

The Fatty Acid–Bile Acid Conjugate Aramchol Reduces Liver Fat Content in Patients With Nonalcoholic Fatty Liver Disease

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BACKGROUND & AIMS: We investigated the effects of the fatty acid–bile acid conjugate 3 β -arachidyl-amido, 7 α -12 α -dihydroxy, 5 β -cholan-24-oic acid (Aramchol; Trima Israel Pharmaceutical Products Ltd, Maa-barot, Israel) in a phase 2 trial of patients with nonalcoholic fatty liver disease (NAFLD).

METHODS: We performed a randomized, double-blind, placebo-controlled trial of 60 patients with biopsy-confirmed NAFLD (6 with nonalcoholic steatohepatitis) at 10 centers in Israel. Patients were given Aramchol (100 or 300 mg) or placebo once daily for 3 months (n = 20/group). The main end point was the difference between groups in the change in liver fat content according to magnetic resonance spectroscopy. The secondary end points focused on the differences between groups in alterations of liver enzyme levels, levels of adiponectin, homeostasis model assessment scores, and endothelial function.

RESULTS: No serious or drug-related adverse events were observed in the 58 patients who completed the study. Over 3 months, liver fat content decreased by 12.57% \pm 22.14% in patients given 300 mg/day Aramchol, but increased by 6.39% \pm 36.27% in the placebo group ($P = .02$ for the difference between groups, adjusted for age, sex, and body mass index). Liver fat content decreased in the 100-mg Aramchol group, by 2.89% \pm 28.22%, but this change was nonsignificant ($P = .35$), indicating a dose–response relationship (P for trend = .01). Groups given Aramchol had nonsignificant improvements over time in endothelial function and levels of alanine aminotransferase and adiponectin, but homeostasis model assessment scores did not change. The appropriateness of a single daily dose was confirmed by pharmacokinetic analysis.

CONCLUSIONS: Three months' administration of the fatty acid–bile acid conjugate Aramchol is safe, tolerable, and significantly reduces liver fat content in patients with NAFLD. The reduction in liver fat content occurred in a dose-dependent manner and was associated with a trend of metabolic improvements, indicating that Aramchol might be used for the treatment of fatty liver disease. ClinicalTrials.gov number: NCT01094158.

Keywords: Clinical Trial; Lipid; Steatosis; Cholic Acid.

Nonalcoholic fatty liver disease (NAFLD) and nonalcoholic steatohepatitis (NASH) are increasingly relevant public health issues because of their close association with the worldwide epidemics of diabetes and obesity.¹ NAFLD/NASH is associated with liver-related and cardiovascular morbidity and mortality, and therefore is a burden on health expenditure in Western countries. In the majority of patients, NAFLD/NASH is associated with additional features of the metabolic syndrome.² Histologically, NAFLD is characterized by the presence of fat infiltration of the liver, expressed by ballooning of hepatocytes. In approximately 20% of cases it is accompanied by inflammation and hepatocyte

injury, defined as NASH, and liver fibrosis progression.² Despite the high prevalence of NAFLD, no safe and effective treatment currently is available.³ Adherence to life-change modifications is difficult to maintain.⁴ Bariatric surgery can be performed in selective obese patients,

Abbreviations used in this paper: ANOVA, analysis of variance; BMI, body mass index; FMD, flow-mediated dilatation; LFC, liver fat content; MRS, magnetic resonance spectroscopy; NAFLD, nonalcoholic fatty liver disease; NASH, nonalcoholic steatohepatitis; PK, pharmacokinetic; SCD1, stearoyl coenzyme A desaturase 1.

