

Lovastatin for Reduction of Leptin in Nondialysis Patients With Type 2 Diabetic Nephropathy

Sharareh Gholamin,^{1,2} Seyed-Mostafa Razavi,^{1,2}
Seyed-Meghdad Taghavi-Garmestani,¹ Amir Ghorbanihaghjo,³
Nadereh Rashtchizadeh,³ Javid Safa,⁴ Amir Mansour Vatankhah,⁴
Tabassom Azizi,⁵ Hassan Argani^{4,5}

¹Clinical Research and Development Center, Shahid Modarres Hospital, Shahid Beheshti University of Medical Sciences, Tehran, Iran

²Institute for Stem Cell Biology and Regenerative Medicine, Department of Neurosurgery, Stanford University, Stanford, CA, USA

³Biotechnology Research Center, Tabriz University of Medical Sciences, Tabriz, Iran

⁴Drug Applied Research Center, Tabriz University of Medical Sciences, Tabriz, Iran

⁵Urology and Nephrology Research Center, Shahid Beheshti University of Medical Sciences, Tehran, Iran

Keywords. lovastatin, leptin, transferrin, albumin, diabetic nephropathy

Introduction. Diabetic Nephropathy (DN) is one of the main complications of diabetes mellitus, mostly ending to end-stage renal disease. Leptin and C-reactive protein (CRP), as inflammatory markers implicated in the progression of DN, increase in diabetes mellitus, while transferrin and albumin, as members of anti-oxidant defense mechanism, are found to decline.

Materials and Methods. In a controlled clinical trial, 65 patients with type 2 DN were assigned to receive lovastatin or placebo, for 3 months, to assess statins' impact on serum levels of leptin, CRP, transferrin, albumin, and lipid profile.

Results. Serum levels of CRP (3.52 ± 4.16 mg/dL to 2.84 ± 3.06 mg/dL, $P = .02$), leptin (10.78 ± 8.30 mg/dL to 7.80 ± 5.41 mg/dL, $P = .006$), low-density lipoprotein cholesterol (116.16 ± 46.54 mg/dL to 85.46 ± 29.22 mg/dL, $P < .001$), and total cholesterol (199.00 ± 43.33 mg/dL to 164.67 ± 35.19 mg/dL, $P < .001$) were lowered after lovastatin therapy. Mean serum level of high-density lipoprotein cholesterol increased (40.00 mg/dL to 42.80 mg/dL, $P = .005$) after the treatment. Lovastatin had no significant effect on albumin and transferrin. Placebo did not change any of the parameters after 3 months.

Conclusions. The effect of statins on the inflammatory markers involved in the development of DN is a new approach to evidence supporting the pleiotropic effect of this drug group.

IJKD 2014;8:201-6
www.ijkd.org

INTRODUCTION

Diabetic nephropathy (DN) is one of the main complications of diabetes mellitus, especially type 2. Advanced stages of the disease can be a substantial underlying factor for glomerulosclerosis and end-stage renal dysfunction.^{1,2} Leptin, derived from adipose tissue, is a known mediator in the development of DN. This 167-amino acid protein plays a striking role in body energy homeostasis, regulation of immune function, hematopoiesis, angiogenesis, and bone development.³ Leptin

level is likely increased in patients with type 2 diabetes mellitus and correlates positively with insulin resistance.^{4,5} In an etiologic survey, leptin seemed to induce overexpression of tumor growth factor- β (TGF- β). Rising of TGF- β is associated with collagen type IV augmentation and results in proteinuria, which is the first step to diabetic glomerulopathy. Endothelial cell proliferation and glomerular extracellular accumulation (mesangial expansion) are other pathologic changes found in the later stages of diabetic glomerulopathy, caused

by a high leptin level.^{6,7}

Diabetes mellitus, like many other diseases such as infectious diseases and cancer, goes with elevated inflammatory factors and decreased antioxidant defense mechanism. Inflammatory status in diabetes accompanies with the decline of anti-inflammatory biomarkers incorporated in antioxidant defense system (albumin and transferrin) and an increase in C-reactive protein (CRP) as a marker of inflammation.⁸⁻¹¹

