

Effect of lifestyle changes and atorvastatin administration on dyslipidemia in hemodialysis patients: a prospective study

Alicja E. Grzegorzewska¹, Leszek Niepolski², Jan Sikora³,
Małgorzata Janków⁴, Paweł P. Jagodziński⁵, Anna Sowińska⁶

¹ Department of Nephrology, Transplantology and Internal Diseases, Poznan University of Medical Sciences, Poznań, Poland

² B. Braun Avitum Dialysis Center, Nowy Tomyśl, Poland

³ Immunology Unit, Department of Clinical Immunology, Poznan University of Medical Sciences, Poznań, Poland

⁴ Eurodial Dialysis Center, Ostrów Wielkopolski, Poland

⁵ Department of Biochemistry and Molecular Biology, Poznan University of Medical Sciences, Poznań, Poland

⁶ Department of Computer Science and Statistics, Poznan University of Medical Sciences, Poznań, Poland

KEY WORDS

atorvastatin, CD36 expression, hemodialysis, lifestyle changes, salusin α

ABSTRACT

INTRODUCTION Atherogenic dyslipidemia accelerates the development of cardiovascular complications and contributes to mortality of hemodialysis (HD) patients.

OBJECTIVES The aim of the study was to evaluate the effects of lifestyle changes followed by treatment with atorvastatin in dyslipidemic HD patients.

PATIENTS AND METHODS Dyslipidemic HD patients ($n = 49$) were enrolled into the prospective study. Forty-two patients completed a 21-week lifestyle intervention. In 34 patients, who continued to be dyslipidemic, atorvastatin was used for 14 weeks. After 4 weeks, the initial dose of atorvastatin of 10 mg/d was increased to 20 mg/d in dyslipidemic patients.

RESULTS The most pronounced effects of lifestyle changes were shown at 14 weeks and included significant differences in high-density lipoprotein cholesterol, low-density lipoprotein (LDL) cholesterol, salusin α , malondialdehyde-oxidized LDL, fructosamine, and monocyte CD36 expression. Immunoglobulin G anti-oxLDL showed the highest values at 21 weeks. Seven patients (16.7%) were nondyslipidemic at 21 weeks. In patients who continued to be dyslipidemic, LDL cholesterol and triglyceride levels significantly decreased, salusin α levels and CD36 expression increased, and dyslipidemia resolved in 59.4% of the patients following atorvastatin treatment.

CONCLUSIONS Lifestyle changes have selective efficacy in the treatment of dyslipidemia in HD patients, while atorvastatin (up to 20 mg/d) may be effective in about 60% of nonresponders to lifestyle changes. Lipid-lowering interventions affect plasma salusin α and monocyte CD36 expression.

INTRODUCTION Atherogenic dyslipidemia accelerates cardiovascular complications related to the development of atherosclerosis.¹ In dyslipidemic patients with chronic kidney disease (CKD), including patients on hemodialysis (HD), lifestyle changes or lipid-lowering medications may affect the levels of low-density lipoprotein (LDL) cholesterol, high-density lipoprotein (HDL) cholesterol, non-HDL cholesterol, or triglycerides (TG).²⁻⁴ However, less recognized factors such as salusin α , cluster of differentiation

(CD) 36, oxidized LDL (oxLDL), and autoantibodies to oxidation-specific epitopes of LDL (anti-oxLDL), may also be involved in the pathogenesis of atherosclerosis, and the effect of lipid-lowering management on their levels is still largely unknown. In recent studies,^{5,6} salusin α has been reported as an endogenous inhibitor of atherosclerosis. The role of CD36 in lipid homeostasis as a scavenger receptor for LDL⁷ and transporter for long-chain fatty acids⁸ has been established. CD36 also mediates the cell uptake

Correspondence to:

Prof. Alicja E. Grzegorzewska,
MD, PhD, Katedra i Klinika
Nefrologii, Transplantologii
i Chorób Wewnętrznych,
Uniwersytet Medyczny
im. Karola Marcinkowskiego,
al. Przybyszewskiego 49,
60-355 Poznań, Poland, phone:
+48-61-869-17-00, fax:
+48-61-869-16-88, e-mail:
alicia_grzegorzewska@yahoo.com
Received: May 13, 2014.
Revision accepted: July 14, 2014.
Published online: July 21, 2014.
Conflict of interest: none declared.
Pol Arch Med Wewn. 2014;
124 (9): 443-451
Copyright by Medycyna Praktyczna,
Kraków 2014

of HDL.^{9,10} oxLDL is a stimulant of CD36 expression, and CD36 binds and internalizes oxLDL.^{11,12} The presence of oxLDL has been demonstrated in atherosclerotic lesions of humans and animals.¹³ High CD36 expression on macrophages, indicating intensive uptake of oxLDL, has been attributed to foam cell formation and atherosclerosis.^{14,15} Anti-oxLDL against malondialdehyde (MDA)-modified LDL has been shown to block the uptake of oxLDL by macrophages¹⁶ and to reduce the progression of atherosclerosis.¹⁷

In this prospective study of dyslipidemic HD patients, we aimed to show to what extent lifestyle changes followed by treatment with atorvastatin are effective in the treatment of dyslipidemia and affect laboratory markers related to lipid and carbohydrate metabolism.

PATIENTS AND METHODS **Subjects** The study design was approved by the Institutional Review Board of the Poznań University of Medical Sciences, Poland, and is available at ClinicalTrials.gov (ClinicalTrials.gov NCT01 448 174). Written informed consent to participate in the study was obtained from all patients.

The inclusion criteria for HD patients were as follows: age ≥ 18 years, HD vintage ≥ 3 months, and presence of dyslipidemia diagnosed according to the recommendations of the National Kidney Foundation / Kidney Disease Outcomes Quality Initiative (KDOQI) clinical practice guidelines.² Patients were excluded if they were treated with statins, fibrates, or other medications that could affect lipid metabolism during the 6 months prior to the study; if they had active thyroid gland disease and/or used thyreotropic medications; if they were treated with corticosteroids, immunosuppressants, or hormones; if they were diagnosed with genetic lipid abnormalities, neoplastic disease, acute coronary syndrome, and/or cerebral stroke during the 6 months prior to the study; if they had surgery during the 3 months prior to the study; if they had plasma activities of alanine transaminase and/or aspartate transaminase exceeding 3 times the upper laboratory normal limit; if they had hemoglobin A_{1c} (HbA_{1c}) levels exceeding 7.5%; and were treated with long-acting insulin (in diabetic patients). Another exclusion criterion was poor adherence to medical prescriptions in routine clinical management.

Design of the study and follow-up The study had a prospective design and consisted of an educational phase (4 weeks of intensive teaching in lifestyle changes), intervention phase I (lifestyle changes: lipid-lowering diet and physical activity, conducted as long as continuous improvement in the serum lipid profile was noted in the entire group), and intervention phase II (treatment with lifestyle changes and atorvastatin in patients who remained dyslipidemic despite lifestyle changes [phase IIa], or continuation of lifestyle changes without atorvastatin in patients who became nondyslipidemic on introduction of lifestyle changes

[phase IIb]). The length of intervention phase II was scheduled to equal the time during which improvements in the serum lipid profile were noted following lifestyle changes in intervention phase I.

A total of 49 dyslipidemic Caucasian HD patients were enrolled into the study (Supplementary material online, *Table S1*). Dyslipidemic HD patients did not differ in the prevalence of diabetes, myocardial infarction, and cigarette smoking from HD patients without dyslipidemia.¹⁸ Dyslipidemic diabetic patients without coronary artery disease (CAD) did not differ from dyslipidemic nondiabetic subjects with CAD in serum lipid concentrations (higher total cholesterol [TC] was shown in diabetics compared with CAD patients in nondialyzed subjects).¹⁹ All patients were dialyzed using low-flux polysulfone-based membranes, and low-molecular-weight heparin was used as an anticoagulant.

All patients were clinically stable when they simultaneously started lifestyle changes in a single dialysis center. Lifestyle changes were introduced in accordance with the KDOQI guidelines.² In brief, the planned diet contained approximately 2000 total calories per day: 7% of the calories as saturated fat; up to 10% of the calories as polyunsaturated fat; up to 20% of the calories as monounsaturated fat; 25% to 35% of the total calories as total fat; 50% to 60% of the total calories as complex carbohydrates; 20 to 30 g of fiber per day; and less than 200 mg of cholesterol per day. Patients consumed only natural food; dietary supplements were prohibited. Individually planned physical activity included 10,000 steps per day or 20- to 30-minute period of activity 3 to 4 times a week, or both interchangeably. Patients were interviewed at least 2 times a week by a nurse educated in nutrition and a nephrologist regarding adherence to the prescribed diet and physical activity. Pedometer measurements were taken. At every interview, patients were motivated to maintain lifestyle changes.

Regular patient evaluation was performed at baseline and 4, 14, and 21 weeks after the implementation of lifestyle changes. This phase was completed in 42 patients (64.8 ± 10.9 y; 18 women; dialysis vintage, 3.18, 0.67–10.9 y) and included, at completion, 35 individuals who continued to be dyslipidemic. These 35 patients were allocated to intervention phase IIa, which was started in 34 patients. Evening treatment with atorvastatin (Atoris, Krka, Poland) was started at a dose of 10 mg/d. After 4 weeks, the initial atorvastatin dose was increased to 20 mg/d in dyslipidemic patients and continued for the next 10 weeks. Thirty-two patients (65.8 ± 10.3 years; 15 women; dialysis vintage, 3.19, 1.02–11.2 y) completed the 14-week intervention phase IIa. Seven patients started (62.4 ± 12.7 y; 3 women; dialysis vintage, 4.14, 1.42–8.2 y) and completed the 14-week intervention phase IIb. The scheduled patient evaluations were performed at 0, 4, and 14 weeks after the implementation of intervention phase II. The flowchart of the study in

HD patients is shown in Supplementary material online (*Figure S1*).

Clinical and laboratory examinations Patients fasting overnight entered the dialysis center close to the start of their mid-week dialysis session. Anthropometric measurements were taken. Blood samples for laboratory analyses were obtained from the arteriovenous fistula just prior to the start of the dialysis session. HD sessions were conducted according to the schedule individually prescribed for each patient.

Blood laboratory analyses included the measurement of salusin α , CD36 monocyte count and expression, TC, HDL cholesterol, TG, MDA-oxLDL, immunoglobulin G (IgG) autoantibodies against oxLDL (IgG anti-oxLDL), insulin, fructosamine, and standard laboratory parameters. The LDL cholesterol concentration was calculated using the Friedewald formula: LDL cholesterol = TC – HDL cholesterol – TG/5.²⁰ Non-HDL cholesterol was the TC minus HDL cholesterol. Homeostasis model assessment–insulin resistance (HOMA-IR) was determined as fasting plasma insulin (μ U/ml) \times fasting plasma glucose (mmol/l) / 22.5.²¹

Laboratory methods Salusin α (Human) RIA Kit (Phoenix Pharmaceuticals, Burlingame, United States) was used for salusin α measurement. The antiserum used for this assay was raised against a synthetic form of the peptide. A centrifugal concentrator (Eppendorf Concentrator 5301, Eppendorf AG, Hamburg, Germany) was used as recommended by the RIA Kit manufacturer. All measurements were done in duplicate. Within-assay coefficient of variation (CV) was automatically calculated for each duplicate. In 6 consecutive study evaluations, %CVs (median and range) were 5.4 (0.1–13.5), 7.6 (0–13.3), 6.8 (0.9–13.6), 6.1 (0.2–12.4), 8.6 (0.2–14.6), and 5.5 (0.3–14.0). The interassay CV for these 6 evaluations was 8.6%.

Antibodies for quantification of CD36 expression on monocytes were purchased from BD Pharmingen, Oxford, United Kingdom. Fluorescein isothiocyanate (FITC) conjugated CD45 antibody and phycoerythrin (PE) conjugated CD14 antibody were used to identify monocytes and the anti-CD36 antibody was conjugated to allophycocyanin (APC). FITC conjugated mouse IgG1, PE conjugated mouse IgM, and APC conjugated mouse IgM were used as isotype-matched negative controls. A whole blood direct immunofluorescence staining technique was used. Data were acquired on a BD FACSCanto (San Jose, California, United States) flow cytometer. For each sample, 10,000 events were collected. During analysis, monocytes were identified by their reactivity with CD14-PE and nonreactivity with CD45-FITC and their distinctive forward scatter and side scatter profile. Mean fluorescence intensity (MFI) values were used as indirect measures of CD36 density. MFI values derived for cells stained with isotype

control antibodies were taken as indicators of autofluorescence, nonspecific antibody binding, or instrument noise.

MDA-oxLDL (MDA-modified apolipoprotein B) and IgG anti-oxLDL were determined in duplicate using enzyme immunoassays (Biomedica, Wien, Austria).

The patient control values for salusin α , monocyte CD36 expression, MDA-oxLDL, and IgG anti-oxLDL were obtained in 43 individuals with normal renal function, matched for sex and age with HD patients. Controls declared that they were healthy and had never taken lipid-lowering medication (Supplementary material online, *Table S1*).

Other parameters were determined using routine laboratory methods.

Statistical analysis The results were shown as mean \pm standard deviation or median and range.

The effects of the prospective study were analyzed using the parameters of patients who finished intervention phases I and IIa. The Friedman test was used if 3 to 4 sets of the results were compared for each phase of the prospective study. If the analysis indicated significance, the post hoc procedure was used. If 2 sets of the prospective results were analyzed, the Wilcoxon test or the paired *t* test was used.

The Multivariate Adaptive Regression Splines (MARSplines) model with generalized cross validation was used to show the parameters that could be useful in the prediction of each intervention's efficacy in patients who were lost during the study.

The Spearman correlation was performed between selected sets of results. The receiving operating characteristic (ROC) curve methodology was applied to find the predictive values of salusin α in regard to serum lipid parameters. Odds ratios (ORs) with 95% confidence intervals (95% CIs) were calculated using the Fisher exact probability test. A *P* value of less than 0.05 was considered statistically significant.

RESULTS Changes during the prospective study

The best serum lipid profile (the lowest LDL cholesterol concomitantly with a significantly higher HDL cholesterol) was shown after 14 weeks of introducing lifestyle changes (*TABLE 1*). This improvement was accompanied by a decrease of salusin α and an increase of MDA-oxLDL, IgG anti-oxLDL, CD36 expression, and fructosamine. During the next 7 weeks, LDL cholesterol increased, while the above parameters shifted towards the initial values (*TABLE 1*). Lifestyle interventions resulted in comparable changes of the metabolic parameters in diabetics and nondiabetics (Supplementary material online, *Figures S2–13*). During intervention phase I, 7 of 42 patients (16.7%) became nondyslipidemic according to the KDOQI guidelines.²

The best serum lipid profile after addition of atorvastatin (the lowest TC, LDL cholesterol,

TABLE 1 Changes in study parameters in patients who finished intervention phase I (n = 42)

Parameter	Lifestyle intervention, wk				P value			
	0	4	14	21	0 vs. 4	0 vs. 14	0 vs. 21	4 vs. 21
total cholesterol, mg/dl	198 (155–316)	201 (134–288)	199 (128–289)	204 (120–290)			0.778	14 vs. 21
HDL cholesterol, mg/dl	37 (24–68)	42.5 (31–68)	41.5 (29–69)	38 (27–71)	<0.05	<0.05	>0.05	>0.05
LDL cholesterol, mg/dl	128 (84–369)	123 (54–350)	114 (54–162)	128 (54–168)	>0.05	<0.05	>0.05	<0.05
non-HDL cholesterol, mg/dl	162 (123–279)	158.5 (89–250)	155.5 (93–249)	164 (93–247)			0.323	
MDA-OxLDL, mg/ml	0.72 (0.05–11.0)	0.65 (0.05–9.20)	1.02 (0.05–14.3)	0.40 (0.05–10.0)	>0.05	<0.05	>0.05	<0.05
IgG anti-OxLDL, mU/ml	102.5 (43–1300)	149 (68–1000)	143 (43–1400)	171 (11–1200)	<0.05	<0.05	>0.05	<0.05
triglycerides, mg/dl	161.5 (93–466)	183 (86–486)	183 (83–438)	170.5 (65–432)			0.050	
salusin α , pmol/l	14.1 (10.3–21.3)	14.1 (7.22–48.3)	6.08 (3.80–14.4)	6.65 (3.80–24.2)			<0.0001	
CD36 expression, MFI	NA	1235 (384–4399)	1626 (610–7360)	1030 (458–3150)	>0.05	<0.0001	<0.0001	>0.05
insulin, mIU/ml	11.1 (1.4–70.8)	11.4 (3.2–61.4)	11.9 (1.4–65.2)	11.7 (2.2–56.5)	NA	NA	NA	<0.05
HOMA-IR	3.7 (0.50–37.0)	3.2 (0.62–35.3)	3.0 (0.29–32.7)	3.1 (0.44–24.2)			0.742	
fructosamine, mmol/l	251 (167–458)	NA	310 (194–716)	284 (156–417)			0.999	
					NA	<0.05	<0.0001	>0.05

Data are presented as median and range. The Friedman test was used with post hoc tests as appropriate. Conversion factors to SI units are as follows: insulin in mIU/ml to pmol/l, 6.945; serum cholesterol in mg/dl to mmol/l, 0.02586; triglycerides in mg/dl to mmol/l, 0.01129.

Abbreviations: anti-OxLDL – autoantibodies to oxidized low-density lipoproteins, HDL – high-density lipoprotein, HOMA-IR – homeostasis model assessment–insulin resistance, IgG – immunoglobulin G, LDL – low-density lipoprotein, MDA-OxLDL – malondialdehyde oxidized low-density lipoprotein, MFI – mean fluorescence intensity, NA – not available

TABLE 2 Changes in study parameters in patients treated with atorvastatin who completed the entire study (n = 32)

Parameter	Atorvastatin treatment, wk			P value			
	0	4	14	0 vs. 4 vs. 14	0 vs. 4	0 vs. 14	4 vs. 14
total cholesterol, mg/dl	214.5 (158–290)	163 (124–251)	159.5 (108–226)	<0.0001 ^a	<0.05	<0.05	<0.05
HDL cholesterol, mg/dl	37 (29–69)	39.5 (23–72)	40 (25–75)	0.198 ^a	–	–	–
LDL cholesterol, mg/dl	135 (90–168)	90 (60–162)	87.5 (43–182)	<0.0001 ^a	<0.05	<0.05	>0.05
non-HDL cholesterol, mg/dl	179.5 (122–247)	119 (84–211)	121.5 (62–174)	<0.0001 ^a	<0.05	<0.05	>0.05
MDA-OxLDL, µg/ml	0.50 (0.05–9.20)	0.81 (0.05–11.0)	0.05 (0.05–11.0)	0.166 ^a	–	–	–
IgG anti-OxLDL, mU/ml	153 (68–1000)	178 (65–1300)	157.5 (48–1300)	0.002 ^a	<0.05	>0.05	>0.05
triglycerides, mg/dl	178 (65–432)	154 (70–313)	147.5 (59–310)	0.033 ^a	>0.05	<0.05	>0.05
salusin α, pmol/l	6.65 (3.80–24.2)	5.14 (4.56–10.3)	65.0 (4.94–134.1)	0.00039 ^a	>0.05	<0.05	<0.05
CD36 expression, MFI	938 (458–3150)	1167 (427–5453)	1433 (517–2971)	0.0001 ^a	>0.05	<0.05	<0.05
insulin, mIU/ml	9.8 (2.2–56.5)	NA	9.8 (2.6–61.5)	–	–	0.254 ^b	–
HOMA-IR	2.8 (0.44–24.2)	NA	2.0 (0.54–26.2)	–	–	0.340 ^b	–
fructosamine, mmol/l	288 ± 58	NA	267 ± 54	–	–	0.025 ^c	–

Results are expressed as mean ± standard deviation or median and range.

a Friedman (with post hoc tests, as appropriate), **b** Wilcoxon, **c** *t* test

Conversion factors and abbreviations: see [TABLE 1](#)

and TG) was observed at the end of the study ([TABLE 2](#)). This improvement in the serum lipid profile was accompanied by an increase in both plasma salusin α and CD36 expression and a decrease in fructosamine ([TABLE 2](#)). Administration of atorvastatin resulted in comparable changes of the analyzed metabolic parameters in diabetics and nondiabetics (Supplementary material online, *Figures S14–25*). At the end of intervention phase IIa, 19 of 32 patients (59.4%) were nondyslipidemic according to the Kidney Disease Outcomes Quality Initiative (KDOQI) criteria.² At the end of intervention phase IIb, 3 of 7 patients were nondyslipidemic according to the KDOQI criteria.² Owing to a small number of patients, a statistical analysis of their clinical and laboratory data was not performed. Therefore, 35 weeks after the start of the study, only in 3 of 39 patients who completed the entire study (7.7%) dyslipidemia was shown to resolve by using lifestyle modifications only.