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but is not recommended for treating NASH.⁵ Vitamin E was found to be efficient for nondiabetic NASH patients without cirrhosis.⁶ The long-term safety and efficacy of pioglitazone are not clear in NASH.^{7,8} Metformin,⁹ ursodeoxycholic acid, omega 3 fatty acids, and statins are not considered as therapy for NAFLD/NASH patients.^{2,8}

Aramchol (3 β -arachidyl-amido, 7 α -12 α -dihydroxy, 5 β -cholan-24-oic acid; Trima Israel Pharmaceutical Products Ltd, Maabarot, Israel) is a novel synthetic lipid molecule obtained by conjugating 2 natural components, cholic acid (bile acid) and arachidic acid (saturated fatty acid), through a stable amide bond. Aramchol significantly reduces hepatic fat content in animals with a high-fat diet model.¹⁰ In *in vitro* models, Aramchol achieves 70% to 83% inhibition of the stearoyl coenzyme A desaturase 1 (SCD1) activity. SCD1 is a key enzyme that modulates fatty acid metabolism in the liver. SCD1 inhibition decreases the synthesis and increases β oxidation of fatty acids, resulting in decreased hepatic storage of triglycerides and fatty acid esters.^{11,12}

This process reduces liver fat and improves insulin resistance in animals.¹³ In addition, Aramchol activates cholesterol efflux by stimulating the adenosine triphosphate-binding cassette transporter A1, a pan-cellular cholesterol export pump,^{10,14} and has shown to have an anti-atherogenic effect in animal studies.¹⁵ In pre-clinical studies, Aramchol in high doses did not cause the severe adverse effects attributed to complete inhibition of SCD1 (skin and eye disturbances, atherosclerosis, and inflammation).^{16,17}

In short-term clinical studies, no adverse events were observed in 41 healthy volunteers, testing single Aramchol doses up to 900 mg and repeated doses up to 300 mg. We hypothesize that the favorable safety profile is attributable to Aramchol's partial inhibition of SCD1 activity and its additional effect on the up-regulation of adenosine triphosphate-binding cassette transporter A1. The current study evaluated Aramchol's effect on liver fat content (LFC) and metabolic parameters in NAFLD patients.

Patients and Methods

Study Design

Our study was a phase 2, multicenter, randomized, double-blind, and placebo-controlled trial for 3 months in 10 Israeli medical centers. It aimed to evaluate the safety, pharmacokinetics (PKs), and effect of 2 Aramchol doses on LFC in 60 subjects with NAFLD and NASH. This study was designed and conducted according to the Good Clinical Practice guidelines and following the protocol approval by each Institutional Review Board and by the Israeli Ministry of Health. Written informed consent was obtained from all patients before any study-specific medical procedures were performed. The trial is registered at <http://clinicaltrials.gov> (NCT01094158) and was conducted from December 2010 to December 2011.

The baseline evaluation included detailed medical history and physical examination, height and weight measurements for body mass index (BMI) calculation, and blood analyses. Eligible patients were randomized within 14 days of recruitment to receive a once-daily oral dose of high-dose Aramchol (300 mg), low-dose Aramchol (100 mg), or placebo. The medication was administered in the morning within 10 minutes after breakfast for 12 weeks. Randomization was performed in 12 balanced blocks of 6 subjects each, with 2 subjects per treatment group in each block.

Subjects attended the clinic every 2 weeks for the first 2 visits followed by monthly visits for a total of 3 months. A follow-up visit was performed 1 month after treatment completion for safety and liver enzyme evaluation. At each study visit, routine blood tests were obtained, body weight and a physical examination were recorded, and the number of pills was counted to document compliance. Liver-related symptoms as well as possible side effects of Aramchol were investigated.

Eligibility

This study was conducted in men and women aged 18 to 75 years with proven histology of NAFLD or NASH. A percutaneous liver biopsy used for the diagnosis of NAFLD or NASH was mandatory within 18 months before the start of the study. All biopsy specimens were assessed by a single hepatopathologist blinded to the clinical and radiologic data, to randomization, and to previous histologic assessments.

A NAFLD activity score, representing the sum of scores for steatosis, lobular inflammation, and ballooning (range, 0–8), was used to differentiate between NAFLD and NASH. Subjects with a NAFLD activity score of 0 to 2 were considered to have NAFLD. An activity score of 3 or more was considered NASH. Fibrosis was staged as follows: 0, none; 1, perisinusoidal or periportal; 2, perisinus and periportal; 3, bridging fibrosis; and 4, cirrhosis.