Dyslipidemia, as a known predisposing factor in deterioration of kidney function and cardiovascular disorders in patients with type 2 diabetes mellitus, is of great concern. Statins are found to be effective contributors in improvement of lipid parameters, eg, lowering low-density lipoprotein cholesterol (LDLC) and triglyceride, and raising high-density lipoprotein cholesterol (HDLC).¹¹⁻¹⁴ Additionally, statin therapy can be effective in reducing inflammation and glomerular filtration dysfunction in patients with chronic kidney disease.¹⁵⁻¹⁸ Recent studies manifested that statins, through non-lipid-lowering property (pleiotropic effect), decreased CRP and increased transferrin and albumin as a contributor to anti-oxidant defense mechanism in patients with diabetes mellitus.¹⁹⁻²²

To our knowledge there has not yet been any study designed focusing on the impact of statins on leptin levels in patients with type 2 DN. As far as high leptin level is responsible for the degree of inflammation and glomerular dysfunction, statins, through their non-lipid-lowering property, could effectuate leptin levels in DN. Moreover, the effect of statins on transferrin and albumin levels engenders a new approach to inflammation in patients with DN. Hence, we were incited to assess the effect of lovastatin on leptin, transferrin, albumin, CRP, and lipid profile in patients with DN.

MATERIALS AND METHODS

A single-blinded, parallel group, placebo-controlled clinical trial was carried out to assess the effect of 3 months of treatment with lovastatin, 20 mg/dL, daily, on the plasma levels of leptin, CRP, transferrin, albumin, fasting blood glucose, urea, creatinine, estimated glomerular filtration rate (GFR), and lipid profile (triglyceride, total cholesterol, LDLC, and HDLC), along with 24-hour urine volume, 24-hour urine protein, and 24-hour urine creatinine in patients with DN. Sixty-five

patients with type 2 DN (GFR, 30 mL/min to 60 mL/min, calculated by the Cockcroft-Gault formula; proteinuria, > 500 mg/d) were recruited. The study population included men with DN, aged between 25 and 60 years. Patients with the following conditions were excluded: smoking habits, extreme physical activity (> 3 hours exercise per week), uncontrolled cardiovascular (diagnosed by symptoms and electrocardiogram) or liver disease (abnormal liver function tests), history of diabetic complications (such as ketoacidosis, hyperosmolar coma, and diabetic foot), poorly controlled type 2 diabetes (hemoglobin A1c > 7.5%), acute myocardial infarction, malnutrition, acute or chronic infection (diabetic foot, hepatitis, etc), hospitalization in the past 6 months, and nonadherence to regular life style (such as regular diet, drugs, and exercise). Women were not enrolled for inflammatory acute-phase markers are altered by female sexual hormones.²³ The study was carried out from February 2006 to March 2008. The ethics committee of Tabriz University of Medical Sciences approved the study protocol and all patients gave written informed consent.

Patients with hypertension were treated with 50 mg/d to 150 mg/d of captopril or 10 mg/d to 40 mg/d of enalapril during the study. Blood pressure was controlled to be lower than 130/90 mm Hg and the blood glucose was kept between 70 mg/dL and 170 mg/dL by oral hypoglycemic agents or insulin injection therapy. Thirty patients were assigned to receive 20 mg/dL/d of lovastatin (Hakim Pharma, Tehran, Iran) and 35 patients received placebo. Peripheral blood samples were drawn after an overnight fasting and stored at -70°C, before and after 3 months of treatment with lovastatin or placebo. Twenty-four-hour urine was collected and assessed for volume, protein, and creatinine at the beginning and end of the study.

Serum levels of total cholesterol, triglyceride, and HDLC were quantified with an automated chemical analyzer (Abbott analyzer, Abbott Laboratories, Abbott Park, Chicago, Illinois, USA). Serum LDLC was calculated by the Friedewald equation. An ultrasensitive solid phase enzyme-linked immunosorbent assay (DRG instruments GmbH, Marburg, Germany; Lot No, RN-28874) was used to determine serum CRP. Transferrin and leptin levels were measured by enzyme-linked immunosorbent assay (EIA-Vendor, batch

No, RD-1505).

