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Heparin-mediated extracorporeal LDL precipitation treating a peripheral arterial disease patient suffering from repeated postoperative bypass occlusion

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Abstract Acute occlusion of a peripheral artery is a serious complication in peripheral arterial disease (PAD). Traditionally open surgical intervention in combination with antithrombotic therapy is the choice for treatment but the beneficial effects of both strategies are limited often by the patient's situation and therapeutic side effects. Heparin-mediated extracorporeal low-density lipoprotein precipitation (H.E.L.P.) apheresis efficiently removes circulating atherogenic lipoproteins, fibrinogen and C-reactive proteins as well as various proinflammatory and procoagulatory factors. We first report H.E.L.P. apheresis treating a PAD patient suffering from repeated postoperative femoropopliteal bypass graft occlusion, first, intensively, followed by weekly intervals. Limb amputation was avoided and the patient is doing well now. Angiography revealed bypass graft remained patent half a year after operation. This case report might help to design the regime for preventing postoperative bypass occlusion in patients with hyperlipidemia or hyperfibrinogenemia.

Introduction

Peripheral arterial disease (PAD) is defined as the tip of the atherosclerotic "iceberg" [1]. Acute peripheral arterial occlusion is a serious compli-

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cation. Traditionally, open surgical intervention remains "gold standard" for treatment despite high rate of perioperative morbidity and mortality [2]. Minimally invasive alternatives to open surgery are thus desirable provided it possesses the potential of efficiently restore arterial perfusion and thrombolysis.

Heparin-mediated extracorporeal low-density lipoprotein (LDL) precipitation (H.E.L.P.) apheresis was first introduced in 1984 as an extracorporeal procedure for selective removal of LDL. As polyanions, heparin molecules are able to form with plasma LDL, VLDL, fibrinogen and lipoprotein (a) [Lp(a)] nonsoluble precipitates at a low pH of 5.12 in the presence of bivalent cations such as Ca^{2+}, etc. A single 2-h H.E.L.P. apheresis (3 to 3.5 l of plasma is treated) reduces plasma LDL-cholesterol, fibrinogen, Lp(a) and high sensitive C-reactive protein (hs-CRP) by 60—70%. The main indications of the apheresis include familial hypercholesterolemia, coronary heart disease (CHD) and after heart transplantation. LDL-apheresis (dextran sulfate/cellulose absorbent column, Kaneka, Japan) has been reported in treating occlusive disorders in PAD patients with or without hypercholesterolemia [4—6]. But using H.E.L.P. apheresis, which simultaneously removes LDL, Lp(a), fibrinogen and CRP, as a main adjuvant therapy in treating acute postoperative peripheral bypass occlusion was not reported yet. Here, we present a case report of treating one PAD patient suffering from repeated femoropopliteal bypass graft occlusion with H.E.L.P. and limb amputation was avoided.

Case report

A 68-year-old woman with clinically established PAD since 1998 underwent percutaneous transluminal angioplasty and subsequently femoropopliteal bypass surgery in 2000. She was first referred to us in January 2001 with symptoms of critical limb ischemia such as progressive pain at rest and strict limitation to walk (<200 m). Clinical examination at that time revealed ankle brachial index (ABI)<0.4, PVD stage III according to Fontaine Classification [7]. Her LDL-cholesterol, HDL-cholesterol, Lp(a) and fibrinogen values were 3.9 mmol/l, 1.2 mmol/l, 130 mg/dl, and 2.6 g/l, respectively. D-dimer, prothrombin time (PT) and activated partial thromboplastin time (aPTT) were within normal range. She was normotensive, nonsmoker and has no history of diabetes or CHD. She was statin- and fibrate-intolerant. Family history was not remarkable. Due to her high Lp(a) levels, we treated the patient with H.E.L.P. apheresis by weekly intervals from January to August 2001. Platelet aggregation inhibitors (aspirin 100 mg/day) was continued. During this period, her symptoms were greatly relieved with ABI 0.9 and PVD stage IIa. Plasma levels of Lp(a) and fibrinogen were maintained at 40 mg/dl and 1 g/l measured at treatment intervals. For some reasons, H.E.L.P. therapy was stopped for 3 months and her symptoms of critical limb ischemia reoccurred.

In December 2001, she was admitted at our surgical department. Physical examination revealed 58 kg, body mass index=21.3, no palpable pulses from right femoral and pedal arteries, ABI<0.4 and PVD stage III. Needle angiography of right leg demonstrated distal occlusion of femoropopliteal bypass. CRP value was 78 mg/l. Coagulatory studies revealed D-dimer 1.3 mg/l, antithrombin 82% (ref. 58—113%), protein C 152% (ref. 70—140%), lupus anticoagulant activity not detectable, cardiolipin antibodies IgG<4.0 GPL-U/ml (ref.<10), IgM<4.0 GPL-U/ml (ref.<10), heterozygous for factor V Leiden R5060 and prothrombin G20210A mutations. A new venous femoropopliteal bypass graft operation was then performed. Intensive antithrombotic therapy started immediately after operation by using high dose of heparin (50000 U/day), cumarine, aspirin (100 mg/day) and clopidogrel (75 mg/day). However, the new graft occluded three times within 1 week requiring three thrombectomies. Reocclusion of graft would have left no choice but amputation.