Efficacy Evaluation

The primary efficacy end point was the difference in LFC, measured by magnetic resonance spectroscopy (MRS), between baseline and the end of treatment. The secondary efficacy end points were the differences between baseline and the end of treatment in serum alanine aminotransferase levels, endothelial function, measured by flow-mediated dilatation (FMD), insulin resistance, measured by homeostasis model assessment, adiponectin levels, hemoglobin A1C levels (HbA1C), weight, and metabolic parameters such as serum cholesterol and triglycerides.

Magnetic Resonance Spectroscopy

Baseline and post-treatment MRS were performed to assess hepatic steatosis. Each MRS was performed using

point-resolved spectroscopy single voxel at the same Medical Center on a GE 3T SignaHDxt scanner (Signa Excite; GE, Milwaukee, WI) to minimize the impact of scanner platform, field strength, and other confounders that frequently corrupt fat content estimations.^{18–20} Quantitative analysis of fat was performed using LC Model software (version 6.3-0L; available at: <http://www.s-provencher.com/pages/lcmodel.shtml>). The change from baseline in LFC was calculated for each subject. The mean change per group was calculated based on the individual changes. The primary end point was the difference between the study groups in the mean LFC changes.

Flow-Mediated Dilatation

A decrease in endothelial function, as observed in NASH patients, plays a key role early in the development of atherosclerosis and is an independent predictor of cardiovascular risk.^{21–23} Endothelial function was assessed at baseline and at the study end by FMD. Brachial artery FMD was measured by induction of reactive hyperemia using the temporary arterial occlusion method. The resultant relative increase in blood vessel diameter then was assessed via ultrasound.

Safety Evaluation

Safety evaluation included medical history, complete physical examination and vital signs, clinical laboratory evaluations (hematology, biochemistry), and electrocardiography. Adverse events were summarized and were stratified by severity, outcome, and relation to study drug.

Pharmacokinetic Analysis

PK assessment was performed on a third of the studied population. Blood was collected at 3, 6, 12, and 24 hours after administration of the first dose of Aramchol and at time 0, 3, 6, 12, 24, and 72 hours after administration of the last dose. Aramchol blood levels were measured by liquid chromatography mass spectrometry at Analyst Laboratories (Rehovot, Israel).

PK analysis was performed using appropriate non-compartmental methods. Standard PK parameters (maximum plasma concentration; time to reach maximum plasma concentration; the area under the concentration vs time curve, calculated as the sum of areas under the curve from time 0 to the last measurable data point; the area under the concentration vs time curve, calculated as the sum of areas under the curve from time 0 to infinity; and elimination half-life) were calculated.

Statistical Analysis

A sample size of 20 subjects per group provided a power of 90% to identify a difference of at least 5% in the triglyceride concentration decrease between the placebo

group and at least one of the treatment groups, with a standard deviation of 5%. The sample size was considered appropriate for the study: taking into consideration possible drop-outs, a sample size of 17 patients in each group still would have 80% power to detect a difference in means of 5.0% reduction, assuming a standard deviation of 5.0, with a 2-sided significance level of 5%.^{24,25}

Baseline parameters were compared between groups using chi-squared test for sex and the number of patients with NASH, and for all other variables using analysis of variance (ANOVA). Efficacy parameters were analyzed using ANOVA and analysis of covariance models. A paired *t* test was applied for testing changes within the study groups in continuous parameters. The ANOVA model using the Dunnett method was applied for testing differences in measurements and changes from baseline between the study groups. The analysis of covariance model was applied for testing the differences between the study groups in change of LFC and secondary outcomes, adjusted for baseline measurement suspected to affect LFC. All tests were 2-tailed, and a *P* value of 5% or less was considered statistically significant. The data were analyzed using SAS version 9.1 (SAS Institute, Cary, NC).

All authors had access to the study data, and reviewed and approved the final manuscript.

Results

Patient Characteristics

Sixty patients with biopsy-proven NAFLD (*n* = 54) and NASH (*n* = 6) were randomized. Two patients from the Aramchol 100-mg group withdrew consent, resulting in a sample size of 58 patients completing the study (Supplementary Figure 1). Baseline parameters were similar in all 3 treatment groups (Table 1).