The reasons for withdrawal from the study are shown in Supplementary material online, *Figure S1*.

Differences in the effects of lifestyle changes and atorvastatin Medication with atorvastatin was more effective than lifestyle changes in the resolution of dyslipidemia (59.4% vs. 16.7%, $P = 0.0001$, χ^2 test). Taking the MARSplines models for the prediction of responsiveness to both interventions in patients who were lost or did not need medication with atorvastatin, the difference was still more pronounced (61.2% vs. 14.3%, $P < 0.0001$, χ^2 test).

Only lifestyle changes were significantly effective in increasing the concentration of HDL cholesterol, but they also increased MDA-oxLDL; only atorvastatin was effective in decreasing

non-HDL cholesterol and TG; both interventions were shown to reduce LDL cholesterol levels and to increase monocyte CD36 expression and IgG anti-oxLDL. The opposite effects of both interventions were shown on salusin α and fructosamine. The differences between the effects of lifestyle changes or atorvastatin on HOMA-IR were not significant ([TABLES 1 and 2](#)).

Selected significant correlations At the beginning of the study, there was a correlation between salusin α and LDL cholesterol ($r = -0.342$, $P = 0.02$, $n = 49$). The ROC curve methodology indicated that a salusin α level of 14.4 pmol/l (Supplementary material online, *Figure S26*) is a cut-off value in the prognosis of serum LDL cholesterol concentration below or over the lower limit of LDL cholesterol values designated as highly borderline (130–159 mg/dl). Dyslipidemic HD patients with salusin α levels of 14.4 pmol/l and higher compared with patients with lower salusin α levels had over 5-fold higher chance of having LDL cholesterol levels of less than 130 mg/dl (OR, 5.14; 95% CI, 1.52–17.4; $n = 49$; $P = 0.01$). There was no significant correlation between salusin α and HDL cholesterol levels in all 49 HD patients. However, when HD patients were divided into subgroups with an HDL cholesterol cut-off value of 40 mg/dl, both subgroups had similar plasma concentrations of salusin α (14.3 ± 2.7 pmol/l for HDL cholesterol <40 mg/dl, 14.3 ± 2.5 pmol/l for HDL cholesterol ≥40 mg/dl), and a correlation between salusin α and HDL cholesterol was present in the subgroup with HDL cholesterol concentration of 40 mg/dl or higher ($r = 0.628$; $P = 0.005$; $n = 18$). The subgroup with HDL cholesterol below 40 mg/dl included 11 CAD patients (73.3% of the subgroup).

At 4 weeks, a correlation between plasma salusin α level and CD36 expression was shown in the entire group of HD patients ($r = 0.392$; $P = 0.007$; $n = 47$) and in non-CAD patients ($r = 0.448$; $P = 0.009$; $n = 33$). In CAD patients, a correlation with CD36 expression did not reach significance ($r = 0.389$; $P = 0.2$; $n = 14$), but a correlation was shown between plasma salusin α and CD36 cell count ($r = 0.717$; $P = 0.006$; $n = 14$). MDA-oxLDL level of 1.5 mg/dl and higher was associated with CD36 MFI >2000 (OR, 12.08; 95% CI, 1.27–114.6; $P = 0.018$). CD36 expression correlated with IgG anti-oxLDL ($r = 0.336$; $P = 0.02$; $n = 47$).

At the end of intervention phase I ($n = 42$), correlations were shown between monocyte CD36 expression and TC ($r = -0.359$; $P = 0.02$), LDL cholesterol ($r = -0.354$; $P = 0.022$), and non-HDL cholesterol ($r = -0.345$; $P = 0.03$). CD36 expression over the median value of 1030 was associated with TC of less than 200 mg/dl and non-HDL cholesterol of less than 160 mg/dl (for both: OR, 10.63; 95% CI, 2.51–44.99; $P = 0.002$).

At the end of intervention phase IIa, the lowest TC concentrations were observed in patients who had the highest monocyte CD36 expression ($r = -0.389$; $P = 0.027$; $n = 32$). A borderline correlation was shown for CD36 expression and non-HDL cholesterol ($r = -0.328$; $P = 0.067$; $n = 32$).

DISCUSSION Lifestyle changes were recommended as the first step in treating dyslipidemia in HD patients,² and they were also advised in dialysis patients with hypertriglyceridemia in the newest KDIGO guidelines.²² However, our study shows that lifestyle modifications have a selective and short-term efficacy in resolution of dyslipidemia in HD patients. Generally, completion rates on diets are not high.^{23,24} In our dyslipidemic HD patients undergoing lifestyle changes, a significant decrease in LDL cholesterol and an increase in HDL cholesterol levels were shown, TG remained elevated, and only about 17% of the patients became nondyslipidemic. Of note, current evidence indicates that the benefit of therapy aimed at increasing HDL cholesterol may be “more than questionable” in CKD patients.²⁵ Moreover, an increase in protein glycation and lipid oxidation was detected in the effective period of the intervention, independently of the diabetic status. Higher levels of fructosamine, a marker of glycemic control alternative to HbA_{1c} in dialysis patients,²⁶ reflected enhanced protein glycation on the lipid-lowering diet that included complex carbohydrates of 50% to 60% of the total calories,² although increases in fasting glucose levels were not observed. Therefore, this effect cannot be shown if glucose is the only parameter applied as an indication of carbohydrate metabolism control. HOMA-IR was not affected by lipid-lowering diet. The study by Ostrowska et al.²⁷ showed no significant evidence for a direct link between nutrition, atherogenic index, and insulin resistance.

Protein glycation and lipid oxidation are overlapping processes²⁸; therefore, a simultaneous increase of plasma MDA-oxLDL concentration was not a surprise. MDA is a natural product of polyunsaturated fatty acid peroxidation.^{29,30} Polyunsaturated fat contributes up to 10% of the entire calorie intake. Moreover, the total antioxidant status decreases in HD patients on a lipid-lowering diet.³¹ Consumption of prooxidants and a decrease in antioxidant capacity may be a plausible explanation for increased MDA-oxLDL.

A decrease in plasma salusin α and an increase of both monocyte CD36 expression and plasma IgG anti-oxLDL levels may represent the counteractive mechanisms occurring during intervention phase I in response to blood changes in the above metabolic parameters.

Nagashima et al.⁵ demonstrated that intravenous infusion of salusin α increased serum HDL cholesterol and decreased serum TC levels without affecting the CD36 expression in exudate peritoneal macrophages of apolipoprotein E-deficient mice. In our study, negative correlations/associations between salusin α and lower LDL cholesterol indicate that dyslipidemic HD patients not treated for lipid abnormalities (start of the study) benefit from lower LDL cholesterol levels if they have higher plasma salusin α levels. A positive correlation between salusin α and HDL cholesterol was shown only in dyslipidemic HD patients with HDL cholesterol of 40 mg/dl or higher. It is possible that the group less affected by lipid disturbances (HDL cholesterol, ≥ 40 mg/dl) was sensitive to salusin α and responded to higher plasma salusin α with higher HDL cholesterol levels. In the second group (HDL cholesterol, <40 mg/dl), salusin α was ineffectively upregulated and patients (73.3% had CAD) continued to have lower HDL cholesterol levels despite similar salusin α levels to those in the group of responders. A lifestyle-induced improvement in the serum lipid profile was expected to be associated with an increase in plasma salusin α level. In fact, the reverse reaction was shown. An increase in lipid oxidation and protein glycation could contribute to the suppression of salusin α secretion, and improvement in HDL and LDL cholesterol levels was a result of lifestyle changes without the involvement of salusin α . On the other hand, if a feedback between serum lipids and salusin α was at least partially maintained in dyslipidemic HD patients, an improvement of the serum lipid profile (decreased LDL cholesterol, increased HDL cholesterol) induced by lifestyle changes could diminish a need for lipid regulatory activity exerted by endogenous bioactive salusin α to keep more favorable serum levels of both cholesterol, and, therefore, its plasma concentration decreased. Further studies are needed to elucidate this problem.

High plasma concentrations of oxLDL have been reported to stimulate CD36 expression.¹¹ In our study, in dyslipidemic HD patients with high MDA-oxLDL levels, a positive association between

oxLDL and CD36 was revealed. Therefore, an increase in MDA-oxLDL induced by lifestyle changes could provoke, in response, a development of IgG anti-oxLDL as well as increased scavenger receptor CD36 expression.

In HD patients that remained dyslipidemic despite lifestyle changes, serum lipid profile was improved by atorvastatin treatment. Nearly 60% of the patients became nondyslipidemic on relatively low atorvastatin doses (10–20 mg/d). Treatment with atorvastatin was not associated with an increase in fructosamine, which was observed after implementation of lifestyle changes. The importance of baseline fructosamine concentrations on simvastatin-induced changes in serum lipids was shown by Miserez et al.³² Reports on the effect of atorvastatin on oxidative stress are controversial.^{33–36} In our study, changes in plasma MDA-oxLDL levels remained nonsignificant. Therefore, enhancement of both protein glycation and lipid oxidation, which were possible inhibitors of salusin α secretion during lifestyle changes, was not observed during treatment with atorvastatin, and increased salusin α levels could contribute to the beneficial effect of atorvastatin on the serum lipid profile, as was demonstrated with intravenous administration of salusin α in mice.⁵ An increase in salusin α during treatment with atorvastatin may be explained by a specific atorvastatin effect on the secretion of salusin α . Further studies in this field could be helpful.

An increase in monocyte CD36 expression during treatment with atorvastatin is also an interesting finding. It occurred without an increase in MDA-oxLDL levels. In 2002, Fuhrman et al.³⁷ demonstrated in hypercholesterolemic patients with normal renal function that atorvastatin-induced decrease in both TC and LDL cholesterol was accompanied by an increase in CD36 expression on freshly seeded monocytes. This finding was commented as interesting, but not further analyzed. Our study indicates that treatment with atorvastatin increases circulating monocyte CD36 expression, which explains why CD36 expression may be increased on freshly seeded monocytes.

The serum lipid profile and insulin resistance seen in the examined dyslipidemic HD patients resemble abnormalities characteristic for CD36 deficiency (increased TG and insulin resistance, decreased HDL cholesterol).³⁸ In fact, monocyte CD36 expression was lower in our HD patients than in controls. The cause of downregulation of CD36 expression may arise from the uremic state and HD treatment: the decreased level of interleukin (IL)-4³⁹ and persistent endotoxemia.⁴⁰ IL-4 is a stimulator of CD36 expression,^{11,39} whereas lipopolysaccharide decreases CD36 expression.⁴¹

Deficiency of receptor CD36 is involved in decreased transport of long-chain fatty acids,⁸ resulting in serum lipid abnormalities (HDL cholesterol, decrease; TG, increase), insulin resistance, metabolic syndrome, and, finally, severe atherosclerotic disease.⁴² Recently, the role of

CD36 as a promoter of atherosclerosis has been questioned.^{42–44} Our study seems to provide further evidence that the CD36 scavenger receptor may be involved in the inhibition of the growth of atherosclerotic lesions. At the end of both intervention phases I and IIa, the lowest levels of cholesterol (TC, LDL cholesterol, or non-LDL cholesterol) were observed in HD patients with the highest monocyte CD36 expression. Moreover, CD36 expression positively correlated with the plasma level of salusin α , which is considered as an inhibitor of atherosclerosis,^{5,6,45–48} and with IgG anti-oxLDL, which reduce the progression of atherosclerosis.¹⁷

In this study, we demonstrated for the first time the effect of lifestyle changes and atorvastatin treatment on salusin α and monocyte CD36 expression in HD patients. We consider a relatively small number of dyslipidemic patients included into the study as the main limitation of our study because the analyses in smaller subgroups, although interesting (an example responders vs. nonresponders to both interventions in terms of dyslipidemia resolution), are questionable from the statistical point of view owing to insufficient sample power.

Acknowledgements This study was funded by the Poznań University of Medical Sciences, Poznań, Poland (grant number, 502-01-02 225 363-03 679) and by the Greater Poland Foundation for Scientific Research and Healthcare.

We would like to express our gratitude to Dr. Margarita Lianeri for her assistance.

Supplementary material online Supplementary material is available with the online version of the article at www.pamw.pl.

REFERENCES

- 1 Musunuru K. Atherogenic dyslipidemia: cardiovascular risk and dietary intervention. *Lipids*. 2010; 10: 907–914.
- 2 National Kidney Foundation. Kidney Disease Outcomes Quality Initiative (K/DOQI) Group. K/DOQI clinical practice guidelines for management of dyslipidemias in patients with dialysis – dependent chronic kidney disease. *Am J Kidney Dis*. 2003; 41: Suppl 3: S3–S91.
- 3 Massy ZA, Ma JZ, Louis TA, et al. Lipid-lowering therapy in patients with renal disease. *Kidney Int*. 1995; 48: 188–198.
- 4 Wanner C, Krane V, März W, et al. Atorvastatin in patients with type 2 diabetes mellitus undergoing hemodialysis. *N Engl J Med*. 2005; 353: 238–248.
- 5 Nagashima M, Watanabe T, Shiraishi Y, et al. Chronic infusion of salusin- α and - β exerts opposite effects on atherosclerotic lesion development in apolipoprotein E-deficient mice. *Atherosclerosis*. 2010; 212: 70–77.
- 6 Watanabe T, Sato K, Itoh F, et al. Endogenous bioactive peptides as potential biomarkers for atherosclerotic coronary heart disease. *Sensors (Basel)*. 2012; 12: 4974–4985.
- 7 Endemann G, Stanton LW, Madden KS, et al. CD36 is a receptor for oxidized low density lipoprotein. *J Biol Chem*. 1993; 268: 11811–11816.
- 8 Febbraio M, Abumrad NA, Hajjar DP, et al. A null mutation in murine CD36 reveals an important role in fatty acid and lipoprotein metabolism. *J Biol Chem*. 1999; 274: 19055–19062.
- 9 Love-Gregory L, Sherva R, Schappe T, et al. Common CD36 SNPs reduce protein expression and may contribute to a protective atherogenic profile. *Hum Mol Genet*. 2011; 20: 193–201.
- 10 Brundert M, Heeren J, Merkel M, et al. Scavenger receptor CD36 mediates uptake of high density lipoproteins in mice and by cultured cells. *J Lipid Res*. 2011; 52: 745–758.

- 11 Feng J, Han J, Pearce SF, et al. Induction of CD36 expression by oxidized LDL and IL-4 by a common signaling pathway dependent on protein kinase C and PPAR γ . *J Lipid Res.* 2000; 41: 688-696.
- 12 Han J, Hajjar DP, Febbraio M, et al. Native and modified low density lipoproteins increase the functional expression of the macrophage class B scavenger receptor, CD36. *J Biol Chem.* 1997; 272: 21654-21659.
- 13 Iä-Herttuala S, Palinski W, Rosenfeld ME, et al. Evidence for the presence of oxidatively modified low density lipoprotein in atherosclerotic lesions of rabbit and man. *J Clin Invest.* 1989; 84: 1086-1095.
- 14 Febbraio M, Hajjar DP, Silverstein RL. CD36: a class B scavenger receptor involved in angiogenesis, atherosclerosis, inflammation, and lipid metabolism. *J Clin Invest.* 2001; 108: 785-791.
- 15 Silverstein RL, Febbraio M. CD36 and atherosclerosis. *Curr Opin Lipidol.* 2000; 11: 483-491.
- 16 Shaw PX, Hökkö S, Tsimikas S, et al. Human-derived anti-oxidized LDL autoantibody blocks uptake of oxidized LDL by macrophages and localizes to atherosclerotic lesions in vivo. *Arterioscler Thromb Vasc Biol.* 2001; 21: 1333-1339.
- 17 Tsimikas S, Miyahara A, Hartvigsen K, et al. Human oxidation-specific antibodies reduce foam cell formation and atherosclerosis progression. *J Am Coll Cardiol.* 2011; 58: 1715-1727.
- 18 Napora M, Graczykowska A, Próchniewska K, et al. Relationship between serum asymmetric dimethylarginine and left ventricular structure and function in patients with end-stage renal disease treated with hemodialysis. *Pol Arch Med Wewn.* 2012; 122: 226-234.
- 19 Opolski G, Strojek K, Kurzelewski M, et al. Cardiovascular therapy, diagnostic procedures, and control of risk factors in patients with diabetes or coronary artery disease in Poland: the Kardia-Pol registry. *Pol Arch Med Wewn.* 2012; 122: 413-421.
- 20 Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem.* 1972; 18: 499-502.
- 21 Matthews DR, Hosker JP, Rudenski AS, et al. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. *Diabetologia.* 1985; 28: 412-419.
- 22 Wanner C, Tonelli M; The Kidney Disease: Improving Global Outcomes Lipid Guideline Development Work Group Members. KDIGO Clinical Practice Guideline for Lipid Management in CKD: summary of recommendation statements and clinical approach to the patient. *Kidney Int.* 2014; 85: 1303-1309.
- 23 Brehm BJ, Seeley RJ, Daniels SR, et al. A randomized trial comparing a very low carbohydrate diet and a calorie-restricted low fat diet on body weight and cardiovascular risk factors in healthy women. *J Clin Endocrinol Metab.* 2003; 88: 1617-1623.
- 24 Jedrzejowski A, Lis D, Majcher A, et al. [Effect of lipid lowering diet on serum lipids in peritoneal dialysis patients]. *Pol Merkurius Lekarski.* 2000; 8: 266-267. Polish.
- 25 Speer T, Zewinger S, Fliser D. Uraemic dyslipidaemia revisited: role of high-density lipoprotein. *Nephrol Dial Transplant.* 2013; 28: 2456-2463.
- 26 Mittman N, Desiraju B, Fazil I, et al. Serum fructosamine versus glycosylated hemoglobin as an index of glycemic control, hospitalization, and infection in diabetic hemodialysis patients. *Kidney Int.* 2010; 117: Suppl: S41-S45.
- 27 Ostrowska L, Witczak K, Adamska E. Effect of nutrition and atherogenic index on the occurrence and intensity of insulin resistance. *Pol Arch Med Wewn.* 2013; 123: 289-296.
- 28 Ravandi A, Kuksis A, Shaikh NA. Glucosylated glycerophosphoethanolamines are the major LDL glycation products and increase LDL susceptibility to oxidation: evidence of their presence in atherosclerotic lesions. *Arterioscler Thromb Vasc Biol.* 2000; 20: 467-477.
- 29 Janero DR. Malondialdehyde and thiobarbituric acid-reactivity as diagnostic indices of lipid peroxidation and peroxidative tissue injury. *Free Radic Biol Med.* 1990; 9: 515-540.
- 30 Halliwell B, Chirico S. Lipid peroxidation: its mechanism, measurement, and significance. *Am J Clin Nutr.* 1993; 57: 5Suppl:715S-724S; discussion 724S.
- 31 Mastalerz-Migas A, Reksa D, Pokorski M, et al. Comparison of a statin vs. hypolipemic diet on the oxidant status in hemodialyzed patients with chronic renal failure. *J Physiol Pharmacol.* 2007; 58: Suppl 5: 363-370.
- 32 Miserez AR, Rossi FA, Keller U. Prediction of the therapeutic response to simvastatin by pretreatment lipid concentrations in 2082 subjects. *Eur J Clin Pharmacol.* 1994; 46: 107-114.
- 33 Sathyapalan T, Shepherd J, Coady AM, et al. Atorvastatin reduces malondialdehyde concentrations in patients with polycystic ovary syndrome. *J Clin Endocrinol Metab.* 2012; 97: 3951-3955.
- 34 Usharani P, Mateen AA, Naidu MU, et al. Effect of NCB-02, atorvastatin and placebo on endothelial function, oxidative stress and inflammatory markers in patients with type 2 diabetes mellitus: a randomized, parallel-group, placebo-controlled, 8-week study. *Drugs R D.* 2008; 9: 243-250.
- 35 Sozer V, Uzun H, Gelisgen R, et al. The effects of atorvastatin on oxidative stress in L-NAME-treated rats. *Scand J Clin Lab Invest.* 2013; 73: 591-597.
- 36 Elmadhun NY, Lassaletta AD, Chu LM, et al. Atorvastatin increases oxidative stress and modulates angiogenesis in Ossabaw swine with the metabolic syndrome. *J Thorac Cardiovasc Surg.* 2012; 144: 1486-1493.
- 37 Fuhrman B, Koren L, Volkova N, et al. Atorvastatin therapy in hypercholesterolemic patients suppresses cellular uptake of oxidized-LDL by differentiating monocytes. *Atherosclerosis.* 2002; 164: 179-185.
- 38 Masuda D, Hirano K, Oku H, et al. Chylomicron remnants are increased in the postprandial state in CD36 deficiency. *J Lipid Res.* 2009; 50: 999-1011.
- 39 Cohen SD, Phillips TM, Khetpal P, et al. Cytokine patterns and survival in haemodialysis patients. *Nephrol Dial Transplant.* 2010; 25: 1239-1243.
- 40 Feroze U, Kalantar-Zadeh K, Sterling KA, et al. Examining associations of circulating endotoxin with nutritional status, inflammation, and mortality in hemodialysis patients. *J Ren Nutr.* 2012; 22: 317-326.
- 41 Yesner LM, Huh HY, Pearce SF, et al. Regulation of monocyte CD36 and thrombospondin-1 expression by soluble mediators. *Arterioscler Thromb Vasc Biol.* 1996; 16: 1019-1025.
- 42 Yuasa-Kawase M, Masuda D, Yamashita T, et al. Patients with CD36 deficiency are associated with enhanced atherosclerotic cardiovascular diseases. *J Atheroscler Thromb.* 2012; 19: 263-275.
- 43 Moore KJ, Kunjathoor VV, Koehn SL, et al. Loss of receptor-mediated lipid uptake via scavenger receptor A or CD36 pathways does not ameliorate atherosclerosis in hyperlipidemic mice. *J Clin Invest.* 2005; 115: 2192-201.
- 44 Collot-Teixeira S, Martin J, McDermott-Roe C, et al. CD36 and macrophages in atherosclerosis. *Cardiovasc Res.* 2007; 75: 468-477.
- 45 Watanabe T, Nishio K, Kanome T, et al. Impact of salusin-alpha and -beta on human macrophage foam cell formation and coronary atherosclerosis. *Circulation.* 2008; 117: 638-648.
- 46 Watanabe T, Suguro T, Sato K, et al. Serum salusin-alpha levels are decreased and correlated negatively with carotid atherosclerosis in essential hypertensive patients. *Hypertens Res.* 2008; 31: 463-468.
- 47 Watanabe T, Sato K, Itoh F, et al. The roles of salusins in atherosclerosis and related cardiovascular diseases. *J Am Soc Hypertens.* 2011; 5: 359-365.
- 48 Ti Y, Wang F, Wang ZH, et al. Associations of serum salusin-alpha levels with atherosclerosis and left ventricular diastolic dysfunction in essential hypertension. *J Hum Hypertens.* 2012; 26: 603-609.