We then treated the patient with H.E.L.P. apheresis immediately after the last thrombectomy: first, one therapy in every 2 days (five therapies in 9 days), followed by weekly intervals, thereafter by biweekly intervals till now. The procedure of apheresis was described previously [3]. In brief, venous blood was drawn from cubital vein catheter, and blood cells were separated form plasma which was mixed with 1:1 ratio acetate—heparin buffer (pH 4.85) to precipitate fibrinogen, LDL and Lp(a). This suspension was continuously recirculated to remove precipitated fibrinogen, LDL and Lp(a). Thereafter excess of heparin was removed by anion-exchange filter. Finally, physiological pH was restored by bicarbonate dialysis and extra fluid was removed by ultrafiltration. Plasma free from LDL, fibrinogen and Lp(a) was mixed with cell-rich blood fraction and returned back to the patient. Approximately 3.3 l of plasma was treated per session. Modulation of circulating parameters by apheresis is shown in the Table 1. The patient's condition improved quickly, and she was dis-

Table 1 Modulation of circulating levels of lipids, proinflammatory and coagulatory parameters by H.E.L.P. apheresis

Parameters	Pre-bypass operation	Intensive H.E.L.P. apheresis after last thrombectomy											Weekly apheresis[b]	
		1st		2nd		3rd		4th		5th		Reduction (%)[a]	pre	post
		Pre[c]	post	pre	post	pre	post	pre	post	pre	post			
Lipoproteins (mmol/l)														
LDL-C	3.3	1.1	0.4	—	—	1.6	0.7	—	—	1.8	0.9	−57	2.8	1.0
HDL-C	1.3	0.6	0.4	—	—	0.7	0.6	—	—	1.2	0.8	−27	1.3	0.9
Lp(a) (mg/dl)	96	40	11	—	—	51	22	—	—	68	33	−60	85	29
Proinflammatory and coagulatory parameters														
Fibrinogen (g/l)	3.0	4.2	1.4	3.0	1.6	3.2	1.3	2.0	1.1	2.6	1.0	−55	1.8	0.8
CRP (mg/l)	<5	78	24	13	8	16	7	—	—	6	2	−58	<5	<5
INR	1.0	1.2	2.5	1.7	2.5	1.8	3.8	1.3	1.7	1.1	1.9	+74	1.0	1.8
PT	0.95	0.75	0.34	0.52	0.34	0.5	0.23	0.7	0.52	0.85	0.46	−43	0.95	0.5

LDL-C: low-density lipoprotein cholesterol; Lp(a): lipoprotein (a); CRP: C-reactive protein; INR: international normalized ratio; PT: prothrombin time.

[a] Denotes the average reduction of the five therapies (− denotes decrease, + denotes increase).

[b] Representative values from one single apheresis 6 months after bypass operation.

[c] Denotes the values just before (pre) and immediately after (post) each apheresis.

charged 3 weeks after bypass operation. Weekly apheresis maintained her Lp(a) and fibrinogen levels on average at 40 mg/dl and 1 g/l (measured at treatment intervals). Aspirin (100 mg/day) and clopidogrel (75 mg/day) are continued till now. Pulse Doppler and ultrasonography was performed half a year later, revealing ABI>1.0 and no relevant stenosis of the bypass. Needle angiography showed that the graft remained patent. The patient is now clinically stable, no symptoms of limb ischemia.

Discussion

The basic pathophysiological processes underlying major complications of PAD are atherogenesis and thrombogenesis. Endothelial dysfunction is directly related to the progression and clinical complications of PAD. It is reported that CRP, fibrinogen and ABI are strongly associated with flow-mediated dilation in brachial artery [8]. CRP stimulates the atherosclerotic vessel by upregulating the complement system and induces expression of adhesion molecules (AMs) and chemokines in endothelial cells [9]. Fibrinogen alters blood viscosity, platelet aggregation and thus hemorheology [10]. It also contributes to vascular dysfunction through its proteolytic degradation products [11].

Therapeutic measures, including antithrombotic drugs, cholesterol-lowering drugs such as statins, and vasodilatory agents, have not proved satisfactory in the management of PAD patients. Surgical intervention, such as amputation recommended, as last approach is associated with morbidity, functional impairement and mortality. H.E.L.P. apheresis, which is unique to other LDL-apheresis systems, removes simultaneously circulating LDL, fibrinogen, Lp(a) and CRP with high efficiency. It removes clotting factors, such as factor V, VII, VIII and IX, by 45—60% and antithrombin by 25% [12]. We found recently that this apheresis also reduces circulating levels of AMs, chemokines, endothelin-1, lipopolysaccharide-binding protein, homocysteine, tissue factor and CD40 ligand [13]. All these factors play an important role in atherosclerosis.