At baseline, grade 1 fibrosis was observed in 4 patients (20%) in the high-dose group, and in 5 patients (25.0%) each in the low-dose group and in the placebo group. Grade 2 fibrosis was observed in 2 patients (10.0%) in the low-dose group and in 3 patients (15.0%) in the placebo group, and grade 3 fibrosis was observed in 2 patients (10.0%) each in the low-dose group and in the placebo group. None of the patients in the high-dose Aramchol group had grade 2 or 3 fibrosis.

Efficacy

A significant dose-response relationship was observed in the relative change in LFC among the 3 groups (*P* for trend by linear regression .01, adjusted for age, sex, and BMI) (Figure 1A). Patients treated with high-dose Aramchol had a significant reduction of $12.57\% \pm 22.14$ in LFC compared with a $6.39\% \pm 36.27$ increase in the placebo group (*P* = .020). Individual patient data on changes in LFC stratified by treatment group are shown in Figure 1B, confirming a different

Table 1. Baseline Characteristics of the Study Population

Characteristic (normal range)	Aramchol 300 mg (n = 20)	Aramchol 100 mg (n = 18)	Placebo ^a (n = 19)	P value ^b
Age, y	38.4 ± 14.6	39.7 ± 11.7	42 ± 11.2	.664
Males, n (%)	14 (70)	13 (72.2)	14 (73.7)	.967
Weight, kg	84.5 ± 16.2	86.7 ± 10.0	83.7 ± 11.4	.768
BMI, kg/m ²	29.1 ± 3.4	29.1 ± 3	28.2 ± 3.5	.646
MRS, g TG/100 g liver	0.27 ± 0.07	0.25 ± 0.07	0.22 ± 0.08	.144
NASH, n (%)	2 (10)	2 (11.1)	2 (10.5)	1.0
Endothelial function, %	4.6 ± 2.5	5.4 ± 3.0	6.4 ± 3.0	.213
ALT level, U/L (10–41)	78.3 ± 41.6	74.4 ± 43.6	63.1 ± 31.2	.459
AST level, U/L (15–40)	47.8 ± 33.8	40.2 ± 15	38.8 ± 16.4	.451
Alkaline phosphatase level, U/L (39–117)	75.7 ± 17.8	72.1 ± 25.6	73.5 ± 24.5	.889
Glucose level, mg/dL (70–105)	95.6 ± 12.9	99.1 ± 16.2	96.1 ± 10.9	.689
Cholesterol level, mg/dL (70–239)	193.6 ± 43.5	178.7 ± 49	187 ± 40.7	.590
LDL, mg/dL	122.3 ± 33.6	114.4 ± 42.3	116.2 ± 34.4	.786
HDL, mg/dL	43.5 ± 19.8	40.5 ± 7.9	41.6 ± 7.6	.776
TG, mg/dL (50–199)	142.4 ± 61.4	122 ± 59.3	148.4 ± 115	.60
Insulin level, μU/mL (5–25)	18.2 ± 11.3	14.6 ± 6.3	18.3 ± 9.8	.404
HOMA score	4.4 ± 3.7	3.7 ± 2.1	4.4 ± 2.7	.700
HbA1C, %	5.5 ± 0.3	5.6 ± 0.4	5.4 ± 0.5	.498
Adiponectin level, μg/mL	3.8 ± 2.8	3.9 ± 2.1	4.2 ± 2.9	.896
Albumin level, g/dL (3.5–5.1)	4.9 ± 0.3	4.7 ± 0.2	4.7 ± 0.4	.098

NOTE. Continuous data are presented as the mean ± standard deviation. Categorical data are presented as n (%).

ALT, alanine aminotransferase; AST, aspartate aminotransferase; HbA1C, hemoglobin A1C; HDL, high-density lipoprotein; HOMA, homeostasis model assessment; LDL, low-density lipoprotein; TG, triglyceride.

^aOne patient who completed the study was not included in the analysis due to major protocol deviations.