The results were expressed as mean ± standard deviation, and statistical analysis was performed using the SPSS software (Statistical Package for the Social Sciences, version 13.0, SPSS Inc, Chicago, Ill, USA). The differences in quantitative variables were tested with paired samples *t* test and correlations with the Pearson correlation test. *P* values less than .05 were assumed significant.

RESULTS

Demographic characteristics for both groups are presented in Table 1. Meaningful reduction in serum levels of CRP (3.52 ± 4.16 mg/dL to 2.84 ± 3.06 mg/dL, *P* = .02), leptin (10.78 ± 8.30 mg/dL to 7.80 ± 5.41 mg/dL, *P* = .006), LDLC (116.16 ± 46.54 mg/dL to 85.46 ± 29.22 mg/dL, *P* < .001), and total cholesterol (199.00 ± 43.33 mg/dL to 164.67 ± 35.19 mg/dL, *P* < .001) were noticeable after 3 months of

treatment with lovastatin; nevertheless, the mean level of HDLC increased prominently (40.00 ± 5.30 mg/dL to 42.80 ± 5.15 mg/dL, *P* = .005). Plasma concentrations of transferrin, albumin, blood urea nitrogen, creatinine, and triglyceride, as well as 24-hour urine volume and 24-hour urine protein-creatinine ratio were not markedly influenced by lovastatin (Table 2). Placebo did not change any of the parameters after 3 months.

Quantitative serum level of CRP before drug administration had a significant positive correlation with leptin serum levels ($r = 0.465$, *P* = .01) and a significant negative correlation with transferrin serum levels ($r = -0.361$, *P* = .05) in the lovastatin group (Figure). The positive correlation between serum CRP and leptin levels remained significant after administration of lovastatin ($r = 0.391$, *P* = .03). The control group demonstrated a positive correlation between CRP and leptin, both

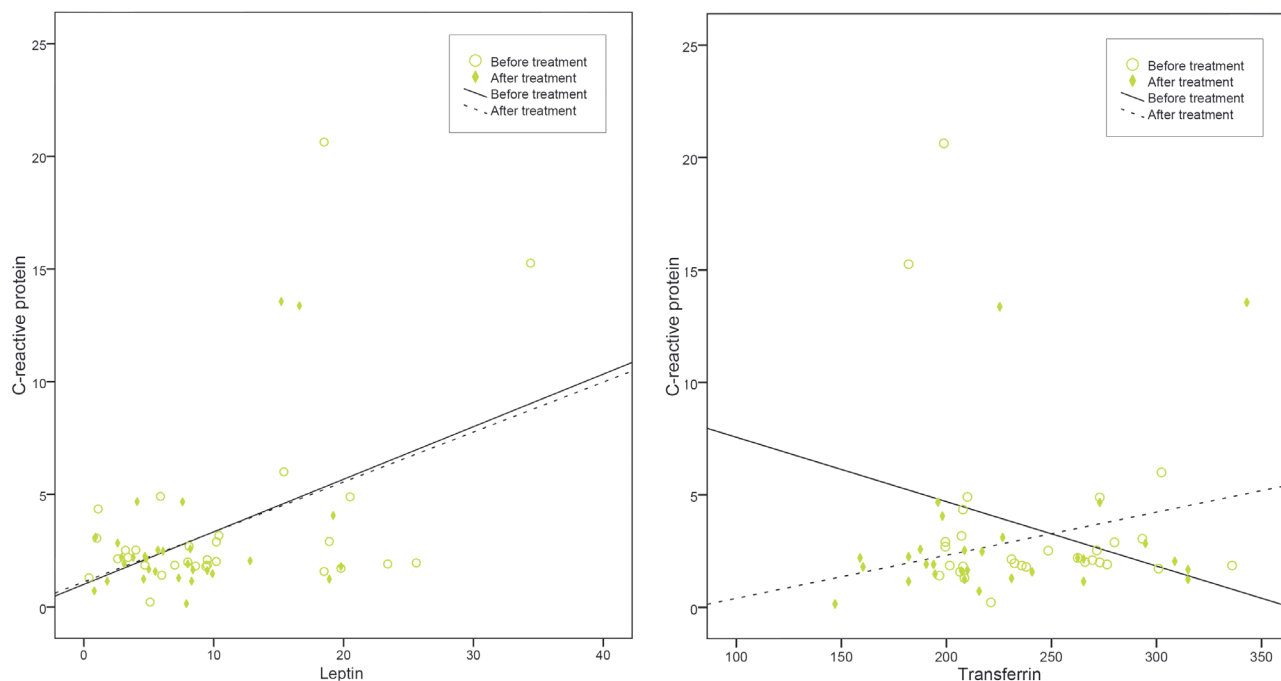
Table 1. Characteristics of Participants in Lovastatin and Placebo Groups

