of DOX dose may decrease serum AST levels in rat models. This was not observed in our study.

We also checked some indirect serum markers of fibrosis, and found that APRI and FIB-4 score were significantly decreased in patients who were pre-treated with higher dose DFO (group 3, 50 mg/kg) (one-sided P values 0.015 and 0.02 respectively). However, statistical adjustment for age, sex and baseline APRI revealed that only treatment in group 2 is independently accompanied with 83% decrease in the risk of DOX-related liver fibrosis. The implication of these circulating factors to predict liver fibrosis is becoming more popular because they are available in many laboratories, they are almost non expensive, and can be repeated several times. Moreover, they may be representative of the fibrosis in the whole liver, thus avoiding small sampling error which is a technical defect in percutaneous liver biopsy [21].

The predictive value of indirect serum markers of liver fibrosis was previously mentioned in conditions such as viral hepatitis and fatty liver disease [1, 22]. Unalp-Ardia similarly reported that APRI can be used to predict liver fibrosis with higher scores indicative of advanced liver disease [23]. Moreover, it has been claimed that the combination of APRI with FibroMeter may show an accurate lower cost alternative to liver biopsy to evaluate fibrosis [24]. A meta-analysis showed that APRI > 1 had a sensitivity of 76% and a specificity of 72% to predict cirrhosis in patients infected with hepatitis C virus (HCV). For significant fibrosis, an APRI threshold of 0.7 was 77% sensitive and 72% specific. They concluded that APRI may obviate the need for staging liver biopsy in a subset of patients with HCV infection [14].

Moreover, FIB-4 index as another non-invasive serum marker to delineate liver fibrosis has been implicated in a variety of illnesses including hepatitis B virus (HBV) infection. Mallet investigated the

accuracy of FIB-4 index in a group of chronic HBV-infected patients and concluded that a cut-off value ≤ 1.45 can differentiate moderate fibrosis from severe fibrosis with a negative predictive value of 86%, a sensitivity of 71.1% and a specificity of 73.1%. He asserted that it is even more precise than APRI to exclude significant fibrosis [1]. Shah et al. reported that in patients with NAFLD, the FIB-4 index is superior to other non-invasive markers of fibrosis [22]. In addition, a systematic review and meta-analysis showed APRI and FIB-4 can identify hepatitis B-related fibrosis with a moderate sensitivity and accuracy. They suggested that an APRI threshold of 0.5 and 1.5 and an FIB4 threshold of 1.45 and 3.25 had acceptable sensitivity and specificity to delineate mild from significant fibrosis [17]. Though we showed that treatment with higher dose DOX (50 mg/kg) may be associated with significant decrease in FIB-4 score (Table 3), we were not able to show its independent association with FIB-4 score.

We also checked FibroTest which is a commercially available algorithm combining different elements, and has been proved to have high predictive value in advanced fibrosis. A cut-off value of 0.58 has been reported to associate with severe fibrosis ($F \geq 3$) [19, 25]. Only one patient who was treated with high-dose DFO (group 3) exceeded this cut-off point, and the results were comparable in the 3 groups.

The same finding was confirmed with TE that none of our patients experienced significant fibrosis following DOX treatment. They all had a METAVIR score in the F0-F1 zones, compatible with no or mild fibrosis. Meanwhile, we could show that DFO even further decreased serum markers of hepatic fibrosis when administered at a dose of 10 times DOX dose. It may be promising that DFO may play a role in decreasing the chance of liver fibrosis in the long-term. Despite normal LFT, METAVIR score, APRI, FIB-4 index, and FibroTest, nobody can guarantee that these patients are protected from liver fibrosis and cirrhosis when they reach their adulthood. Therefore, there is always a concern that survivors of pediatric malignancy may suffer from multi-organ damage in the future particularly if they are treated with toxic agents such as anthracyclines with proven cardiac and possibly hepatic and renal complications. It is also important to consider that non-invasive predictors of liver fibrosis such as serum markers and FibroScan are more reliable for detection of advanced fibrosis, and are less sensitive in the early stages of liver fibrosis ($F \leq 2$) [21]. Although none of our patients suffered from severe fibrosis as a result of chemotherapy effect, it is highly recommended to follow these children for at least a decade to investigate the long-term toxicity of chemotherapy agents on the liver and other organs.