^bP values for sex and the number of patients with NASH were determined by chi-squared test; P values for all other variables were determined by ANOVA.

pattern of relative change in LFC, predominantly a decrease in the high-dose Aramchol group.

Differences between the 3 groups were analyzed for secondary end points with adjustment for age, sex, and baseline BMI. There were no statistically significant differences among the 3 treatment groups for any of the secondary end points (Table 2). There was a nonsignificant trend of mild weight reduction ($P = .1$) in the high-dose Aramchol group (Table 2). Serum adiponectin levels increased ($0.2 \pm 1.7 \mu\text{g/mL}$) in the high-dose Aramchol group but decreased in the low-dose ($-0.3 \pm 1.5 \mu\text{g/mL}$) and placebo groups ($-0.7 \pm 1.3 \mu\text{g/mL}$) ($P = .088$

for trend of dose-response relationship by linear regression) (Figure 2). FMD increased nonsignificantly by $1.28\% \pm 2.92\%$ in the high-dose group, by $0.34\% \pm 3.54\%$ in the low-dose group, and by $0.46\% \pm 2.28\%$ in the placebo group.

Safety

The frequency of adverse events was similar in all treatment groups, but none of them were considered to be related to the treatment. All adverse events were mild

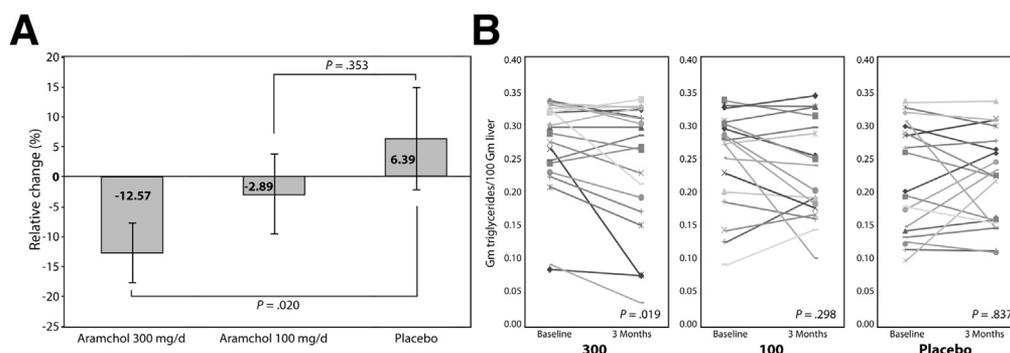


Figure 1. Relative change in MRS from baseline after 12 weeks of treatment. (A) The relative change (percentage) ± standard error of the mean LFC between baseline and end of treatment (measured by MRS). P value was determined by analysis of covariance using the Dunnett method for the difference between the Aramchol groups and placebo adjusted for age, sex, and baseline BMI. (B) Individual patient absolute changes in LFC (measured by MRS, triglycerides [in grams]/100 g liver) between baseline and 12 weeks of treatment stratified by the treatment group. P values refer to within-group comparisons of absolute change from baseline, after 12 weeks of treatment.