Characteristic	Lovastatin Group	Placebo Group
Mean age, y	54.47 ± 6.11	50.53 ± 8.25
Mean body mass index, kg/m ²	25.55	25.72
Mean systolic blood pressure, mm Hg	139.72	137.11
Mean diastolic blood pressure, mm Hg	76.00	73.46
Mean duration of diabetes, y	9.77 ± 3.11	8.77 ± 3.46
Hypertension, n (%)	13 (43.3)	12 (40.0)

Table 2. Mean Levels of Recorded Parameters in Lovastatin and Placebo Groups*

Parameter	Lovastatin Group		Placebo Group	
	Before	After	Before	After
Plasma				
Albumin, g/dL	4.41	4.47	4.69	4.64
Triglyceride, mg/dL	175.3	152.1	172.5	165.0
Total cholesterol, mg/dL	199.0†	164.7†	194.3	198.8
LDLC, mg/dL	116.2†	85.5†	124.1	121.1
HDLC, mg/dL	40.0†	42.8†	39.8	35.6
Urea, mg/dL	55.2	51.8	54.2	53.2
Creatinine, mg/dL	1.66	1.61	1.68	1.60
Uric acid, mg/dL	5.9	6.2	6.0	6.6
Fasting glucose, mg/dL	157.83	151.53	146.37	150.91
CRP, mg/dL	3.52†	2.84†	2.75	3.61
Transferrin, mg/dL	241.26	227.47	248.85	245.32
Leptin, ng/mL	10.78†	7.80†	9.95	9.42
GFR, mL/min/1.73 m ²	54.78	54.57	53.35	54.28
Urine				
24-hour volume, ml/d	2138	2229	2217	2260
24-hour protein, mg/d	681	618	675	683
24-hour creatinine, mg/d	1218	1239	1246	1297

*LDLC indicates low-density lipoprotein cholesterol; HDLC, high-density lipoprotein cholesterol; GFR, glomerular filtration rate; and CRP, C-reactive protein.
†*P* < .01



Scatter plots show correlations of leptin and transferrin with C-reactive protein before and after treatment with lovastatin.

before ($r = 0.468$, $P = .005$) and after ($r = 0.416$, $P = .01$) placebo administration and no significant correlation between CRP and transferrin either before or after taking placebo. No significant correlation was found between leptin and transferrin serum levels in either group.

DISCUSSION

Statins are the first-line lipid-lowering therapy in diabetic patients. They reduce lipid production by inhibition of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase.^{24,25} Recent studies investigated the anti-inflammatory aspect of this drug group beyond its lipid lowering property,^{19,26} though no trial has reported the effect of statins on leptin, transferrin, albumin, and CRP in patients with type 2 DN. This study showed that a 3-month administration of lovastatin significantly reduced serum levels of total cholesterol, LDLC, and leptin and raised serum levels of HDLC in patients with DN. Recent data demonstrated that statins lower total cholesterol, LDLC, and triglyceride and significantly increase HDLC levels.^{14,15,27,28} Nevertheless, in our study, no significant change in triglyceride levels was seen.