SUPPLEMENTARY MATERIAL ONLINE

Table S1 Comparison of hemodialysis patients with dyslipidemia and controls at baseline

Parameter	Patients n = 49	Controls n = 43	P value
male sex, n, % of all	28 (57.1)	21 (42.9)	0.635 ^b
age, y	64.2 ±10.7	62.0 ±9.9	0.319 ^c
cigarette smoking >5 packs/year, n, % of all	6 (12.2)	10 (23.3)	0.265 ^d
alcohol consumption >30 g/d, n, % of all	0 (0)	1 (2.3)	0.467 ^e
CAD, n, % of all	15 (30.6)	0 (0)	<0.0001 ^e
– myocardial infarction, n, % of all	10 (20.4)	0 (0)	0.001 ^e
type 2 diabetes, n, % of all	17 (34.7)	0 (0)	<0.0001 ^e
myocardial infarction or cerebral stroke in parents or siblings, n, % of all	7 (16.7)	6 (13.9)	0.799 ^d
history of lipid-lowering therapy, n, % of all	38 (77.6)	0 (0)	<0.0001 ^e
systolic blood pressure, mmHg	144 (90–180)	130 (100–160)	0.174 ^f
diastolic blood pressure, mmHg	80 (60–90)	80 (60–95)	0.804 ^f
cerebral stroke, n, % of all	9 (18.4)	0 (0)	0.003 ^e
RRT vintage, years	2.61 (0.22–10.4)	–	–
urine volume, ml/24 hours	800 (0–3000)	NA	–
BMI, kg/m ²	30.4 (20.2–51.2)	29.7 (20.5–39.6)	0.616 ^f
Hs-CRP, mg/l	8.0 (4.0–143.0)	1.80 (0.10–9.30)	<0.0001 ^f
fasting glucose, mg/dl	112 (58–240)	99 (85–126)	0.199 ^f
administration of ESA, units/kg/wk	37.9 (0–106.5)	0 (0)	–
urea, mg/dl	114 ±38	30.4 ±7.4	<0.0001 ^g
creatinine, mg/dl	7.4 (2.4–13.1)	0.7 (0.5–1.2)	<0.0001 ^f
MUAC, cm	33.4 ±4.5	31.1 ±2.9	0.004 ^g
MUAMC, cm	29.6 ±3.7	26.3 ±3.7	0.00005 ^c
TSF, cm	1.0 (0.2–3.2)	1.1 (0.3–4.0)	0.169 ^f
waist-to-hip ratio	0.98 ±0.07	0.91 ±0.08	0.0001 ^c
waist- to-height ratio	0.65 ±0.10	0.60 ±0.08	0.014 ^c
insulin, µIU/ml	11.5 (1.4–70.8)	7.9 (3.1–20.1)	0.021 ^f
HOMA-IR	3.52 (0.35–37.0)	1.70 (0–5.55)	0.013 ^f
fructosamine, µmol/l	256 (168–459)	227 (172–348)	0.030 ^f
Total cholesterol, mg/dl	199 (155–316)	229 (147–323)	0.005 ^f
LDL cholesterol, mg/dl	130 (84–369)	136 (60–208)	0.501 ^f
MDA-oxLDL, µg/ml	0.76 (0.05–11.0)	0.72 (0.05–13.1)	0.341 ^f
IgG anti-oxLDL, mU/ml	103 (38–1300)	207 (58–1730)	0.004 ^f
HDL cholesterol, mg/dl	37 (24–68)	62 (31–109)	<0.0001 ^f
non-HDL cholesterol, mg/dl	165 (123–279)	165 (77–256)	0.863 ^f
triglycerides, mg/dl	162 (93–466)	113 (45–342)	0.00002 ^f
salusin α, pmol/l	14.1 (10.3–21.3)	4.94 (3.80–61.6)	<0.0001 ^f
monocyte, cells/µl	472 (215–1102) ^a	348 (93–1040)	0.002 ^f
CD36, cells/µl	375 (75–824) ^a	302 (93–940)	0.061 ^f
CD36 as percent of monocyte	82.8 (22.4–99.6) ^a	90.4 (47.1–100)	0.008 ^f
CD36 expression, MFI	1286 (384–4399) ^a	1768 (714–9324)	0.0005 ^f

Results are expressed as a number of patients in the entire group (n), a percentage of patients in the entire group, mean ± standard deviation, or median and range.

SUPPLEMENTARY MATERIAL ONLINE

a n = 47 (evaluation with the use of results obtained after 4 weeks from the start of the study)

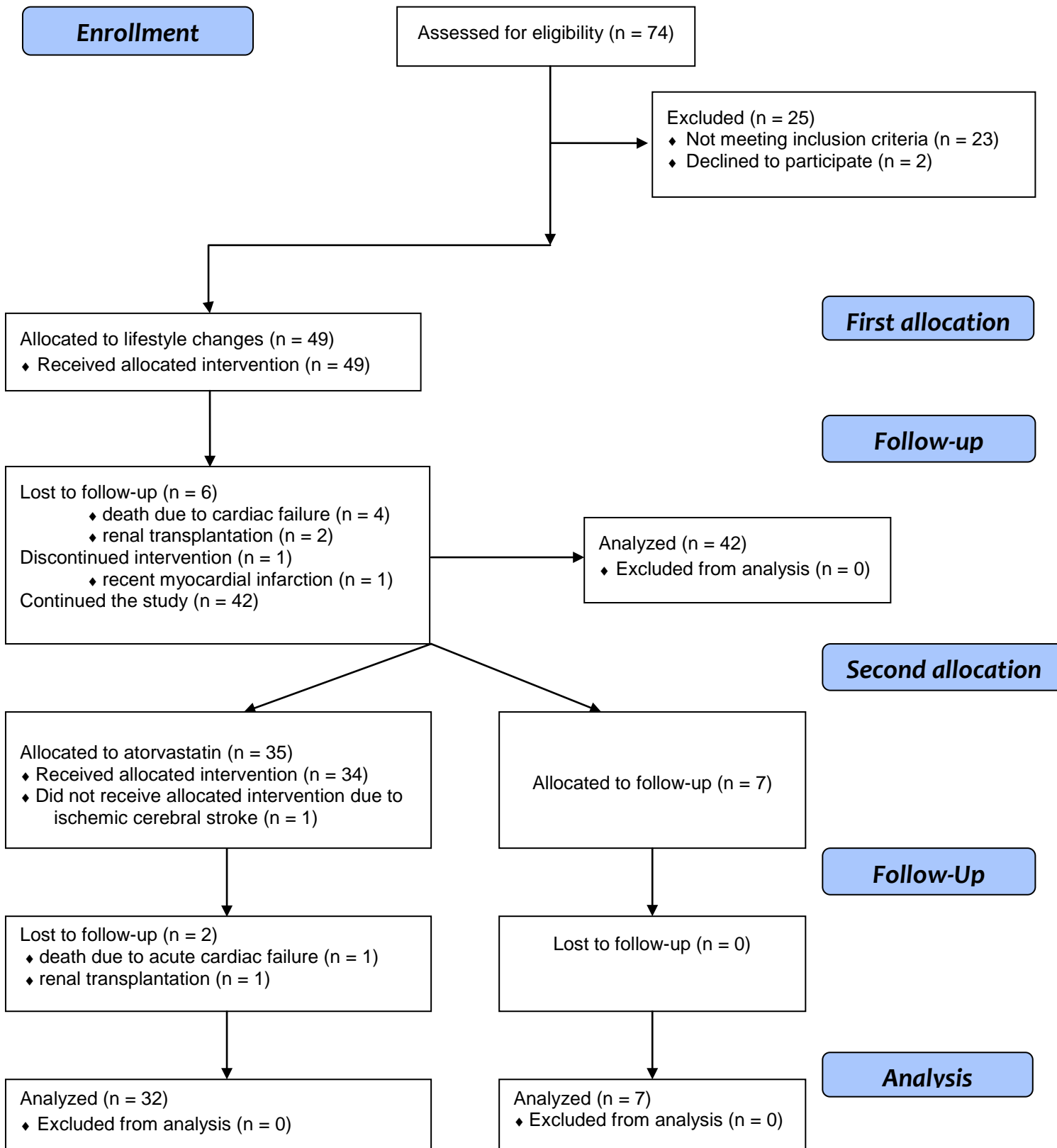
b – χ^2 test, **c** – *t* test, **d** – χ^2 test with Yates correction, **e** – Fisher exact test, **f** – Mann–Whitney test, **g** – Cochran Cox

Conversion factors to SI units: creatinine in mg/dl to $\mu\text{mol/l}$, $\times 88.4$; glucose in mg/dl to mmol/l, $\times 0.0555$; insulin in $\mu\text{IU/ml}$ to pmol/l, $\times 6.945$; serum cholesterol in mg/dl to mmol/l, $\times 0.02586$; triglycerides in mg/dl to mmol/l, $\times 0.01129$

Abbreviations: BMI – body mass index, CAD – coronary artery disease, ESA – erythropoietin-stimulating agents, HDL – high-density lipoprotein, HOMA-IR – homeostasis model assessment–insulin resistance, hsCRP – high-sensitivity C-reactive protein, LDL – low density lipoprotein, MDA-oxLDL – malondialdehyde oxidized low-density lipoproteins, MFI – mean fluorescence intensity, MUAC – mid-upper arm circumference, MUAMC – mid upper arm muscle circumference, RRT – renal replacement therapy, TSF – triceps skinfold thickness

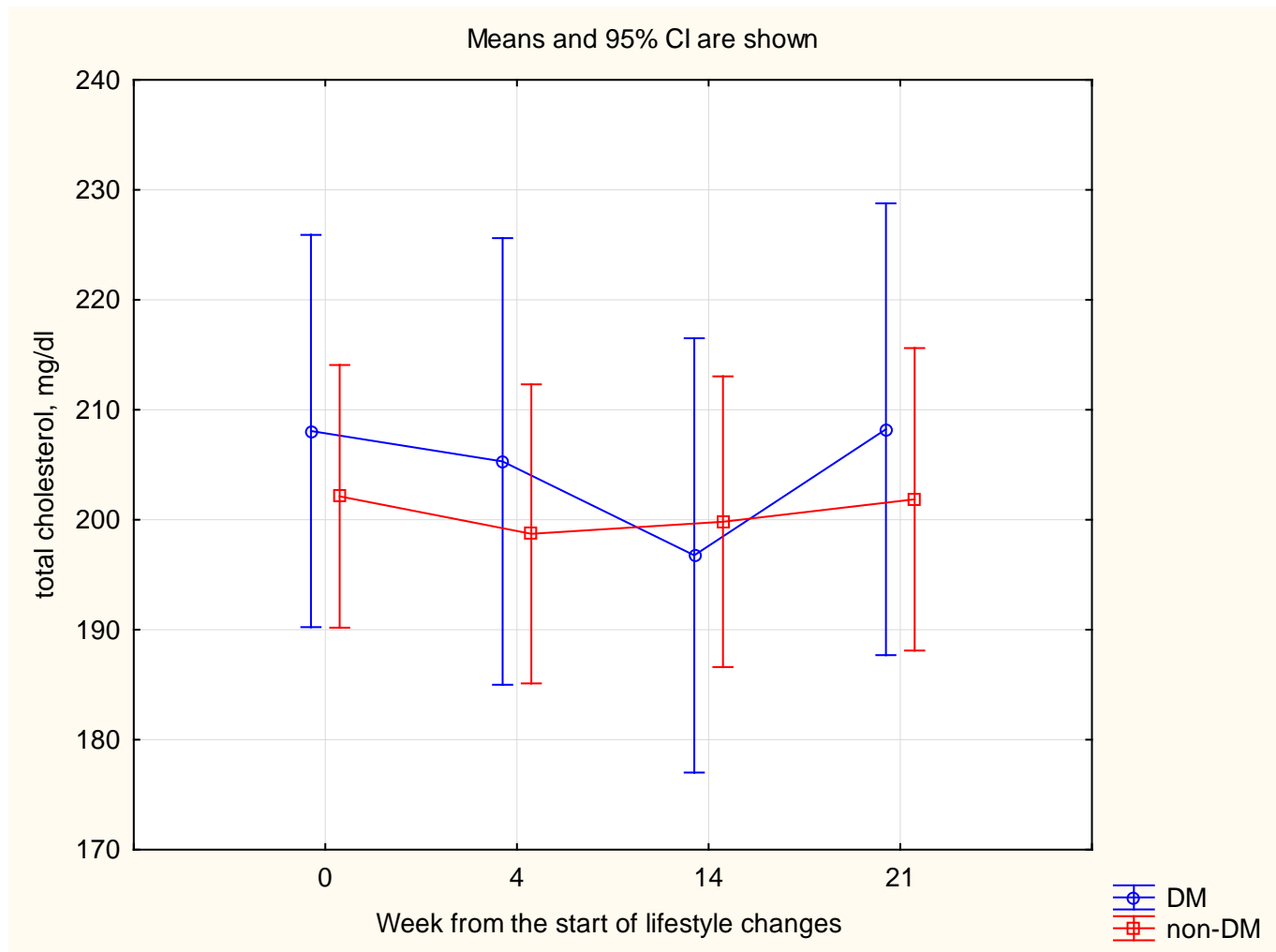
Figure S1

Flowchart of hemodialyzed patients



SUPPLEMENTARY MATERIAL ONLINE

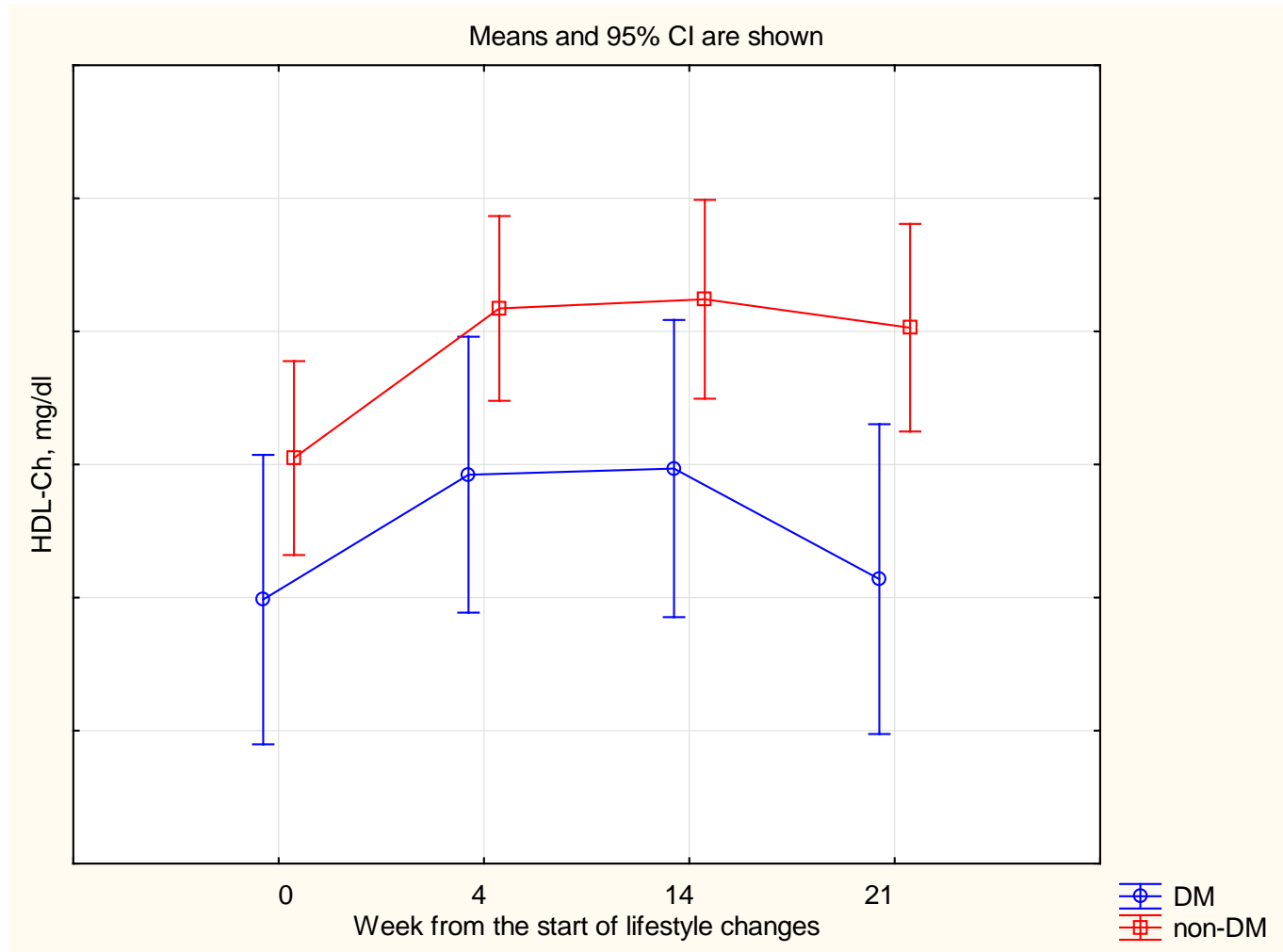
Figure S2 Comparison of the effect of lifestyle changes on total cholesterol in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on total cholesterol between DM and non-DM patients ($P = 0.705$).