Table 2. Secondary Outcomes Change From Baseline, After 12 Weeks of Treatment

Variable	Aramchol 300 mg (n = 20)	Aramchol 100 mg (n = 18)	Placebo (n = 19) ^b	P value ^a	
				300 mg vs placebo	100 mg vs placebo
FMD	1.28 ± 2.92	0.34 ± 3.54	0.46 ± 2.28	.726	.987
ALT level, U/L	-8.6 ± 26.2	-7.8 ± 30.5	-10.4 ± 22.2	.926	.995
AST level, U/L	-5.6 ± 20.98	-1.56 ± 13.55	-5.83 ± 11.51	.923	.820
Alkaline phosphatase level, U/L	-2.7 ± 12.56	-2.06 ± 7.34	1.67 ± 6.16	.109	.218
Glucose level, mg/dL	0.1 ± 12.03	-0.72 ± 15.99	-1.06 ± 9.77	.992	.999
Cholesterol, mg/dL	6.6 ± 31.4	7.2 ± 26	-0.5 ± 24.3	.652	.631
LDL, mg/dL	7.3 ± 34.9	-1.4 ± 27	2 ± 22.7	.780	.898
HDL, mg/dL	0.7 ± 7	2.5 ± 6.5	-0.9 ± 3.7	.852	.257
TG, mg/dL	-8.1 ± 73	28.9 ± 66.5	-7.6 ± 67	.995	.186
Insulin, μ U/mL	-2.83 ± 12.32	4.95 ± 11.70	-0.24 ± 4.73	.612	.292
HOMA score	-0.80 ± 3.81	1.13 ± 3.42	-0.18 ± 1.43	.733	.395
Hemoglobin A1C, %	0.12 ± 0.34	0.02 ± 0.29	0.11 ± 0.34	.921	.430
Adiponectin, μ g/mL	0.2 ± 1.7	-0.3 ± 1.5	-0.7 ± 1.3	.163	.714
Weight, kg	-1.15 ± 2.25	0.08 ± 2.41	0.21 ± 2.29	.185	.990

ALT, alanine aminotransferase; AST, aspartate aminotransferase; HDL, high-density lipoprotein; HOMA, homeostasis model assessment; LDL, low-density lipoprotein; TG, triglyceride.

^aP values were calculated with ANCOVA using the Dunnett method adjusted for age, sex, and BMI.

^bOne patient who completed the study was not included in the analysis due to major protocol deviations.

or moderate and none were serious (Table 3). None of the patients withdrew as a result of adverse events.

Pharmacokinetics

PK analysis of both Aramchol doses was performed on day 1 upon initiation of treatment as well as at the end of treatment at week 13 (Figure 3).

Serum levels of the high-dose Aramchol were almost twice the levels of the low-dose Aramchol. The predose (trough) Aramchol serum levels of both doses remained stable for the duration of treatment.

Discussion

In this randomized, double-blind, placebo-controlled clinical trial, Aramchol was found to be safe and effective

in reducing LFC, as measured by MRS, in NAFLD patients after 12 weeks of daily administration of 300 mg. Because of the short therapeutic protocol, it is premature to assess the clinical significance of the effect of Aramchol on liver fat reduction. However, liver fat reduction alone has been shown to reverse NASH in studies with effective weight reduction regimens²⁶ and in patients undergoing bariatric surgery.^{25,27} A regression in NASH has been shown to accompany the decrease in LFC in patients treated with thiazolidinedione, despite the fact that the treatment did not result in a reduction in body weight.²⁴

A strong association exists between NAFLD and obesity, reduced glucose tolerance, type 2 diabetes mellitus, arterial hypertension, and hypertriglyceridemia. NAFLD/NASH is not only considered the hepatic manifestation of the metabolic syndrome, but also an

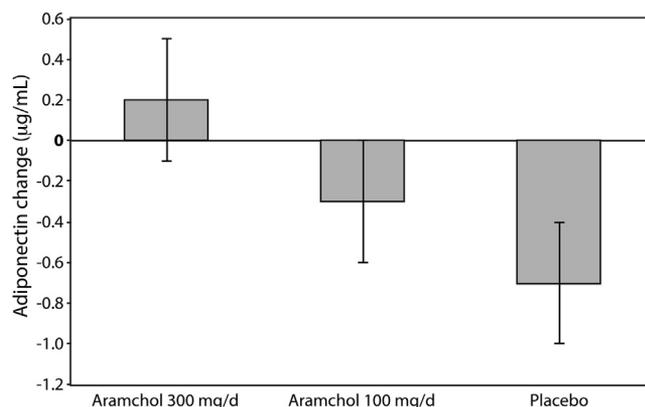


Figure 2. Change in serum adiponectin levels from baseline in each visit during 12 weeks of treatment: P value = .088 for the trend of dose-response relationship by linear regression adjusted for age, sex, and baseline BMI.