Statins can inhibit mesangial cell proliferation through influencing LDLC, oxidized LDL, high levels of glucose, and many other factors

mediating the progression of kidney disease.^{1,29} Available studies demonstrate the role of leptin in glomerulosclerosis and development of kidney damage in diabetic patients. The potential impact of leptin on renal pathophysiology reveals that high leptin level induces glomerular endothelial cell proliferation, TGF- β 1, and collagen type IV overproduction and mesangial overexpansion. These changes discriminate leptin as a causal factor for development of DN.^{5,7,30} However, leptin level rise assuages glomerular injury in at least certain types of DN.^{3,5-7,31} The effect of statins on leptin levels seems to be ambiguous and needs further research. In the study by von Eynatten and colleagues, atorvastatin, 40 mg/d, did reduce leptin levels in type 2 diabetic patients,³² while Chu and colleagues showed that atorvastatin (10 mg/d, 20 mg/d, and 40 mg/d) did not affect leptin levels in hyperlipidemic type 2 diabetes mellitus.³³ Gannagé-Yared and colleagues found that a 12-week treatment with pravastatin, 40 mg/d, does not change leptin and adiponectin levels in healthy volunteers.³⁴ The result by Gouni-Berthold and colleagues also demonstrated that simvastatin therapy, 40 mg/d, did not change leptin levels in healthy men.³⁵ Statin treatment significantly decreased plasma lipids and leptin levels in coronary heart disease patients.²⁶ In a study on

hypercholesterolemic rabbits, atorvastatin, 2.5 mg/kg/d, decreased serum leptin levels by reducing leptin mRNA expression.²⁸

Here in this study, leptin levels were significantly decreased in patients with type 2 DN. Meanwhile, all of our patients received angiotensin-converting enzyme inhibitors as an antihypertensive or antiproteinuric agent during the study. Angiotensin II, like leptin, promotes fibroses in diabetic kidneys by decreasing TGF- β .^{7,36} Treatment by angiotensin-converting enzyme inhibitors may have a similar effect as statin on reducing leptin level.

Inflammatory biomarkers are associated risk factors participating in the pathophysiology of morbidity and mortality in diabetic patients.^{8,10,37,38} Diabetes mellitus, as an inflammatory condition, is associated with decreased serum levels of negative acute-phase proteins (eg, transferrin and albumin) and increased positive acute-phase proteins (eg, CRP and leptin).^{8,9} Transferrin, as a prototype of negative acute-phase protein, was correlated negatively with CRP, as the prototype of positive acute phase protein, before lovastatin therapy. Statin therapy through its pleiotropic character can decline CRP and improve serum albumin level¹⁹⁻²²; though lovastatin made no statistically significant changes in transferrin and albumin in our study. El Haggan and colleagues found positive correlation between CRP and leptin in early phase of post kidney transplantation.³⁹ We found the same relationship between leptin and CRP pretreatment and posttreatment with lovastatin.

Limitations of our study point to the need for further research. In particular, it would be beneficial to carry out similar studies with larger patient populations, administration of higher doses of statins, longer durations of therapy, and inclusion of both men and women. Given the menstrual cycle in females causing changes in blood levels of a number of inflammatory factors including C reactive protein, in this study we only recruited the male population. Since we have the evident effect of lovastatin on leptin in the men population, one can include the women in future studies to expand the understanding of this effect.

CONCLUSIONS

The effect of lovastatin on reducing leptin as a proinflammatory factor in pathogenesis of DN favors the so called “pleiotropic effect” of statins.

Positive correlation of CRP with leptin and its negative correlation with transferrin confirm leptin as an inflammatory and transferrin as a negative inflammatory marker. The impact of statins on serum albumin level and transferrin needs further research and is still drawn in uncertainty.

CONFLICT OF INTEREST

None declared.