SUPPLEMENTARY MATERIAL ONLINE

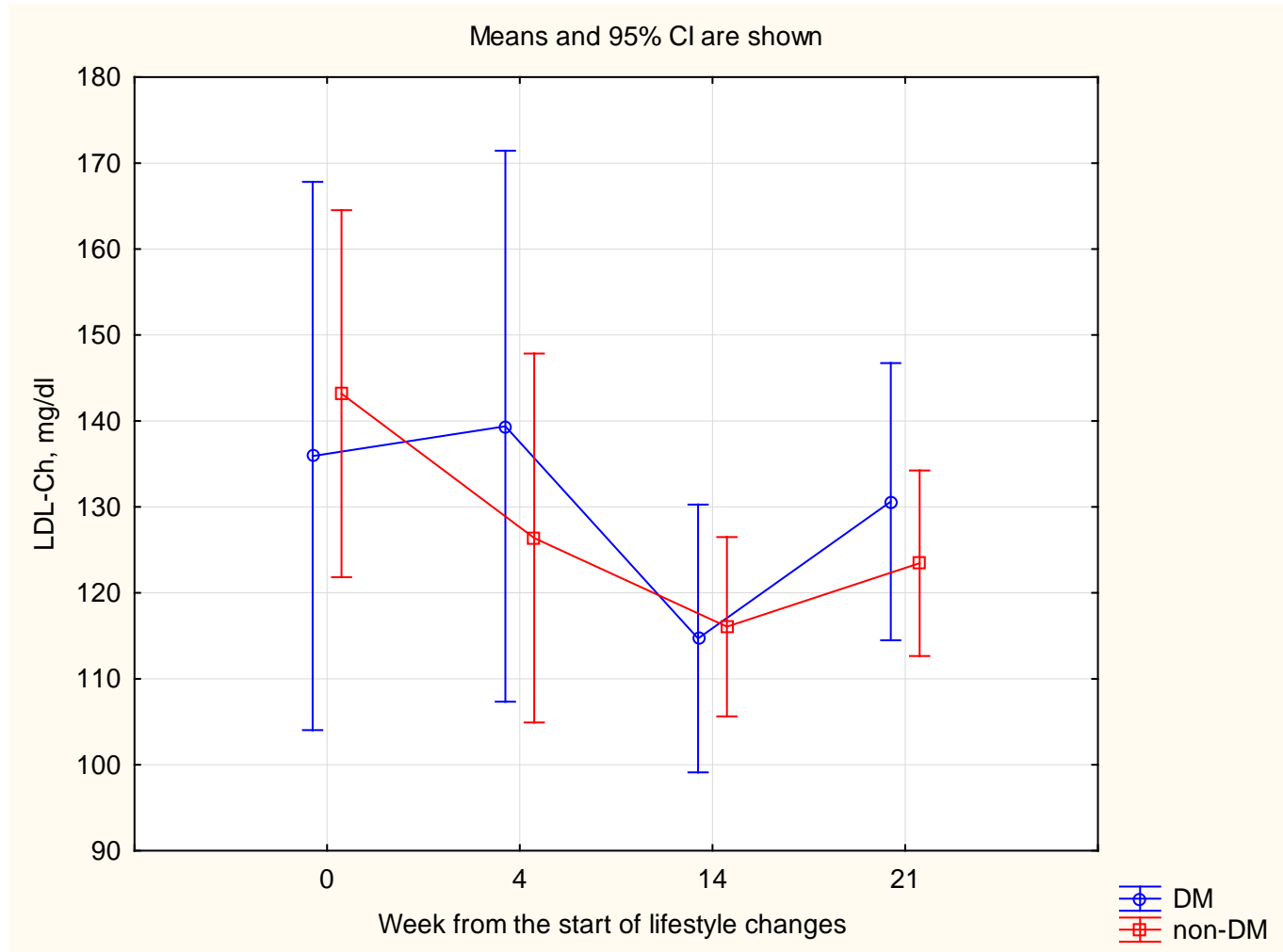
Figure S3 Comparison of the effect of lifestyle changes on high-density lipoprotein (HDL) cholesterol in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on HDL cholesterol between DM and non-DM patients ($P = 0.490$).

SUPPLEMENTARY MATERIAL ONLINE

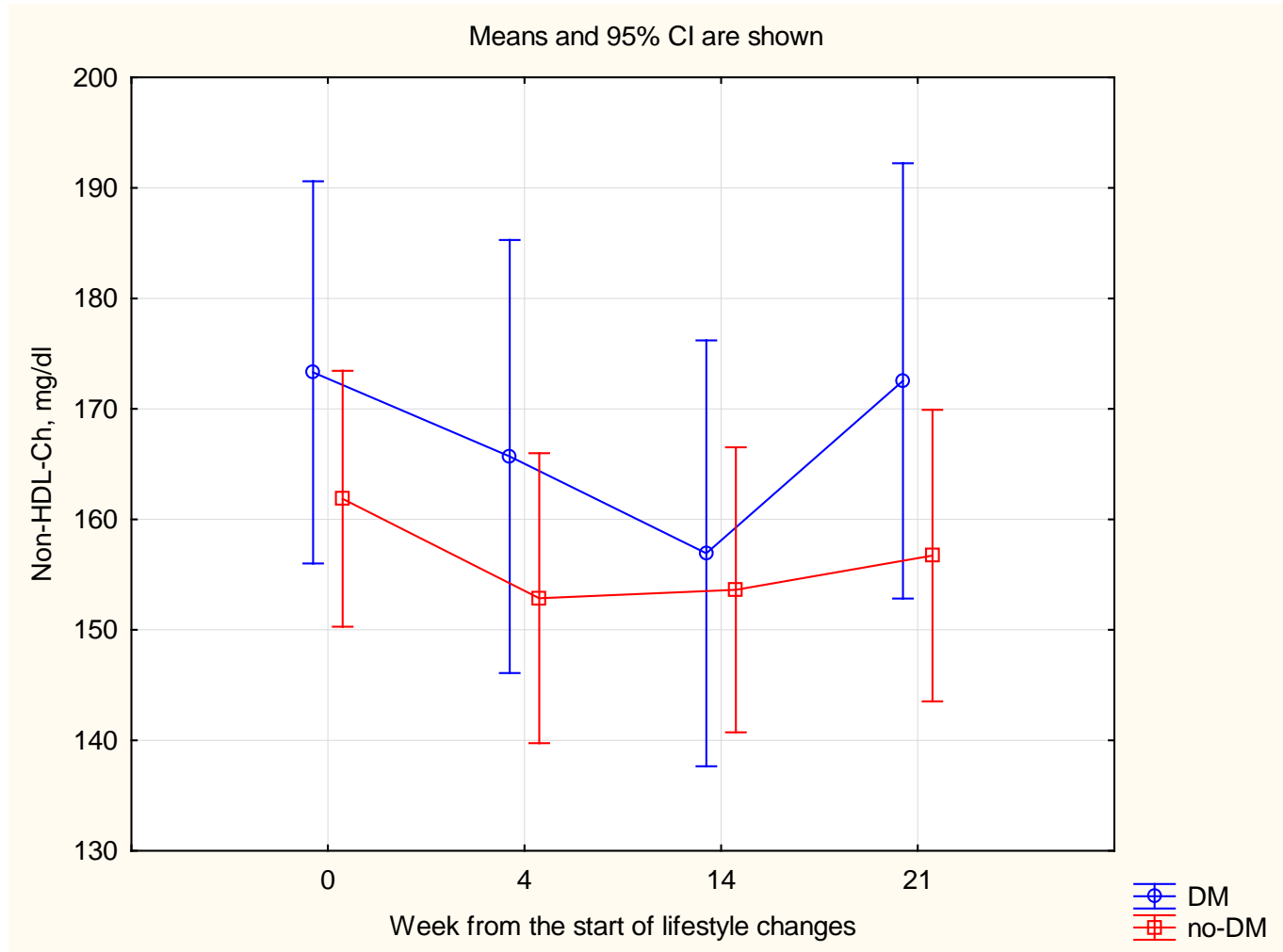
Figure S4 Comparison of the effect of lifestyle changes on low-density lipoprotein (LDL= cholesterol in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on LDL cholesterol between DM and non-DM patients ($P = 0.525$).

SUPPLEMENTARY MATERIAL ONLINE

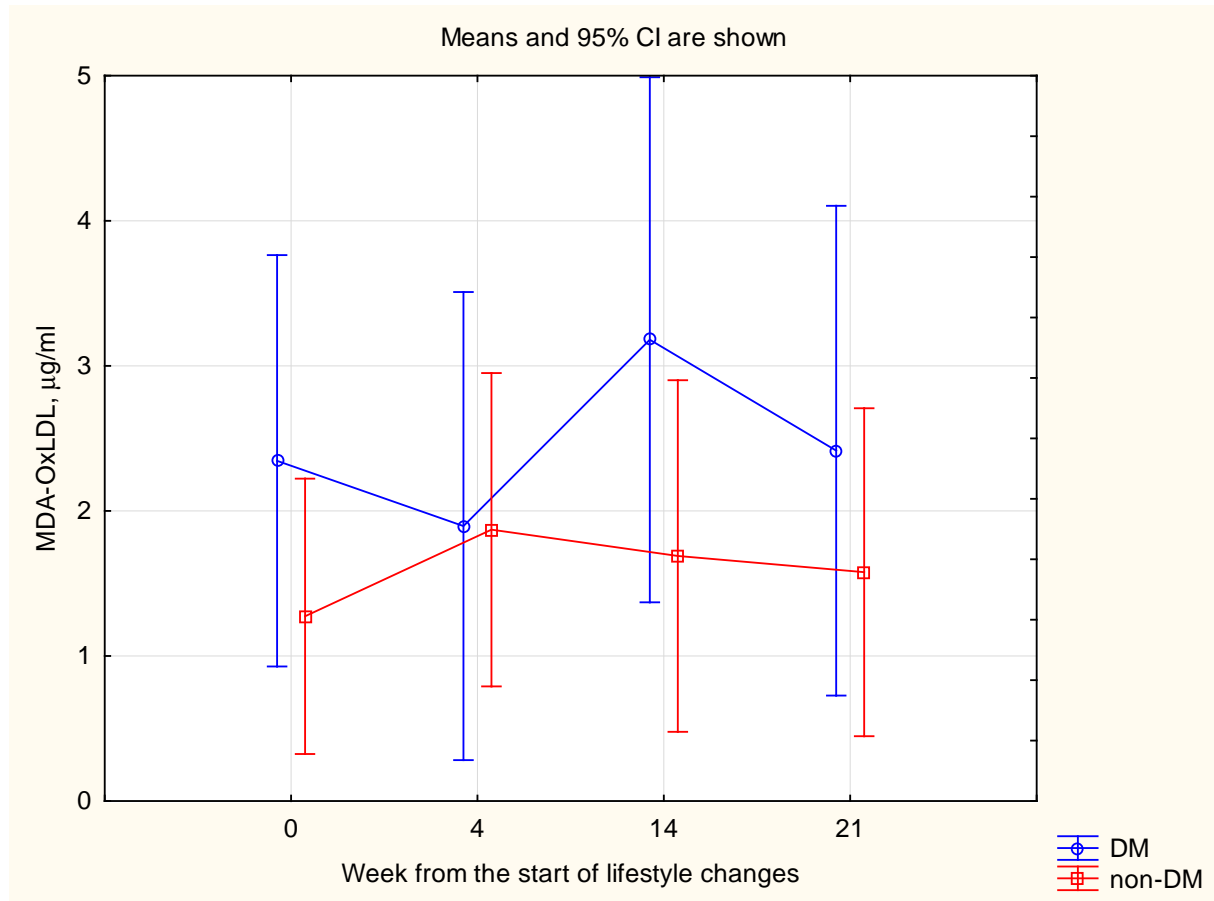
Figure S5 Comparison of the effect of lifestyle changes on non-high-density lipoprotein (non-HDL) cholesterol in dyslipidemic hemodialysis patients with diabetes (DM, $n = 13$) and without diabetes (non-DM, $n = 29$)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on non-HDL cholesterol between DM and non-DM patients in consecutive evaluations ($P = 0.296$).

SUPPLEMENTARY MATERIAL ONLINE

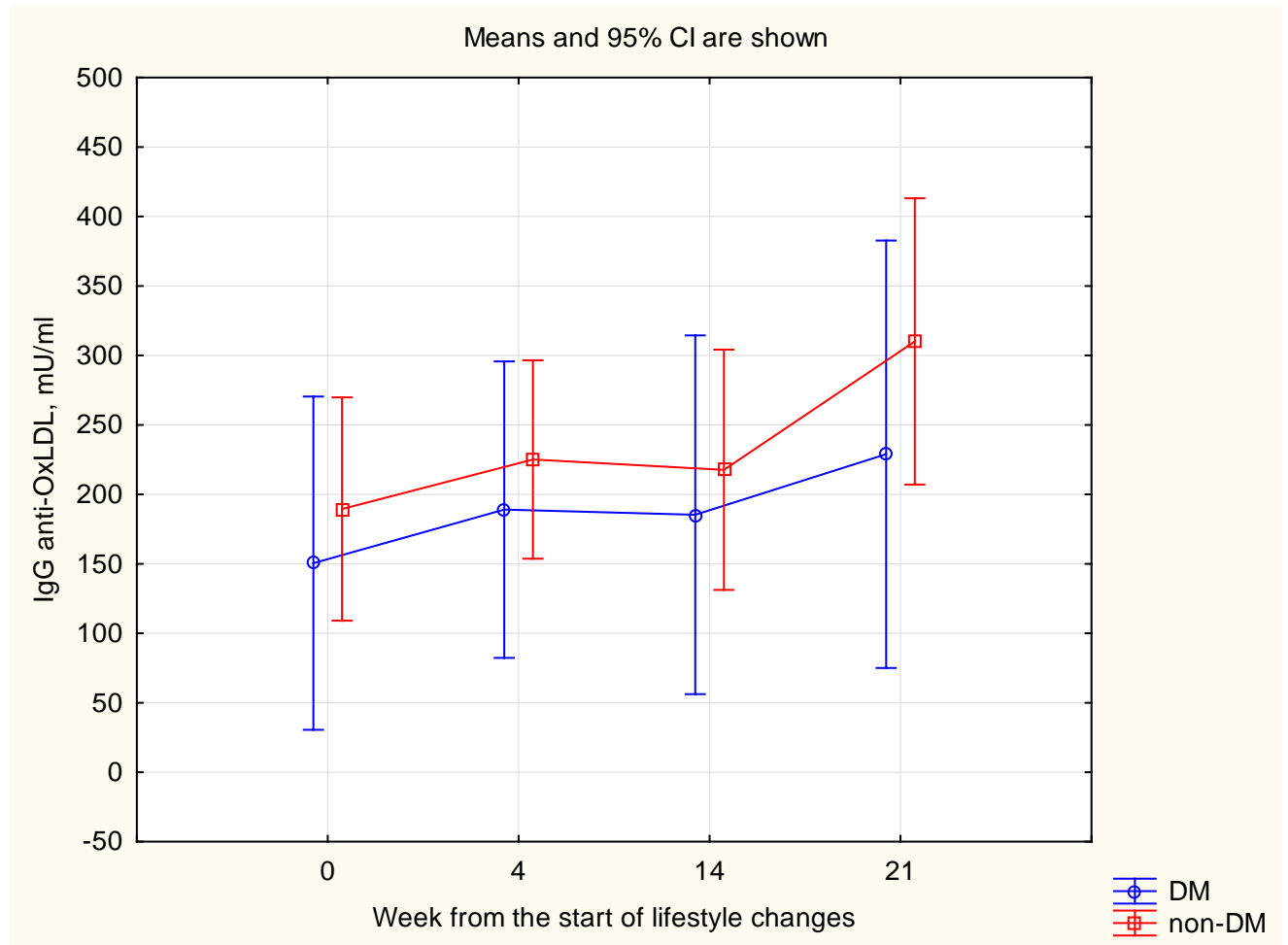
Figure S6 Comparison of the effect of lifestyle changes on malondialdehyde oxidized low-density lipoproteins (MDA-oxLDL) in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on MDA-oxLDL between DM and non-DM patients in consecutive evaluations ($P = 0.157$).

SUPPLEMENTARY MATERIAL ONLINE

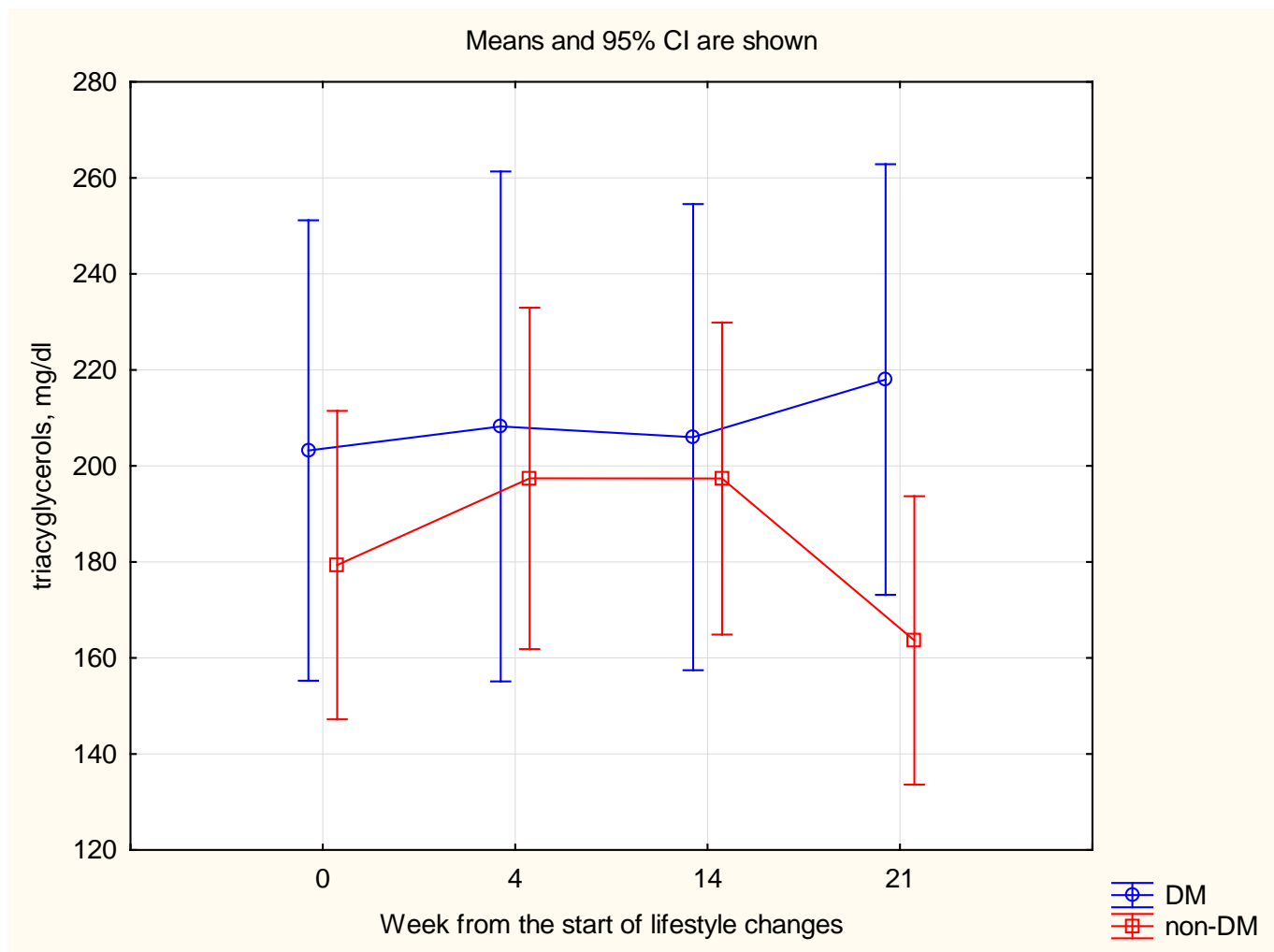
Figure S7 Comparison of the effect of lifestyle changes on immunoglobulin G antibodies to malondialdehyde oxidized low-density lipoproteins (IgG anti-oxLDL) in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on IgG anti-Ox-LDL between DM and non-DM patients in consecutive evaluations ($P = 0.737$).