Our study had some strengths and limitations. Regarding the strengths, it was the first randomized clinical trial in patients with cancer and especially in the pediatric age group which assessed the role of DFO as a rescue therapy to prevent liver damage induced by DOX. Up to now, only limited animal studies were conducted and this issue makes out our study as the first study of its own. Secondly, we tried to assess liver fibrosis with different modalities including FibroScan, LFT and non-invasive markers of liver fibrosis such as FibroTest, APRI and FIB-4 index. This probably helped us to increase the accuracy of our assessment.

On the other hand, the study faced some limitations. First of all, the small number of our cases and the short period of follow up hinder the generalizability of the results. A larger multi-center study with longer

follow up is required to assess the reproducibility of our results. Secondly, the heterogeneity of the study population in terms of their primary diagnosis is another issue that may have confounded the results as the chemotherapy protocols were not the same in all participants. Therefore, the interaction of other chemotherapy agents with DOX and their hepatic side effects cannot be overlooked. Lastly, we didn't have tissue biopsy to assess liver fibrosis in our patients because of ethical issues and the potential hazards of liver biopsy in cancer patients. Though FibroScan, and non-invasive serum markers of fibrosis are good alternative to liver biopsy, they are more informative in advanced fibrosis rather than early stages of liver fibrosis [26].

At the end, considering all these pros and cons of the study, our results may shed light for future researches to solve the dark sides of this puzzle. It is highly advised to run a large multi-center trial in a cohort of patients with similar malignancy such as leukemia to know whether DFO may prove beneficial in long-term protection of liver injury related to anthracyclines.

Conclusion

DOX did not lead to severe fibrosis in the liver provided that not exceeding the maximum allowed cumulative dose. DFO at the dose of 10 times of DOX dose may have a potential protective role against liver fibrosis. Larger multi-center studies are warranted to further assess this hypothesis.

Abbreviations

Doxorubicin (**DOX**)

Deferoxamine (**DFO**)

Liver function tests (**LFT**)

Hepatitis C virus (**HCV**)

γ -glutamyltransferase (**γ GT**)

α 2 -macroglobulin (**α 2 MG**)

Apo lipoprotein A1 (**apoA1**)

Declarations

Ethics approval and consent to participate

Written informed consent was obtained from all participants or their parents. The Ethics Committee of Shiraz University of Medical Sciences approved the study with the Ethical code IR.SUMS.REC.1394.193.

The trial was registered in the Iranian Registry of Clinical Trials with registration No. IRCT2016080315666N4.

Consent for publication

Not applicable.

Availability of data and materials

The data that support the findings of this study are available from Research Center of Shiraz University of Medical Science but restrictions apply to the availability of these data, which were used under license for the current study, and so are not publicly available. Data are however available from the authors upon reasonable request and with permission of Research Center of Shiraz University of Medical Science

Competing interests

All authors declare that they have no conflict of interest.

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Not applicable.

Authors' contributions

Study conception and design were done by MB, NS, MF

SH, NS, MS and analyzed and interpreted the data.

Acquisition of data was done by MF, H MV, MB, NS and GF

GF, MB and MD drafted the manuscript.

The critical revision was done by all authors.

All authors reviewed and approved the final manuscript.