REFERENCES

1. Thorp ML. Diabetic nephropathy: common questions. *Am Fam Physician*. 2005;72:96-9.
2. Dronavalli S, Duka I, Bakris GL. The pathogenesis of diabetic nephropathy. *Nat Clin Pract Endocrinol Metab*. 2008;4:444-52.
3. Briley LP, Szczech LA. Leptin and renal disease. *Semin Dial*. 2006;19:54-9.
4. Gulturk S, Cetin A, Erdal S. Association of leptin with insulin resistance, body composition, and lipid parameters in postmenopausal women and men in type 2 diabetes mellitus. *Saudi Med J*. 2008;29:813-20.
5. Ballermann BJ. A role for leptin in glomerulosclerosis? *Kidney Int*. 1999;56:1154-5.
6. Javor ED, Moran SA, Young JR, et al. Proteinuric nephropathy in acquired and congenital generalized lipodystrophy: baseline characteristics and course during recombinant leptin therapy. *J Clin Endocrinol Metab*. 2004;89:3199-207.
7. Suganami T, Mukoyama M, Mori K, et al. Prevention and reversal of renal injury by leptin in a new mouse model of diabetic nephropathy. *FASEB J*. 2005;19:127-9.
8. Memişoğullari R, Bakan E. Levels of ceruloplasmin, transferrin, and lipid peroxidation in the serum of patients with Type 2 diabetes mellitus. *J Diabetes Complications*. 2004;18:193-7.
9. Memişoğullari R, Taysi S, Bakan E, et al. Antioxidant status and lipid peroxidation in type II diabetes mellitus. *Cell Biochem Funct*. 2003;21:291-6.
10. Van Campenhout A, Van Campenhout C, Lagrou AR, et al. Impact of diabetes mellitus on the relationships between iron-, inflammatory- and oxidative stress status. *Diabetes Metab Res Rev*. 2006;22:444-54.
11. Rashtchizadeh N, Argani H, Ghorbanihaghjo A, et al. C-reactive protein level following treatment and withdrawal of lovastatin in diabetic nephropathy. *Iran J Kidney Dis*. 2009;3:93-8.
12. Gentile S, Turco S, Guarino G, et al. Comparative efficacy study of atorvastatin vs simvastatin, pravastatin, lovastatin and placebo in type 2 diabetic patients with hypercholesterolaemia. *Diabetes Obes Metab*. 2000;2:355-62.
13. Motomura T, Okamoto M, Kitamura T, et al. Effects of pitavastatin on serum lipids and high sensitivity C-reactive protein in type 2 diabetic patients. *J Atheroscler Thromb*. 2009;16:546-52.
14. Strippoli GF, Navaneethan SD, Johnson DW, et al. Effects