Table 3. Adverse Events: the Overall and the Most Frequent Events (>2 Patients in Any Group)

	Aramchol 300 mg (n = 20), n (%)	Aramchol 100 mg (n = 20), n (%)	Placebo (n = 20), n (%)
Patients with adverse events	9 (45.0)	8 (40.0)	11 (55.0)
Preferred term			
Abdominal pain	1 (5.0)	1 (5.0)	2 (10.0)
Abdominal upper pain	-	2 (10.0)	1 (5.0)
Back pain	-	-	3 (15.0)
Constipation	-	-	2 (10.0)
Asthenia	-	-	2 (10.0)
Upper respiratory tract infection	2 (10.0)	-	-

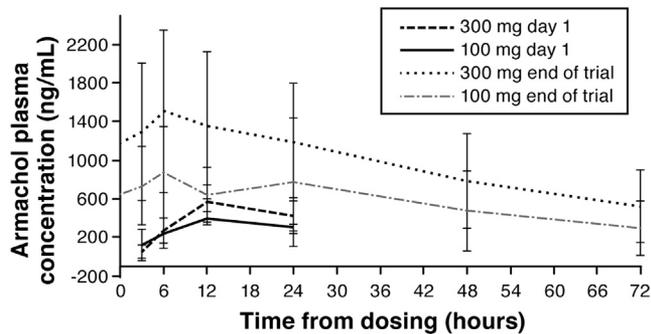


Figure 3. Average Aramchol plasma concentration vs time profiles. PK analysis was performed in a third of the studied population assigned by a centralized blinded randomization as described in the [Patients and Methods](#) section.

aggravating factor for the insulin resistance that is its key pathophysiological hallmark.^{28–30}

Hyperinsulinemia, caused by insulin resistance, results in an increased hepatic de novo lipogenesis and impaired inhibition of adipose tissue lipolysis, which ultimately leads to an increased efflux of free fatty acids from the adipose tissue to the liver.³¹ After the initial fat infiltration, the liver becomes vulnerable to a series of hits, leading to hepatocyte injury and finally progressing from steatosis to NASH and fibrosis. Such multiple pathogenetic factors include oxidative damage, dysregulated hepatocyte apoptosis, activation of the profibrogenic transforming growth factor β pathway, dysregulation of adipokines, and hepatic stellate cell activation.^{28,29}

Our study measured several indicators of the metabolic syndrome. No statistically significant differences were found between either of the 2 Aramchol treatment groups and the placebo group for any of them ([Table 2](#)), but an increase in adiponectin levels and a trend of improvement in endothelial function were observed in the high-dose treatment arm. Adiponectin has anti-oxidant, anti-inflammatory, and antifibrotic effects on the liver and improves insulin sensitivity through peroxisome proliferator-activated receptor activation.^{32,33} Hence, an adiponectin increase might mediate the beneficial Aramchol mechanism.

Adiponectin levels and endothelial function are decreased in NAFLD and NASH patients and the observed trends of improvement after Aramchol treatment point at possible positive metabolic effects of the compound, in addition to its effect of LFC reduction.

Therefore, longer-term studies, with histologic and metabolic end points, are warranted to assess Aramchol's effect on NASH and its metabolic complications.

PK studies confirmed dose-proportional increases of Aramchol in plasma levels with constant steady-state levels throughout the study period and a long elimination half-life, confirming suitability for once-daily administration.

However, some trial limitations might be addressed in the next planned phases, mainly the study population.

The current study included patients with mild NAFLD/NASH disease. This might constitute a suboptimal target population to test reduction of LFC and a significant effect on other metabolic derangements. Moreover, a repeated liver biopsy within the short duration of the study was considered unwarranted and thus no histologic outcomes, including the important effect on fibrosis, can be discussed in this trial.

In conclusion, Aramchol at a dose of 300 mg was found to be safe and effective in reducing LFC in NAFLD and NASH patients and inducing trends of improvement in metabolic parameters. This makes Aramchol a candidate for the treatment of fatty liver-related diseases, currently an unmet medical need. Longer Aramchol trials in patients with NASH and metabolic complications are warranted for the study of metabolic and histologic benefits.