SUPPLEMENTARY MATERIAL ONLINE

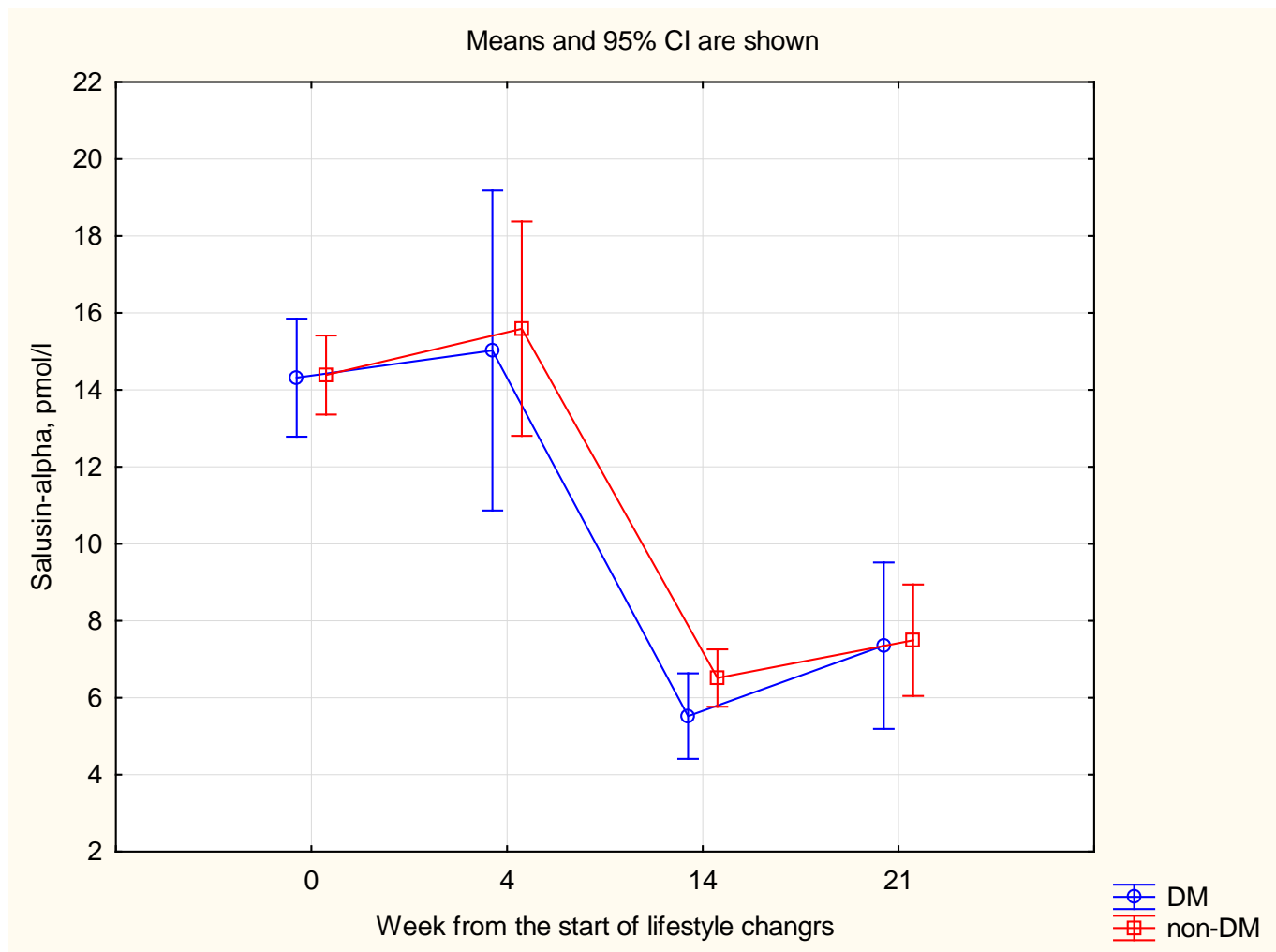
Figure S8 Comparison of the effect of lifestyle changes on triglycerides in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on triglycerides between DM and non-DM patients in consecutive evaluations ($P = 0.107$).

SUPPLEMENTARY MATERIAL ONLINE

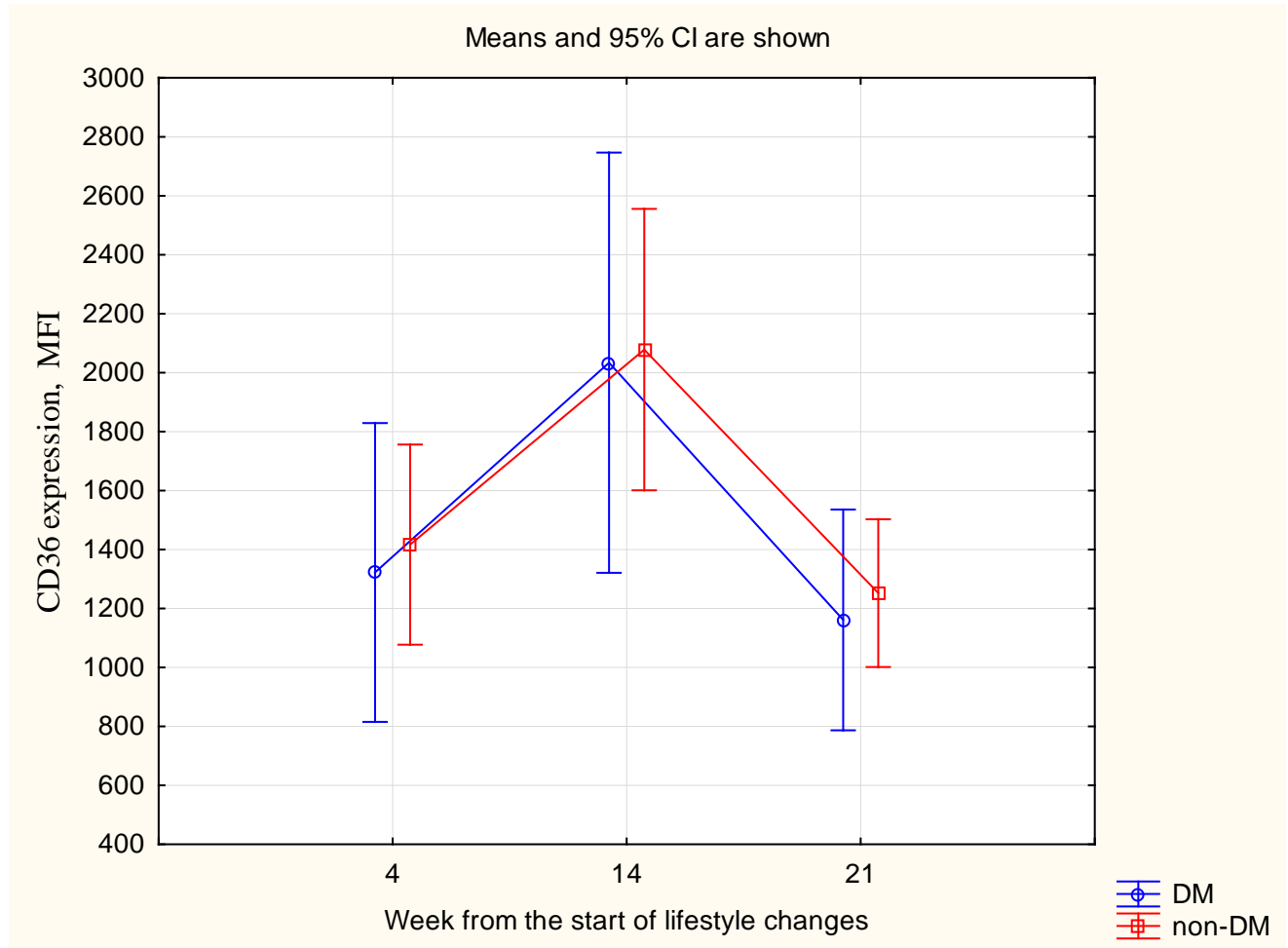
Figure S9 Comparison of the effect of lifestyle changes on salusin α in dyslipidemic hemodialysis patients with diabetes (DM, $n = 13$) and without diabetes (non-DM, $n = 29$)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on salusin α between DM and non-DM patients in consecutive evaluations ($P = 0.660$).

SUPPLEMENTARY MATERIAL ONLINE

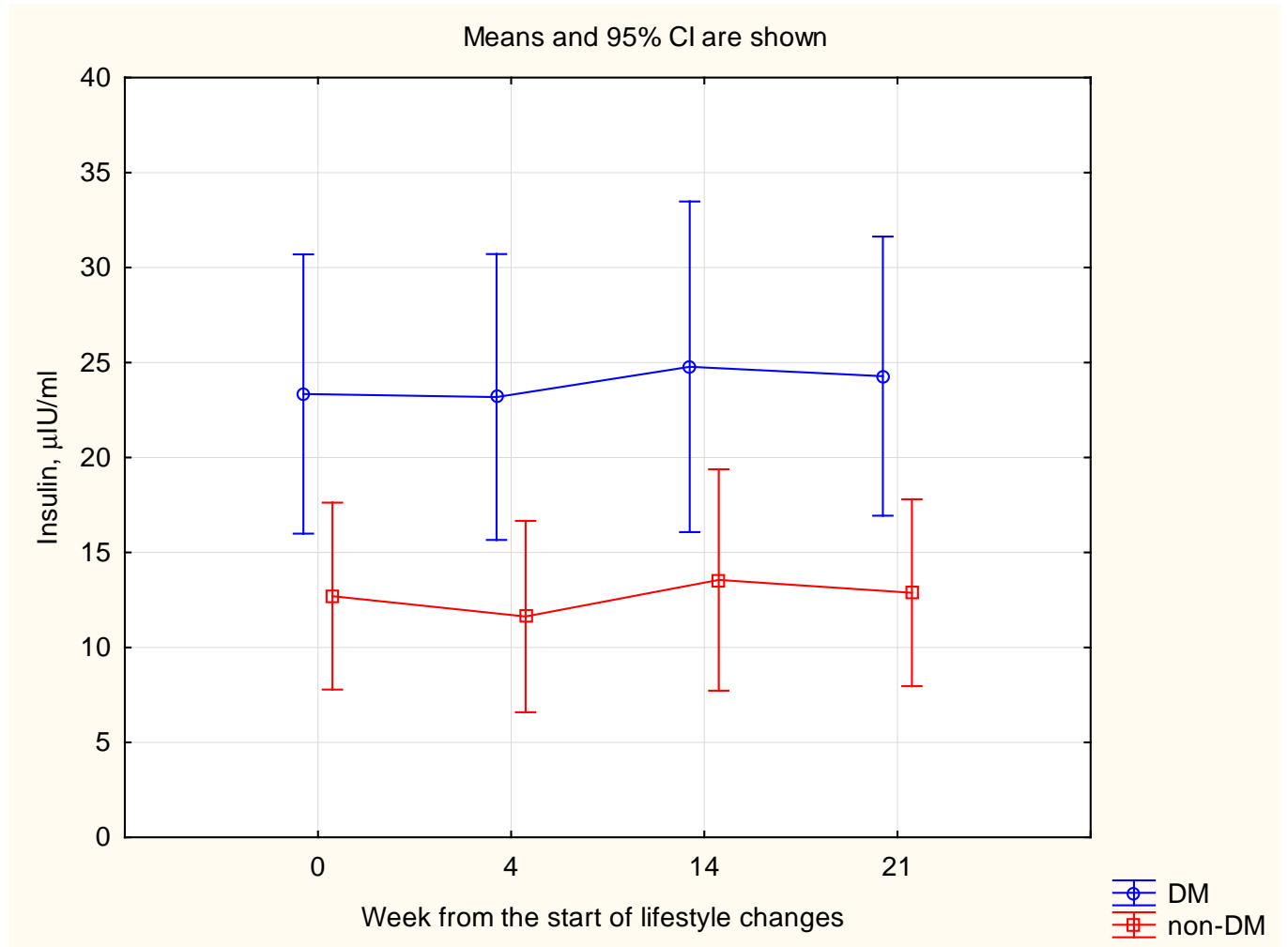
Figure S10 Comparison of the effect of lifestyle changes on CD36 expression (MFI - mean fluorescence intensity) in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on CD 36 expression (MFI) between DM and non-DM patients in consecutive evaluations ($P = 0.982$).

SUPPLEMENTARY MATERIAL ONLINE

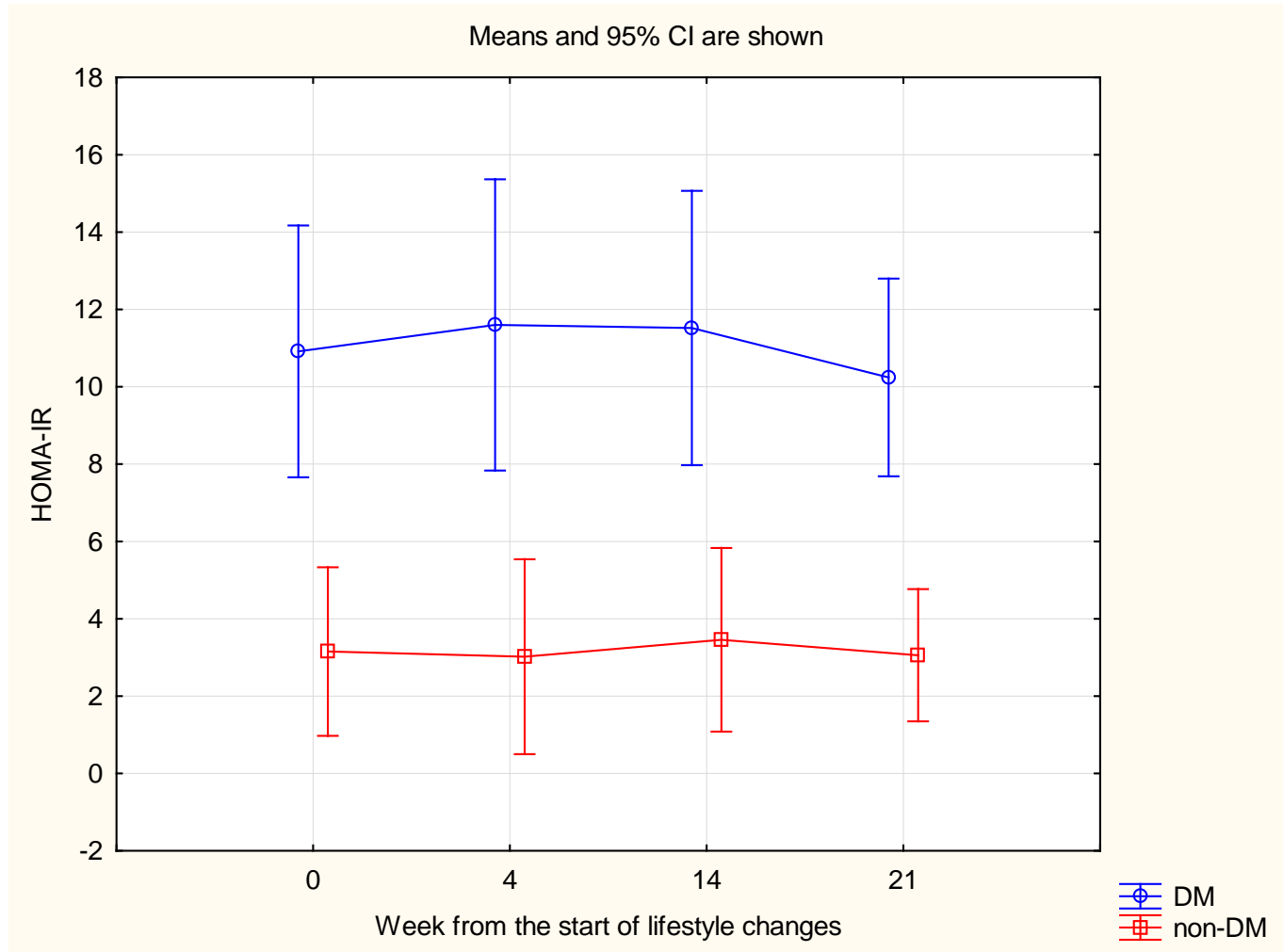
Figure S11 Comparison of the effect of lifestyle changes on insulin in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on insulin between DM and non-DM patients ($P = 0.995$).