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Figures

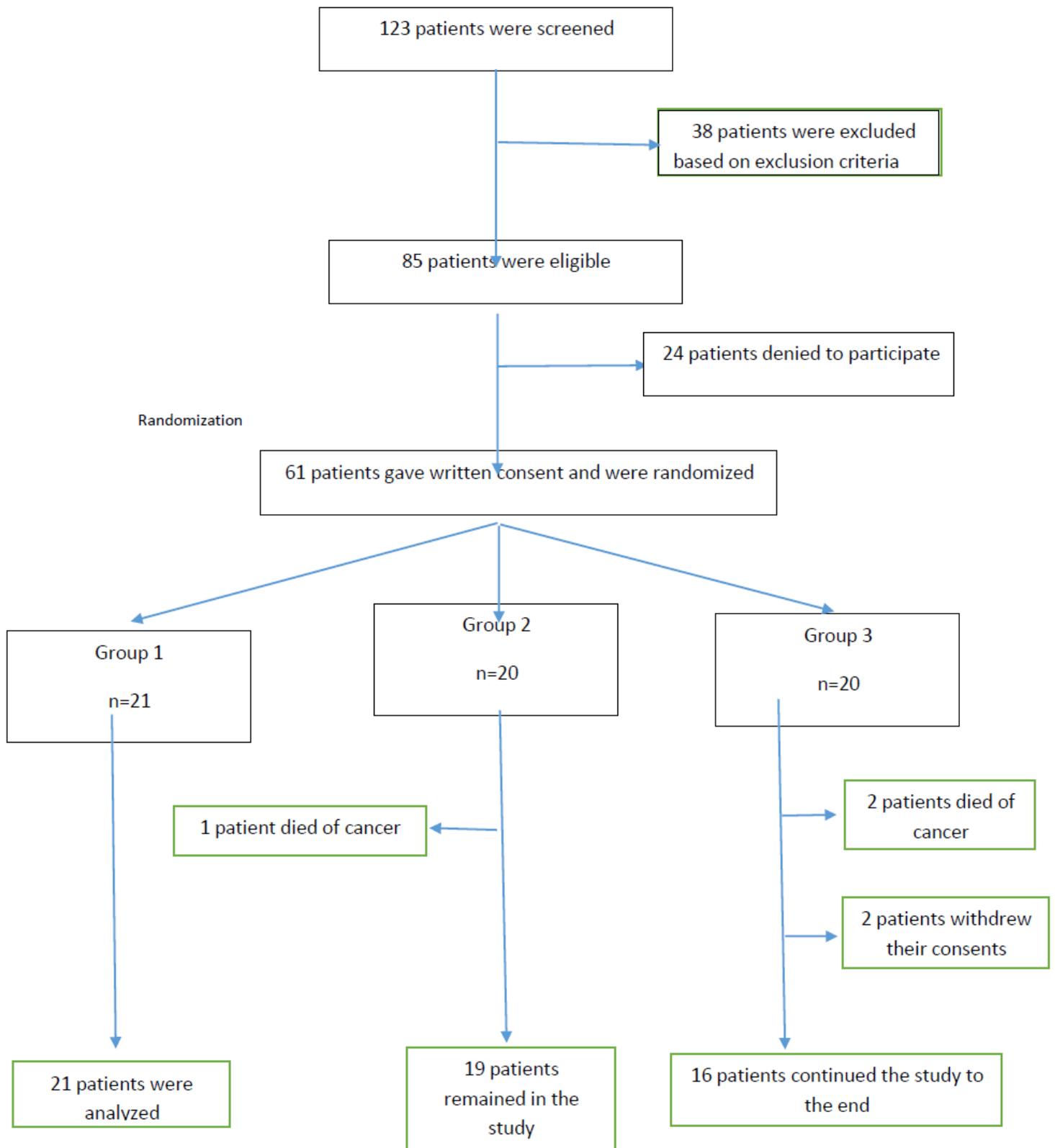


Figure 1

Flowchart of the study population screening and their randomization