Supplementary Material

Note: To access the supplementary material accompanying this article, visit the online version of *Clinical Gastroenterology and Hepatology* at www.cghjournal.org, and at <http://dx.doi.org/10.1016/j.cgh.2014.04.038>.

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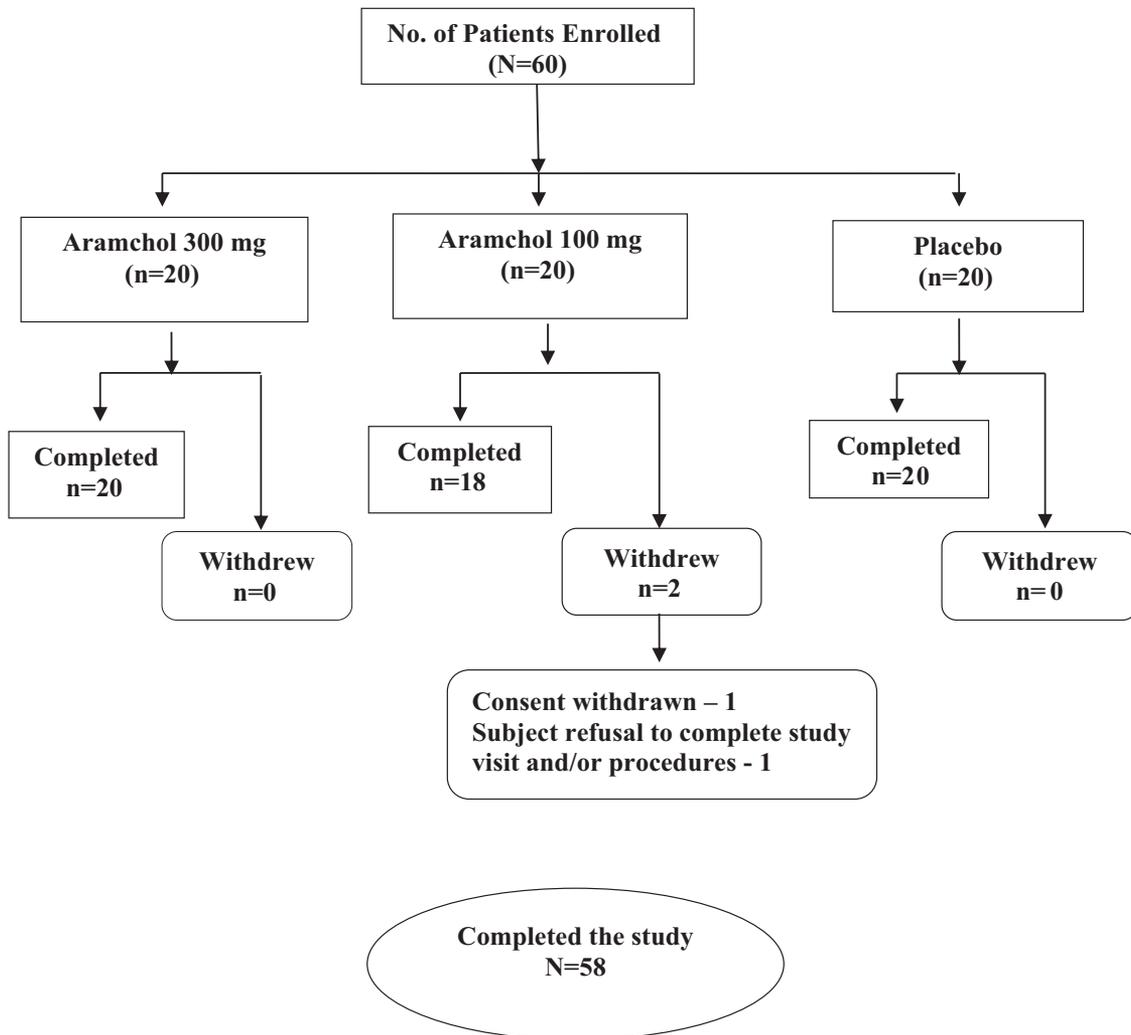
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Conflicts of interest

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Supplementary Figure 1. Consort diagram.