SUPPLEMENTARY MATERIAL ONLINE

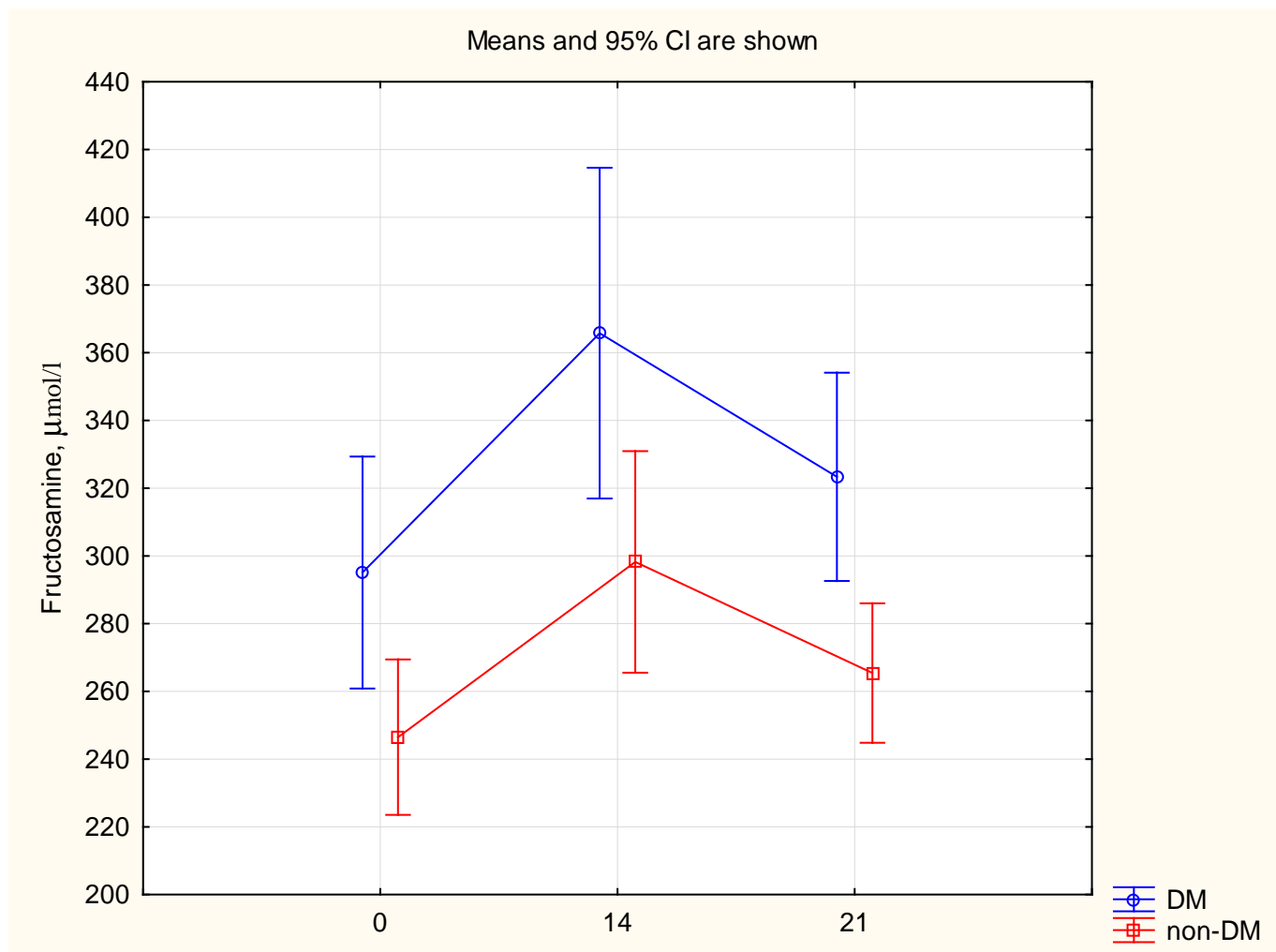
Figure S12 Comparison of the effect of lifestyle changes on homeostasis model assessment - insulin resistance (HOMA-IR) in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on HOMA-IR between DM and non-DM patients ($P = 0.893$).

SUPPLEMENTARY MATERIAL ONLINE

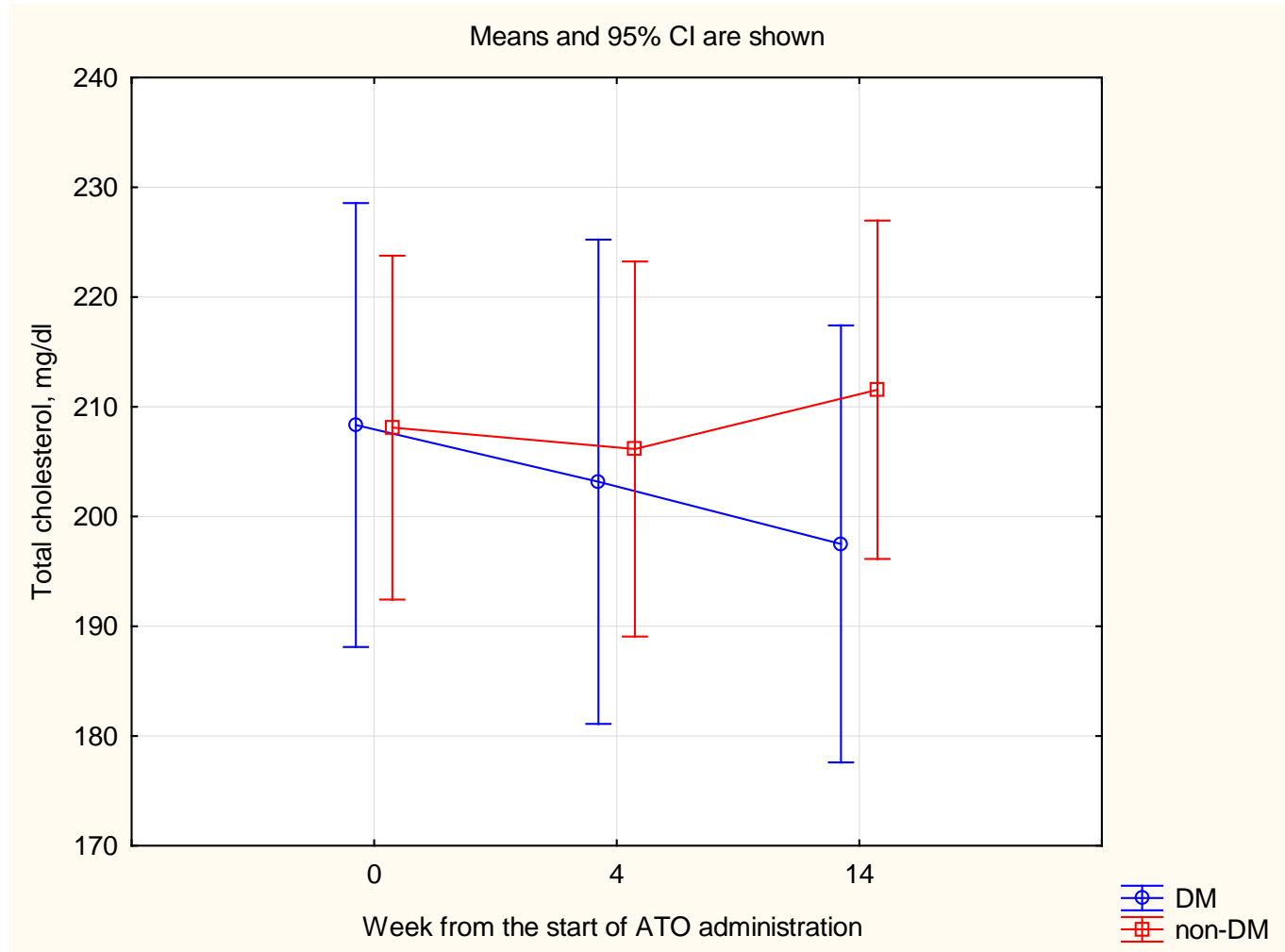
Figure S13 Comparison of the effect of lifestyle changes on fructosamine in dyslipidemic hemodialysis patients with diabetes (DM, n = 13) and without diabetes (non-DM, n = 29)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of lifestyle changes on fructosamine between DM and non-DM patients ($P = 0.781$).

SUPPLEMENTARY MATERIAL ONLINE

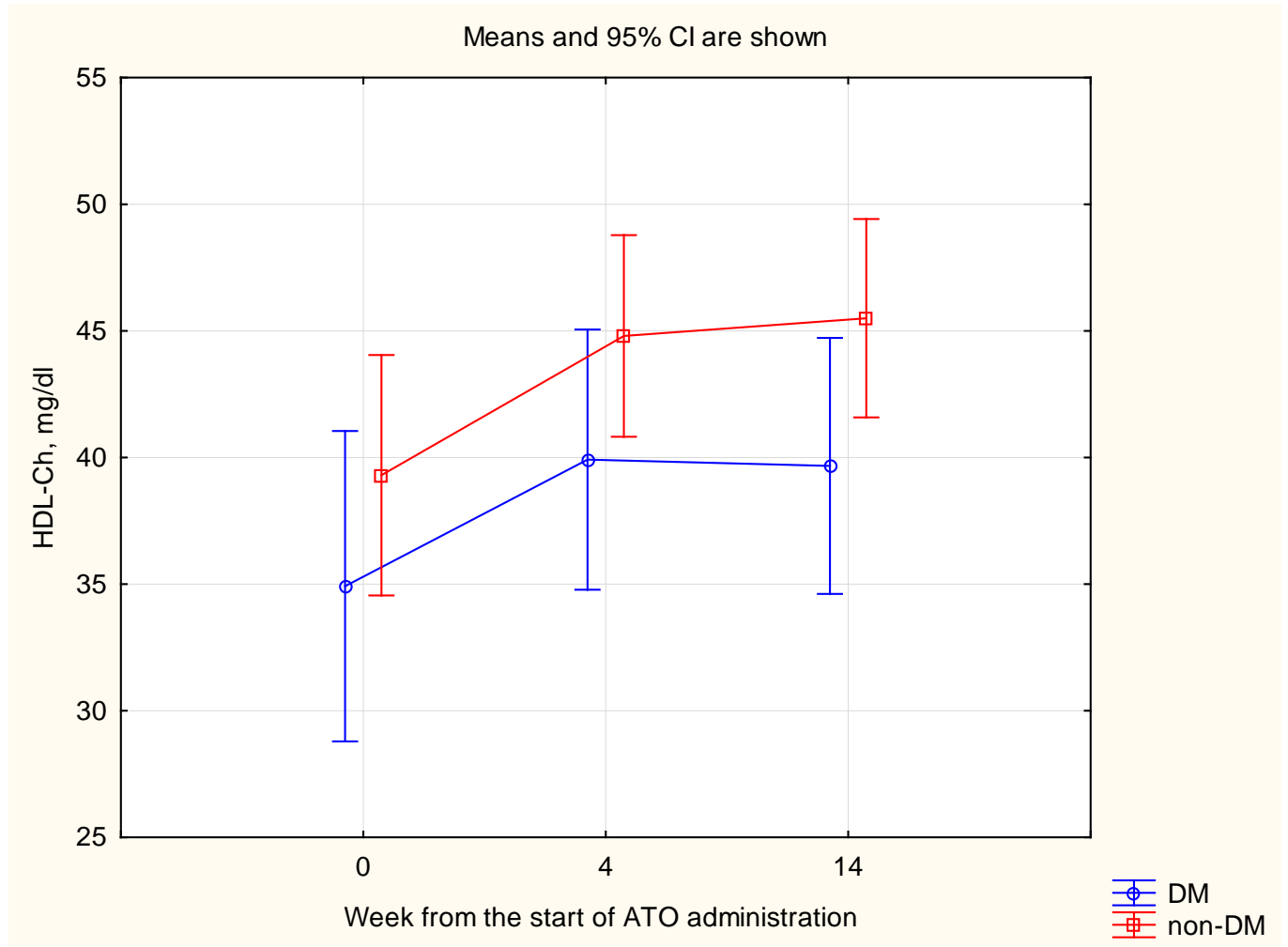
Figure S14 Comparison of the effect of atorvastatin (ATO) on total cholesterol in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of atorvastatin (ATO) on total cholesterol between DM and non-DM patients in consecutive evaluations ($P = 0.301$).

SUPPLEMENTARY MATERIAL ONLINE

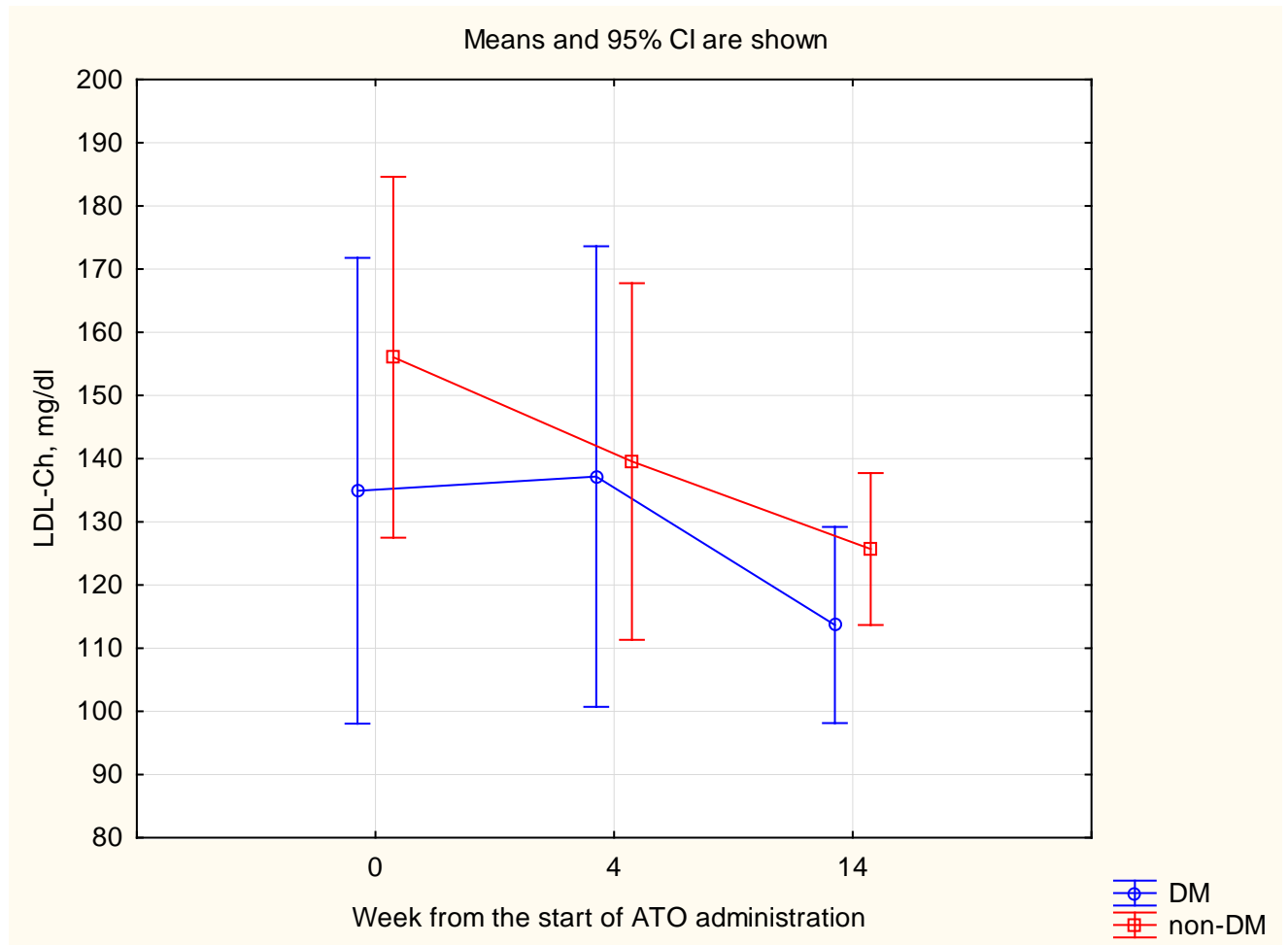
Figure S15 Comparison of the effect of atorvastatin (ATO) on high density lipoprotein cholesterol (HDL-Ch) in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show a significant difference in the effect of atorvastatin (ATO) on HDL-Ch between DM and non-DM patients in consecutive evaluations ($P = 0.890$).

SUPPLEMENTARY MATERIAL ONLINE

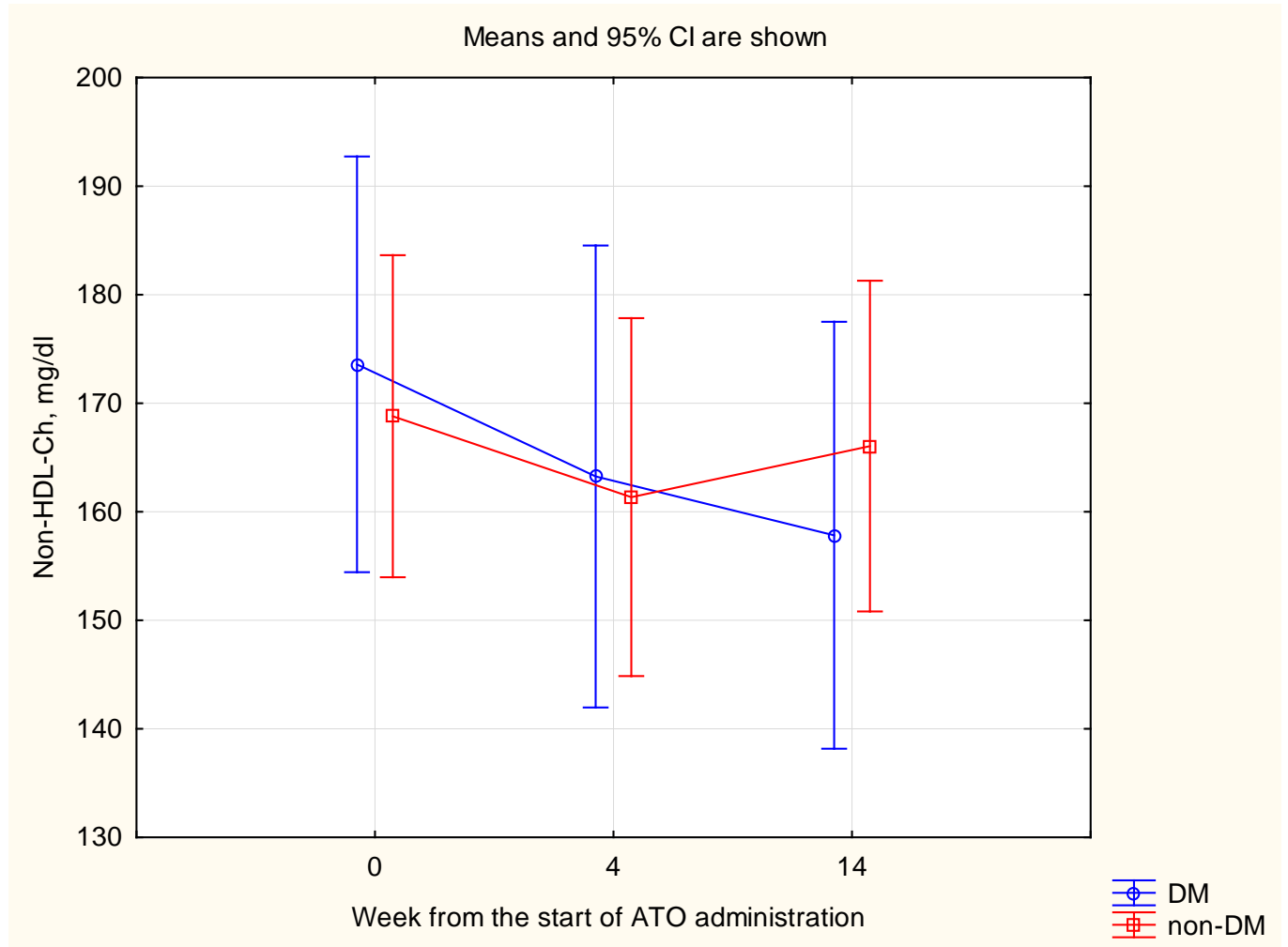
Figure S16 Comparison of the effect of atorvastatin (ATO) on low density lipoprotein cholesterol (LDL-Ch) in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on LDL-Ch between DM and non-DM patients in consecutive evaluations ($P = 0.827$).