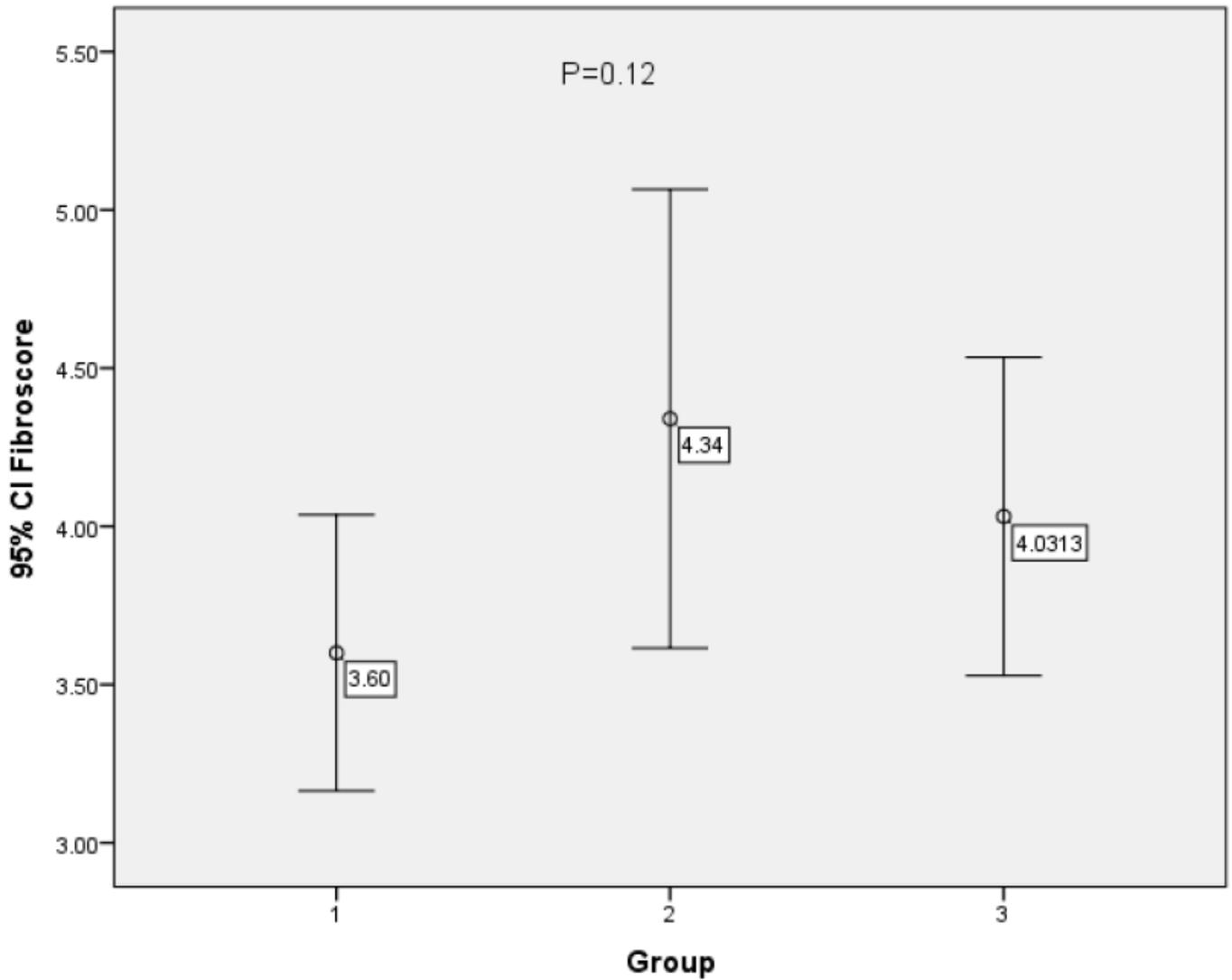


Figure 2

Assessment of liver stiffness by FibroScan in the 3 groups post-Deferoxamine treatment

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