- of statins in patients with chronic kidney disease: meta-analysis and meta-regression of randomised controlled trials. *BMJ*. 2008;336:645-51.
15. Kasiske B, Cosio FG, Beto J, et al. Clinical practice guidelines for managing dyslipidemias in kidney transplant patients: a report from the Managing Dyslipidemias in Chronic Kidney Disease Work Group of the National Kidney Foundation Kidney Disease Outcomes Quality Initiative. *Am J Transplant*. 2004;4 (Suppl 7):13-53.
 16. Shojaei MH, Djalali M, Siassi F, Khatami MR, Boroumand MA, Eshragian MR. Serum levels of lipoprotein(a) and homocysteine in patients on hemodialysis who take hydroxymethylglutaryl-CoA reductase inhibitors, vitamin B6, and folic acid. *Iran J Kidney Dis*. 2009;3:141-4.
 17. Soliemani A, Nikouejad H, Tabatabaizade M, Mianehsaz E, Tamadon M. Effect of hydroxymethylglutaryl-CoA reductase inhibitors on low-density lipoprotein cholesterol, interleukin-6, and high-sensitivity C-reactive protein in end-stage renal disease. *Iran J Kidney Dis*. 2011;5:29-33.
 18. Shojaei M, Djalali M, Khatami M, Siassi F, Eshragian M. Effects of carnitine and coenzyme Q10 on lipid profile and serum levels of lipoprotein(a) in maintenance hemodialysis patients on statin therapy. *Iran J Kidney Dis*. 2011;5:114-8.
 19. Schönbeck U, Libby P. Inflammation, immunity, and HMG-CoA reductase inhibitors: statins as antiinflammatory agents? *Circulation*. 2004;109(Suppl 1):II18-26.
 20. Sirken G, Kung SC, Raja R. Decreased erythropoietin requirements in maintenance hemodialysis patients with statin therapy. *ASAIO J*. 2003;49:422-5.
 21. Afzali B, Haydar AA, Vinen K, et al. From Finland to fatland: beneficial effects of statins for patients with chronic kidney disease. *J Am Soc Nephrol*. 2004;15:2161-8.
 22. Rashtchizadeh N, Argani H, Ghorbanihaghjo A, Nezami N, Safa J, Montazer-Saheb S. C-reactive protein level following treatment and withdrawal of lovastatin in diabetic nephropathy. *Iran J Kidney Dis*. 2009;3:93-8.
 23. Jilma B, Dirnberger E, Loscher I, et al. Menstrual cycle associated changes in blood levels of interleukin-6, alpha1 acid glycoprotein, and C-reactive protein. *J Lab Clin Med*. 1997;130:69-75.
 24. Haffner SM, Lehto S, Rönnemaa T, et al. Mortality from coronary heart disease in subjects with type 2 diabetes and in nondiabetic subjects with and without prior myocardial infarction. *N Engl J Med*. 1998;339:229-34.
 25. Chaiyakunapruk N, Boudreau D, Ramsey SD. Pharmacoeconomic impact of HMG-CoA reductase inhibitors in type 2 diabetes. *J Cardiovasc Risk*. 2001;8:127-32.
 26. Sun YM, Li J, Luan Y, et al. Effect of statin therapy on leptin levels in patients with coronary heart disease. *Peptides*. 2010;31:1205-7.
 27. Law MR, Wald NJ, Rudnicka AR. Quantifying effect of statins on low density lipoprotein cholesterol, ischaemic heart disease, and stroke: systematic review and meta-analysis. *BMJ*. 2003;326:1423.
 28. Zhao SP, Wu ZH. Atorvastatin reduces serum leptin concentration in hypercholesterolemic rabbits. *Clin Chim Acta*. 2005;360:133-140.
 29. Campese VM, Nadim MK, Epstein M. Are 3-hydroxy-3-methylglutaryl-CoA reductase inhibitors renoprotective? *J Am Soc Nephrol*. 2005;16(Suppl 1):11-7.
 30. Wolf G, Chen S, Han DC, et al. Leptin and renal disease. *Am J Kidney Dis*. 2002;39:1-11.
 31. De Block CE, De Leeuw IH, Van Gaal LF. Impact of overweight on chronic microvascular complications in type 1 diabetic patients. *Diabetes Care*. 2005;28:1649-55.
 32. von Eynatten M, Schneider JG, Hadziselimovic S, et al. Adipocytokines as a novel target for the anti-inflammatory effect of atorvastatin in patients with type 2 diabetes. *Diabetes Care*. 2005;28:754-5.
 33. Chu CH, Lee JK, Lam HC, et al. Atorvastatin does not affect insulin sensitivity and the adiponectin or leptin levels in hyperlipidemic Type 2 diabetes. *J Endocrinol Invest*. 2008;31:42-7.
 34. Gannagé-Yared MH, Azar RR, Amm-Azar M, et al. Pravastatin does not affect insulin sensitivity and adipocytokines levels in healthy nondiabetic patients. *Metabolism*. 2005;54:947-51.
 35. Gouni-Berthold I, Berthold HK, Chamberland JP, et al. Short-term treatment with ezetimibe, simvastatin or their combination does not alter circulating adiponectin, resistin or leptin levels in healthy men. *Clin Endocrinol (Oxf)*. 2008;68:536-41.
 36. Michli E, Gulmi FA, Chou SY, et al. Atorvastatin preserves renal function in chronic complete unilateral ureteral obstruction. *J Urol*. 2007;177:781-5.
 37. Ritchie RF, Palomaki GE, Neveux LM, et al. Reference distributions for the negative acute-phase serum proteins, albumin, transferrin and transthyretin: a practical, simple and clinically relevant approach in a large cohort. *J Clin Lab Anal*. 1999;13:273-9.
 38. Riaz S, Alam SS, Akhtar MW. Proteomic identification of human serum biomarkers in diabetes mellitus type 2. *J Pharm Biomed Anal*. 2010;51:1103-1107.
 39. El Haggan W, Chauveau P, Barthe N, et al. Serum leptin, body fat, and nutritional markers during the six months post-kidney transplantation. *Metabolism*. 2004;53:614-9.

Correspondence to:
 Sharareh Gholamin, MD
 Institute for Stem Cell Biology and Regenerative Medicine, Lorry I. Lokey Stem Cell Research Building, Stanford, CA 94305, USA
 Tel: +1 650 417 8615
 E-mail: sharar@stanford.edu

Received April 2013
 Revised October 2013
 Accepted October 2013