SUPPLEMENTARY MATERIAL ONLINE

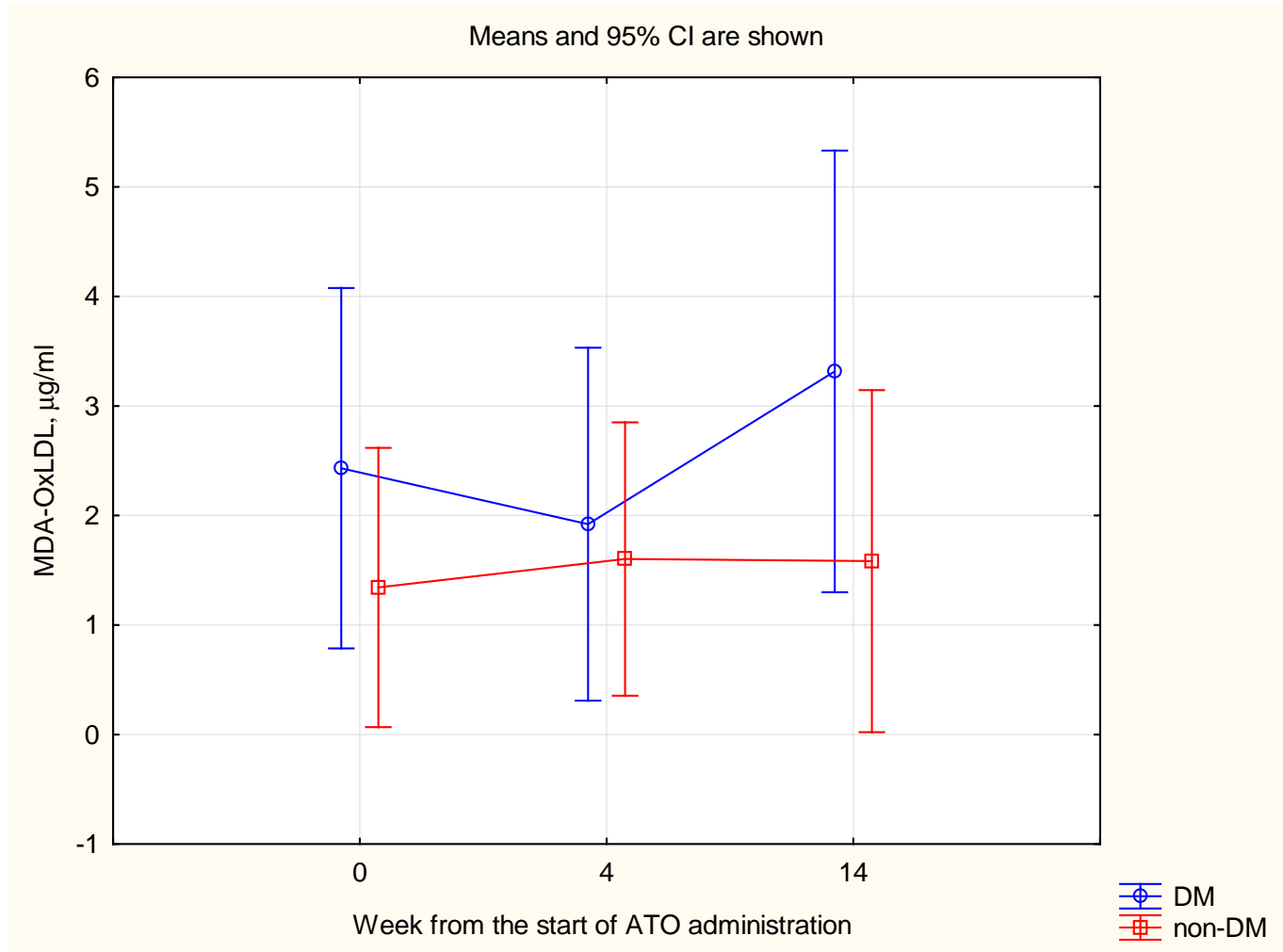
Figure S17 Comparison of the effect of atorvastatin (ATO) on non-high density lipoprotein cholesterol (non-HDL-Ch) in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on non-HDL-Ch between DM and non-DM patients in consecutive evaluations ($P = 0.365$).

SUPPLEMENTARY MATERIAL ONLINE

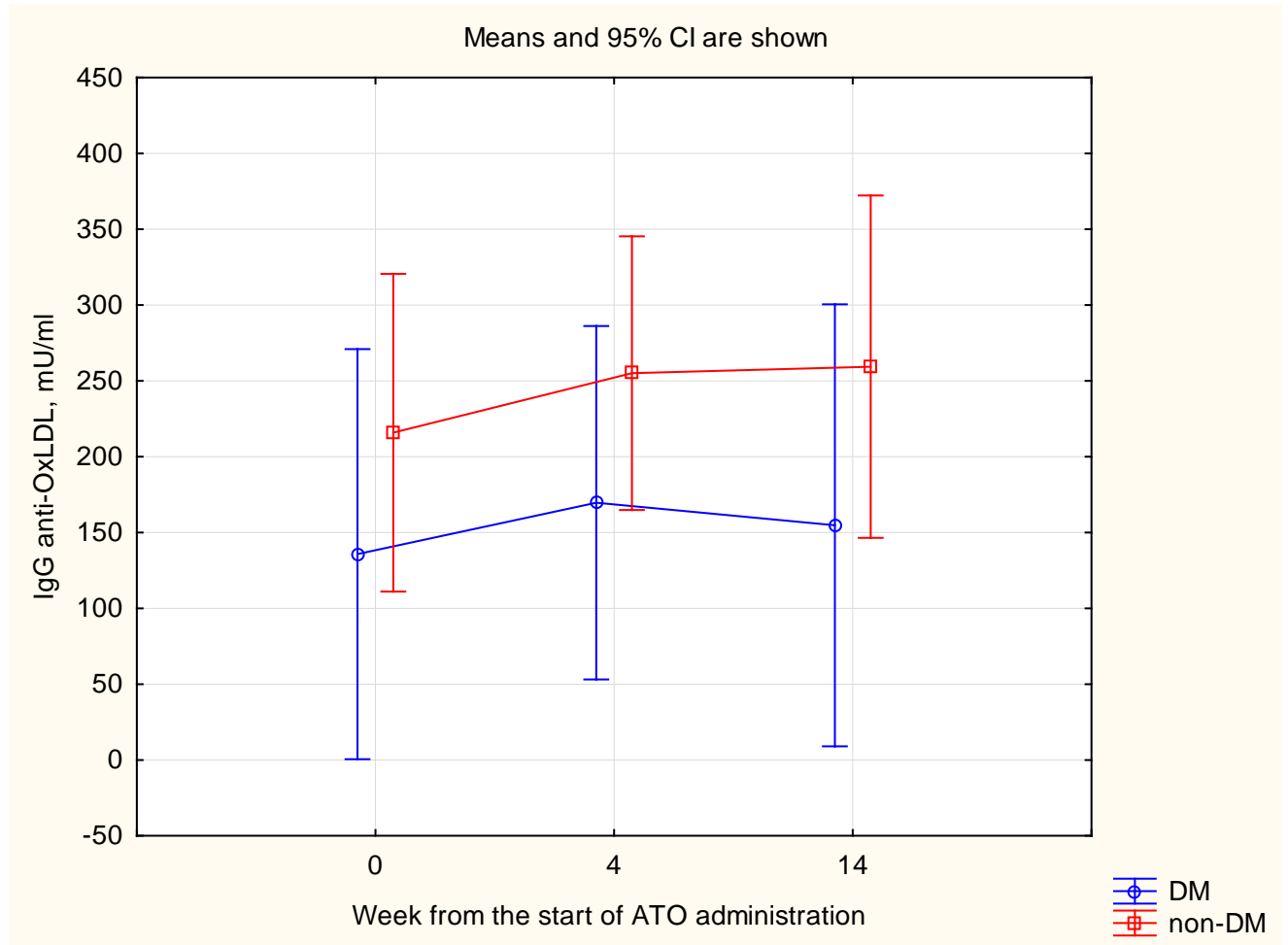
Figure S18 Comparison of the effect of atorvastatin (ATO) on malondialdehyde oxidized low density lipoproteins (MDA-oxLDL) in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on MDA-Ox-LDL between DM and non-DM patients in consecutive evaluations ($P = 0.150$).

SUPPLEMENTARY MATERIAL ONLINE

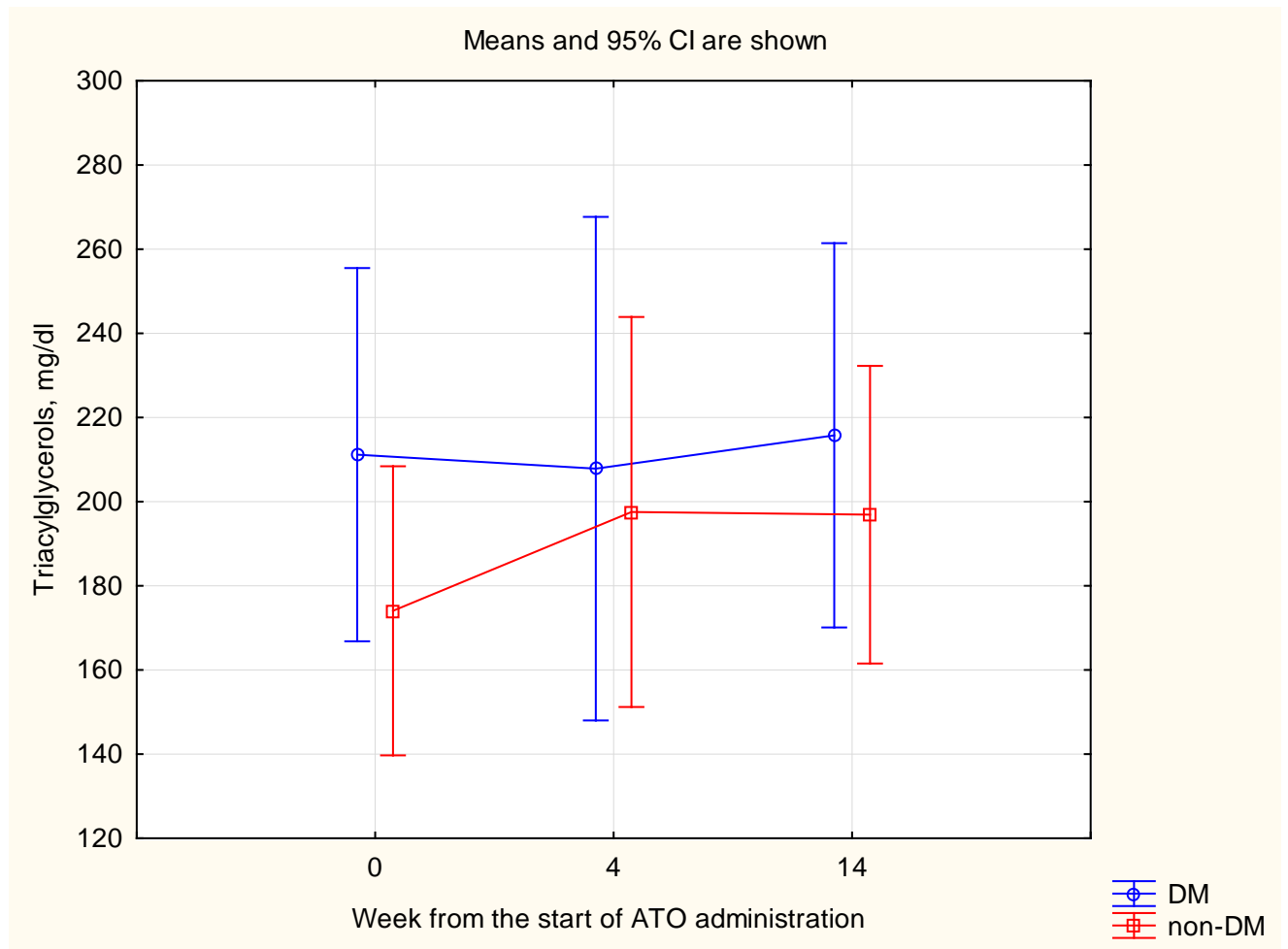
Figure S19 Comparison of the effect of atorvastatin (ATO) on immunoglobulin G antibodies to malondialdehyde oxidized low density lipoproteins (IgG anti-oxLDL) in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on IgG anti-Ox-LDL between DM and non-DM patients in consecutive evaluations ($P = 0.606$).

SUPPLEMENTARY MATERIAL ONLINE

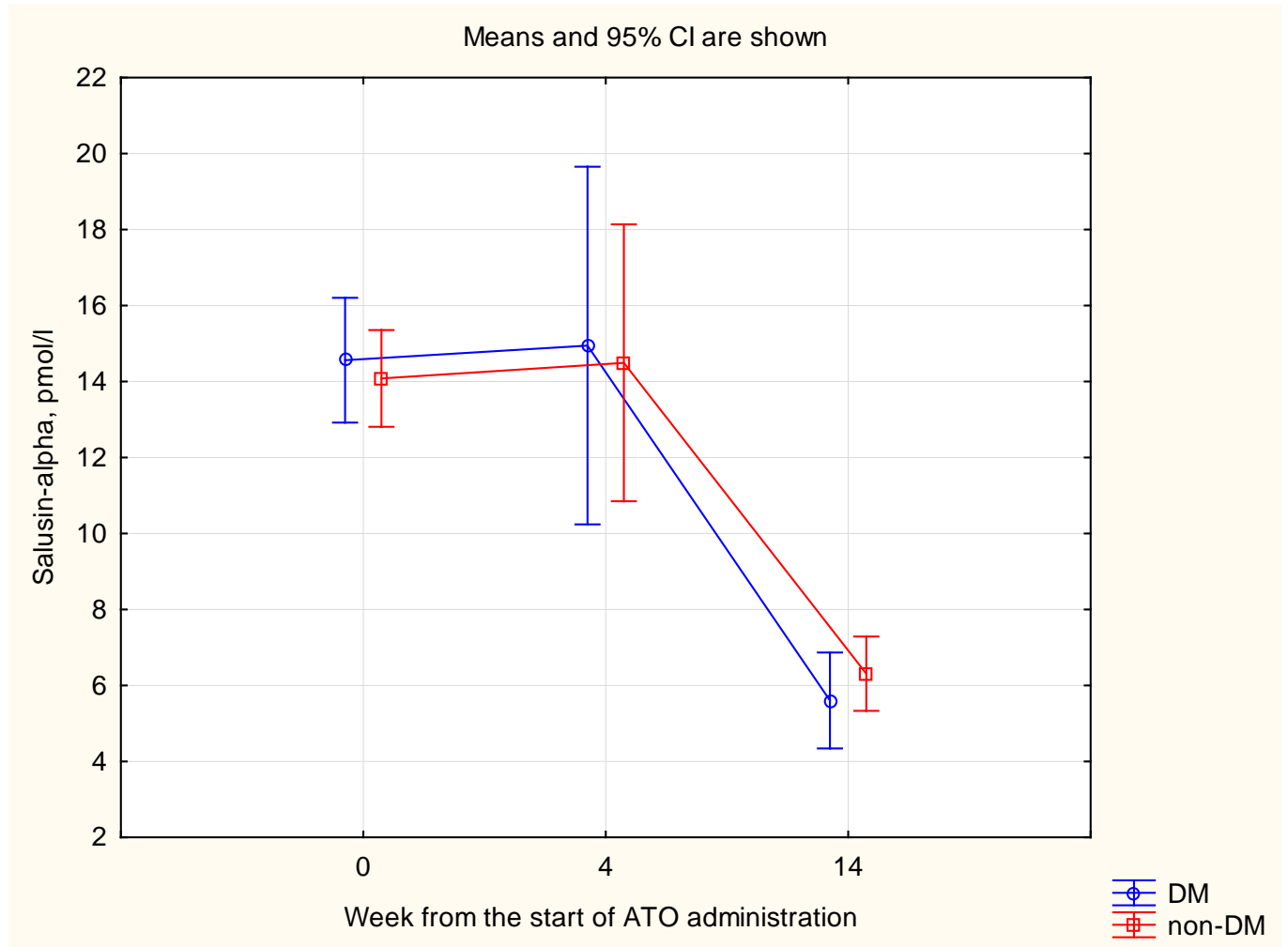
Figure S20 Comparison of the effect of atorvastatin (ATO) on triglicerydes in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on triglicerydes between DM and non-DM patients in consecutive evaluations ($P = 0.722$).

SUPPLEMENTARY MATERIAL ONLINE

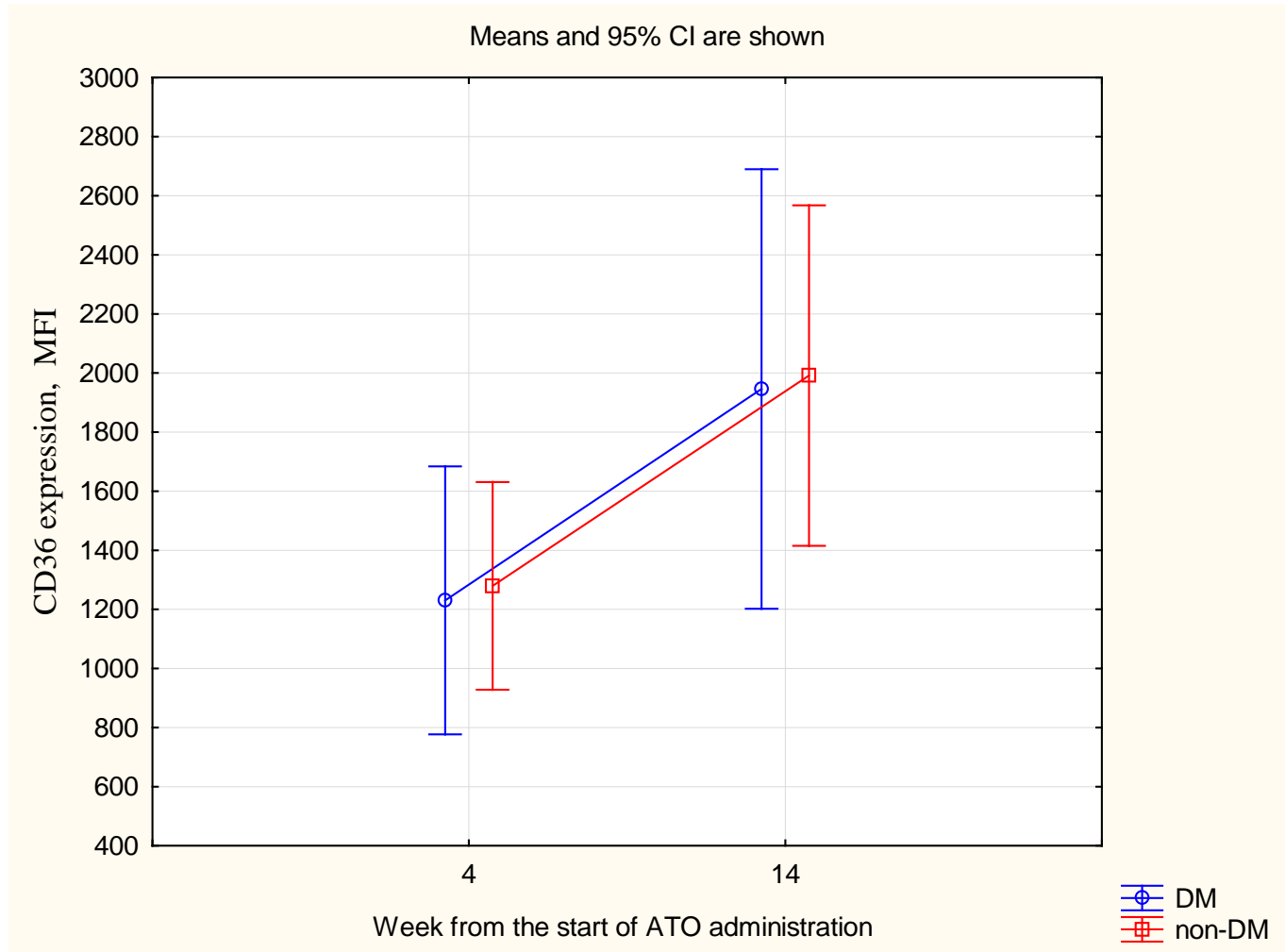
Figure S21 Comparison of the effect of atorvastatin (ATO) on *salusin α* in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on *salusin α* between DM and non-DM patients in consecutive evaluations ($P = 0.503$).

SUPPLEMENTARY MATERIAL ONLINE

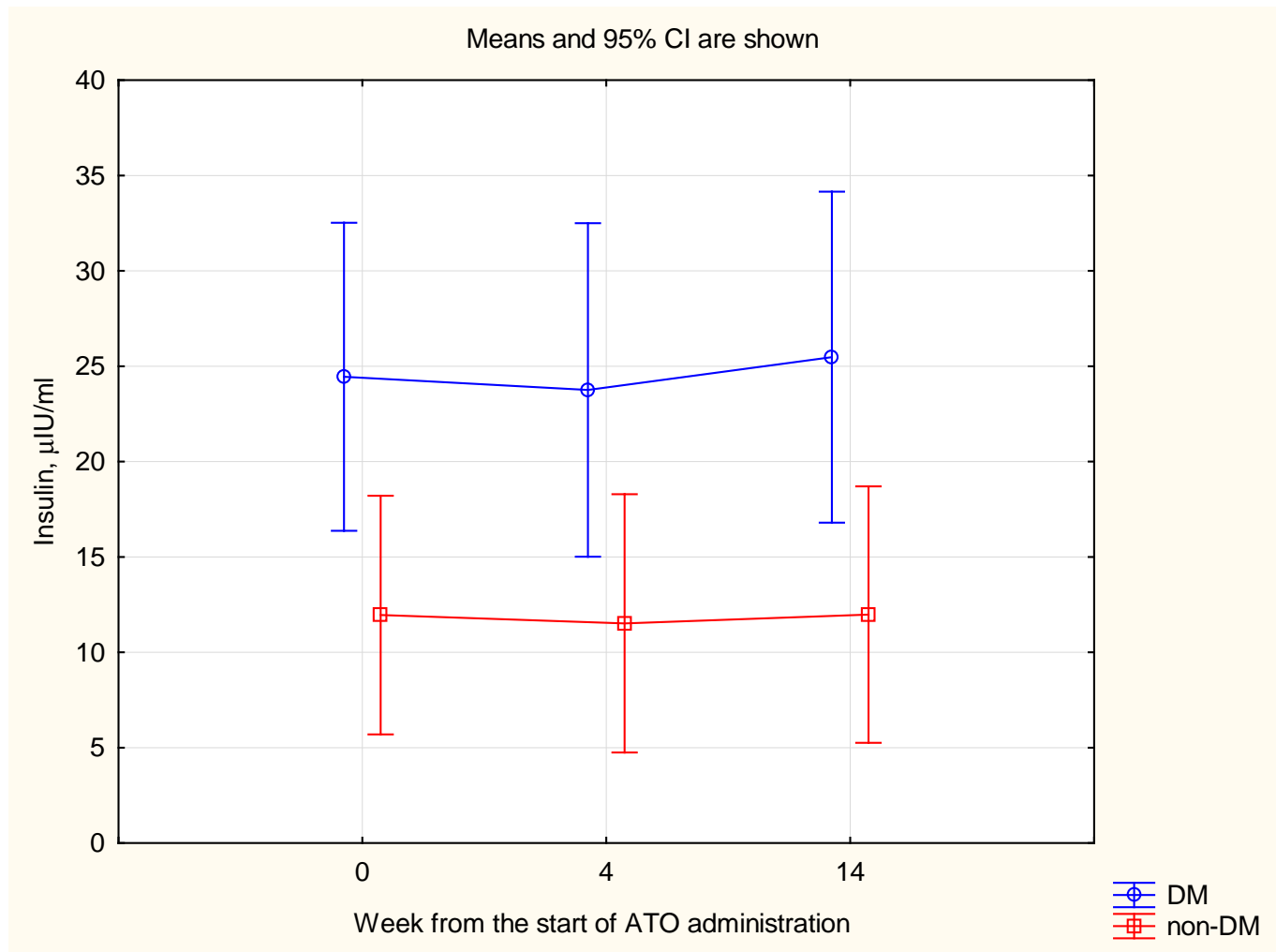
Figure S22 Comparison of the effect of atorvastatin (ATO) on CD36 expression (MFI - mean fluorescence intensity) in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on CD36 expression (MFI) between DM and non-DM patients in consecutive evaluations ($P = 0.988$).

SUPPLEMENTARY MATERIAL ONLINE

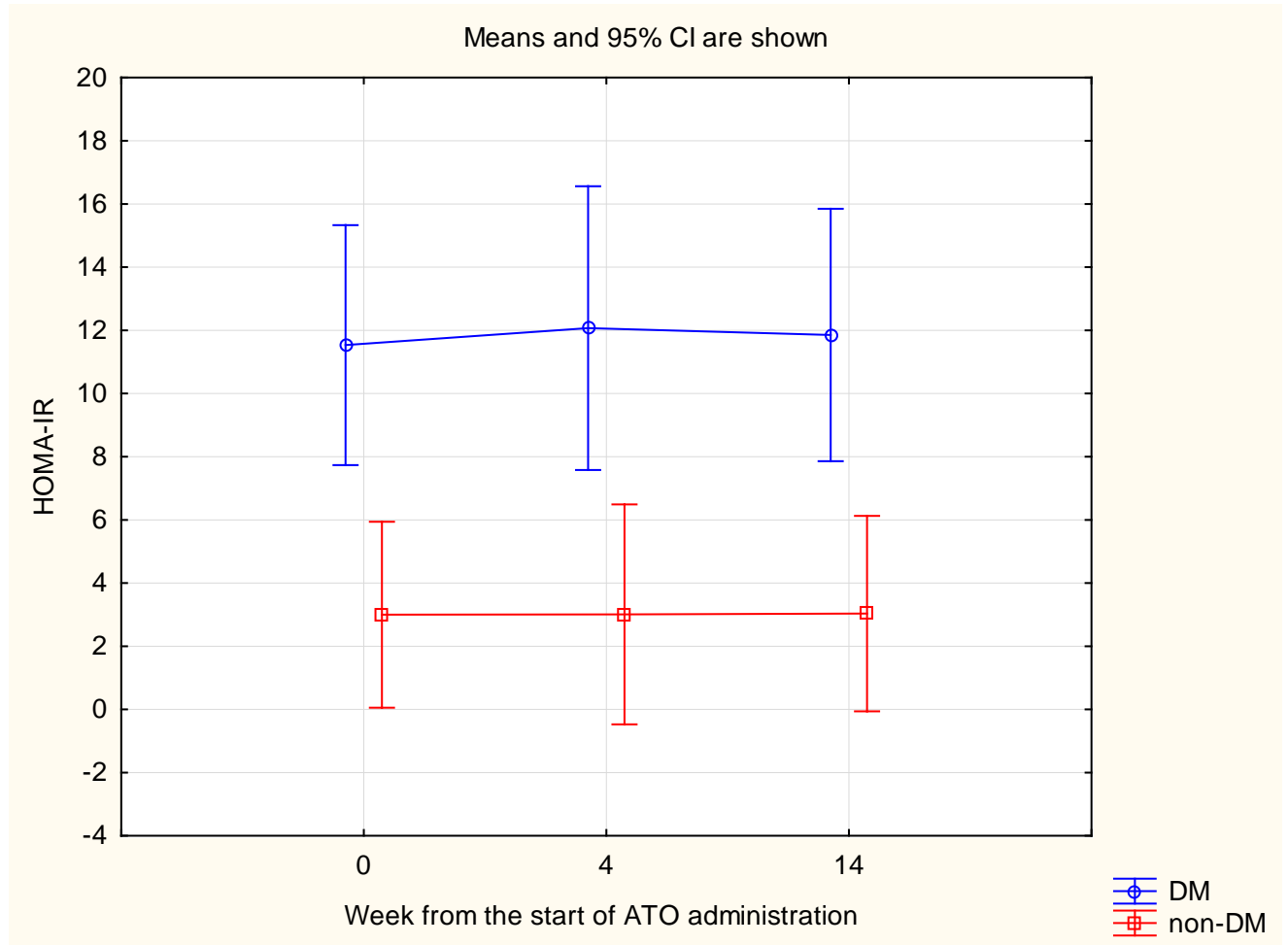
Figure S23 Comparison of the influence of atorvastatin (ATO) on insulin in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on insulin between DM and non-DM patients in consecutive evaluations ($P = 0.803$).

SUPPLEMENTARY MATERIAL ONLINE

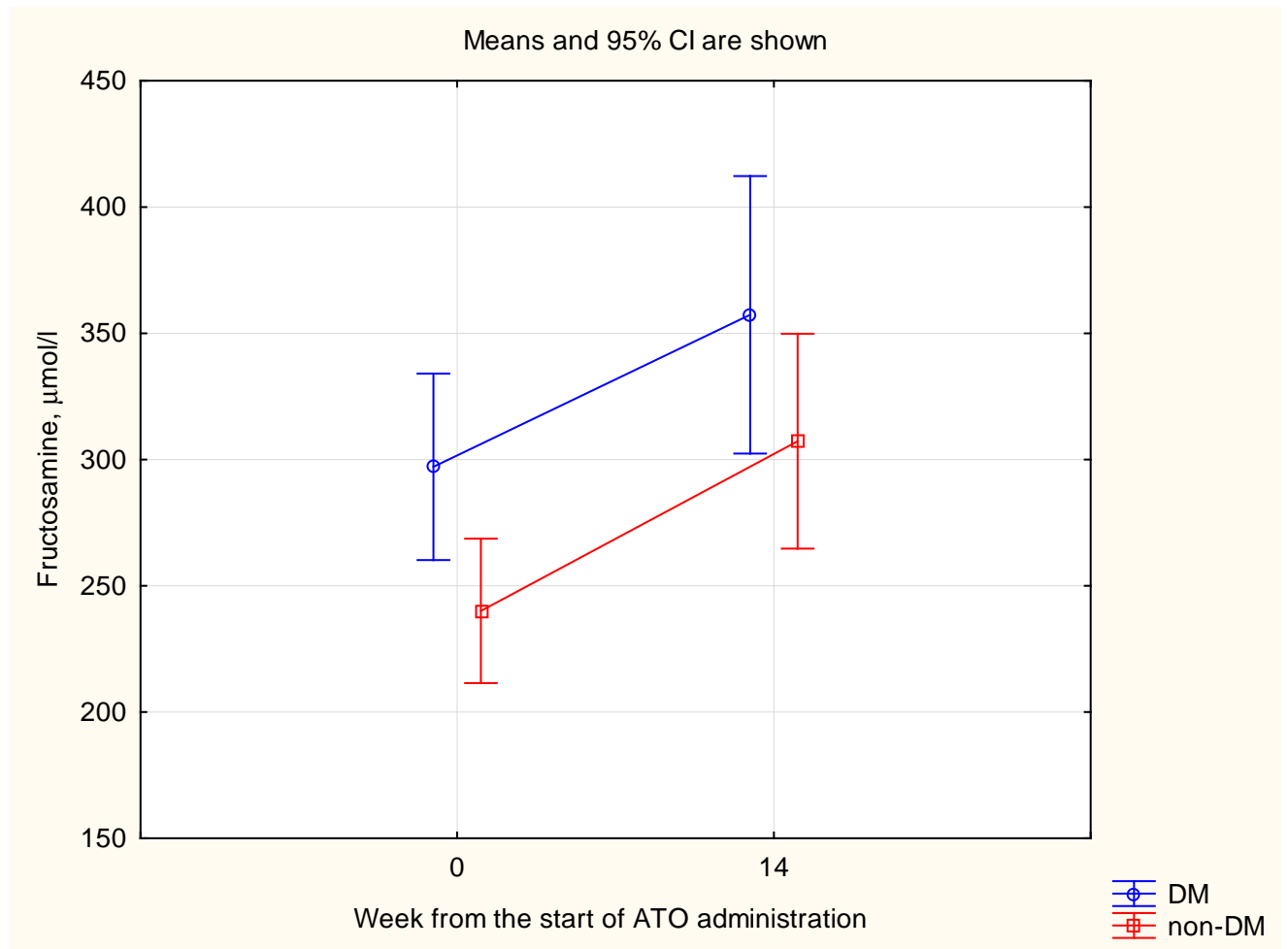
Figure S24 Comparison of the effect of atorvastatin (ATO) on homeostasis model assessment - insulin resistance (HOMA-IR) in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on HOMA-IR between DM and non-DM patients in consecutive evaluations ($P = 0.951$).

SUPPLEMENTARY MATERIAL ONLINE

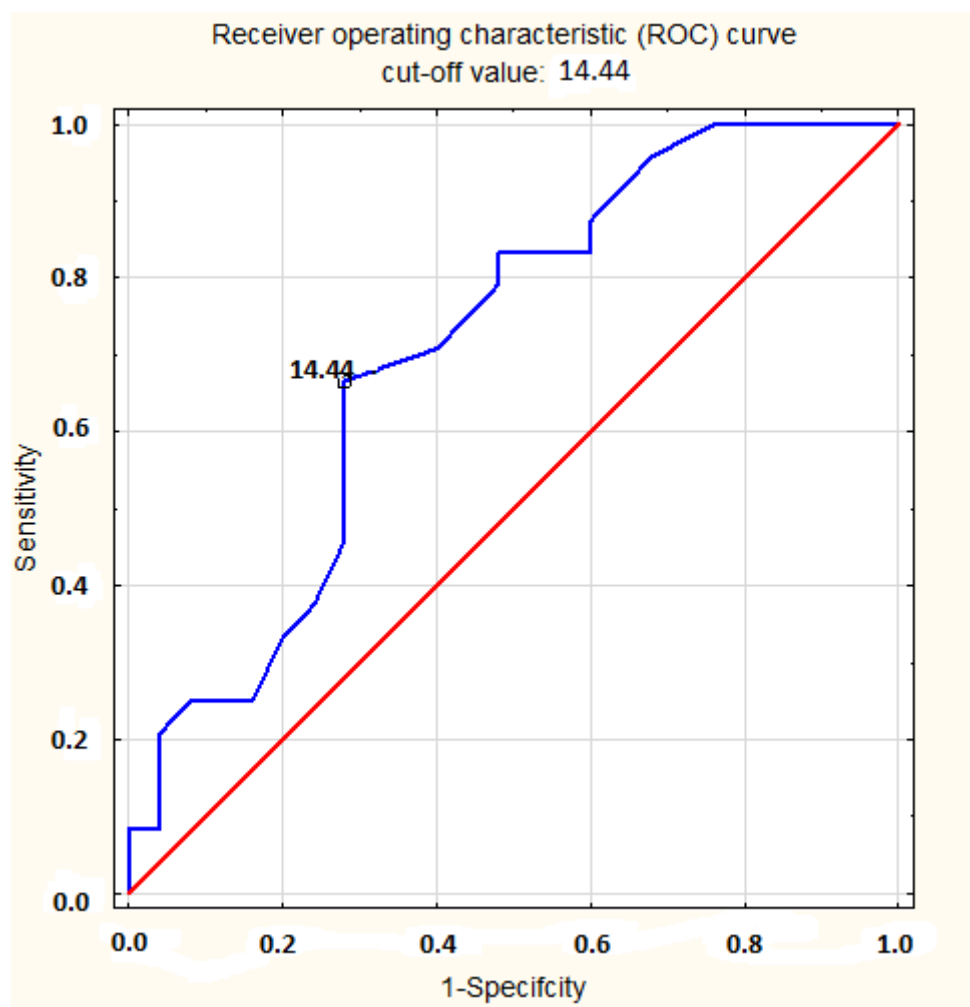
Figure S25 Comparison of the effect of atorvastatin (ATO) on fructosamine in dyslipidemic hemodialysis patients with diabetes (DM, n = 12) and without diabetes (non-DM, n = 20)



A general linear model multi-way repeated measures analysis did not show significant differences in the effect of atorvastatin (ATO) on fructosamine between DM and non-DM patients in consecutive evaluations ($P = 0.813$).

SUPPLEMENTARY MATERIAL ONLINE

Figure S26 Receiver operating characteristic curve methodology indicated that a salusin α level of 14.4 pmol/l is a cut-off value in the prognosis of the serum low-density lipoprotein cholesterol concentration below or over the lower limit of low-density lipoprotein cholesterol values designated as high borderline (130–159 mg/dl).



	AUC	SE	Lower 95% CI	Upper 95% CI
1	0.709	0.074	0.564	0.855

AUC – area under the curve

CI - confidence interval

SE – standard error

Wpływ zmian stylu życia i stosowania atorwastatyny na dyslipidemię u hemodializowanych chorych – badanie prospektywne

Alicja E. Grzegorzewska¹, Leszek Niepolski², Jan Sikora³,
Małgorzata Janków⁴, Paweł P. Jagodziński⁵, Anna Sowińska⁶

¹ Katedra i Klinika Nefrologii, Transplantologii i Chorób Wewnętrznych, Uniwersytet Medyczny im. Karola Marcinkowskiego w Poznaniu, Poznań

² Stacja Dializ B. Braun Avitum, Nowy Tomyśl

³ Zakład Immunologii, Katedra Immunologii Klinicznej, Uniwersytet Medyczny im. Karola Marcinkowskiego w Poznaniu, Poznań

⁴ Stacja Dializ Eurodial, Ostrów Wielkopolski

⁵ Katedra i Zakład Biochemii i Biologii Molekularnej, Uniwersytet Medyczny im. Karola Marcinkowskiego w Poznaniu, Poznań

⁶ Katedra i Zakład Informatyki i Statystyki, Uniwersytet Medyczny im. Karola Marcinkowskiego w Poznaniu, Poznań

SŁOWA KLUCZOWE

atorwastatyna,
ekspresja CD36,
hemodializa,
salusyna α , zmiany
stylu życia

STRESZCZENIE

WPROWADZENIE Aterogenność dyslipidemia przyspiesza rozwój powikłań sercowo-naczyniowych i przyczynia się do śmiertelności hemodializowanych (HD) chorych.

CELE Celem badania była ocena wpływu zmian stylu życia i stosowania atorwastatyny na dyslipidemię u HD chorych.

PACJENCI I METODY HD chorzy wykazujący dyslipidemię ($n = 49$) zostali włączeni do badania prospektywnego. Czterdziestu dwóch badanych ukończyło 21-tygodniową interwencję polegającą na zmianach w stylu życia. U 34 osób nadal wykazujących dyslipidemię zastosowano atorwastatinę przez okres 14 tygodni. Po 4 tygodniach zwiększono początkową dawkę atorwastatyny z 10 mg/dobę do 20 mg/dobę u chorych wykazujących nadal dyslipidemię.

WYNIKI Najwyraźniejsze skutki zmian stylu życia wykazano po 14 tygodniach od ich początku. Obejmowały one istotne różnice w stężeniu cholesterolu frakcji lipoprotein o dużej gęstości, cholesterolu frakcji lipoprotein o małej gęstości (*low-density lipoprotein* – LDL), salusyny α , MDA-OxLDL i fruktozaminy, a także w ekspresji CD36 na monocytach krwi. IgG anty-OxLDL wykazały najwyższe stężenia w 21. tygodniu. U 7 chorych (16,7%) nie stwierdzono dyslipidemii w 21. tygodniu badania. U chorych wykazujących nadal dyslipidemię włączenie i podawanie atorwastatyny wiązało się z istotnym obniżeniem stężenia cholesterolu LDL i triglicerydów, zwiększeniem stężenia salusyny α i ekspresji CD36 oraz ustąpieniem dyslipidemii u 59,4% chorych.

WNIOSKI Zmiany stylu życia wykazują ograniczoną skuteczność w leczeniu dyslipidemii u HD chorych, podczas gdy atorwastatyna (w dawce do 20 mg/dobę) może być skuteczna u około 60% chorych nieodpowiadających na zmiany stylu życia. Interwencje mające na celu obniżenie stężenia lipidów wpływają na stężenie salusyny α w osoczu oraz na ekspresję CD36 na monocytach.

Adres do korespondencji:
prof. dr hab. n. med. Alicja E.
Grzegorzewska, Katedra i Klinika
Nefrologii, Transplantologii
i Chorób Wewnętrznych,
Uniwersytet Medyczny
im. Karola Marcinkowskiego,
al. Przybyszewskiego 49, 60-355
Poznań, tel.: 61-869-17-00,
fax: 61-869-16-88, e-mail:
alicia_grzegorzewska@yahoo.com
Praca wpłynęła: 13.05.2014.
Przyjęta do druku: 14.07.2014.
Publikacja online: 21.07.2014.
Nie zgłoszono sprzeczności
interesów.
Pol Arch Med Wewn. 2014;
124 (9): 443-451
Copyright by Medycyna Praktyczna,
Kraków 2014