Our patient came with moderate levels of LDL and fibrinogen but high levels of Lp(a) which deserves attention. Lp(a), a homologue to plasminogen, competes with its binding site on fibrin clots resulting in disturbance of fibrinolysis. Lp(a) levels are genetically determined and cannot be remarkably reduced either by diet or statins [14]. Apheresis proved its beneficial effects by keeping her LDL, Lp(a) and fibrinogen levels low during January to August in 2001. Cessation of the apheresis caused recurrence of her symptoms.

The new femoropopliteal bypass graft occluded repeatedly after operation. Combination of intensive surgical intervention and antithrombotic therapy could not benefit her furthermore. H.E.L.P. apheresis was then applied adjuvant to antithrombotic therapy. The rationale of first intensive apheresis treatment (once in every 2 days) was based on several observations during our H.E.L.P. practice: a significant increase of fibrinogen levels was seen in CHD patients immediately after coronary bypass operation [15]; hyperlipidemic patients had a quicker recovery of fibrinogen and Lp(a) levels compared to healthy individuals after single H.E.L.P. apheresis [16]; reduction of plasma viscosity immediately after one apheresis is 20% while only 10% after 24 h [17]. In this patient, aggressive and repeated removal of fibrinogen, clotting factors, atherogenic Lp(a) and various proinflammatory and procoagulatory factors might attenuate the acute thrombogenesis, reduce plasma viscosity and platelet aggregation, reestablish swiftly the perfusion of occluded artery and restore the oxygen and nutrient supply to the critically ischaemic leg. The continuous regular apheresis kept above parameters in low levels, which may stabilize and even gradually diminish the existing atherosclerotic plaques and thrombus, maintain bypass graft patent.

Our patient was also heterozygous for factor V Leiden R506Q and prothrombin G20210A mutations, which are most common inherited risk factors for venous thromboembolism. However, it is still controversial whether these mutations are associated with postoperative graft occlusion [18]. Major side effects about H.E.L.P. apheresis include hypotension, vasovagal reaction and gastrointestinal pains. Incidences vary from 0.3% to 2.5%, respectively [3]. But none of these side effects happened during the treatment of this patient. This case report may help to design the regime for preventing postoperative bypass occlusion in patients with hyperlipidemia or hyperfibrinogenemia.

References

[1] Futterman LG, Lemberg L. Peripheral arterial disease is only the tip of the atherosclerotic "iceberg". *Am J Crit Care* 2002;**11**:390—4.

[2] Kenneth O. Peripheral arterial disease. *Lancet* 2001;**358**: 1257—64.

[3] Blessing F, Wang Y, Nagel D, Seidel D. The efficacy and safety of the new heparin-induced extracorporeal low-density lipoprotein precipitation system (Plasmat Futura) in

comparison with the currently used system (Plasmat Secura). *Therap Apher Dial* 2004;**8**:31—6.
[4] Agishi T, Naganuma S, Nakasato S, Kitajima K, Ota K, Ban K, et al. Treatment of arteriosclerotic obstruction by LDL adsorption. *Angiology* 1993;**44**:222—7.
[5] Fuda Y. Efficacy of LDL apheresis on the treatment of patients with arteriosclerotic obliterans without LDL hypercholesterolemia. *Jpn J Artif Organs* 1994;**23**(suppl):S118.
[6] Onoyama O. Case report on LDL apheresis treatment for a patient with peripheral arterial occlusive disease without hypercholesterolemia. *J Jpn Soc Dial Ther* 1996;**29**:850 (suppl.).
[7] Fontaine R, Kim M, Kieny R. Surgical treatment of peripheral circulation disorders. *Helv Chir Acta* 1954;**21**: 499—533.
[8] Brevetti G, Silvestro A, Di Giacomo S, Roxana B, Di Donato A, Schiano V, et al. Endothelial dysfunction in peripheral arterial disease is related to increase in plasma markers of inflammation and severity of peripheral circulatory impairment but not to classic risk factors and atherosclerotic burden. *J Vasc Surg* 2003;**38**:374—9.
[9] Moriarty PM, Gibson CA, Shih J, Matias MS. C-reactive protein and other markers of inflammation among patients undergoing HELP LDL apheresis. *Atherosclerosis* 2001;**158**: 495—498.
[10] Schuff-Werner P, Schutz E, Seyde WC, Eisenhauer T, Janning G, Armstrong VW, Seidel D. Improved haemorheology associated with a reduction in plasma fibrinogen and LDL in patients being treated by heparin-induced extracorporeal LDL precipitation (HELP). *Eur J Clin Invest* 1989;**19**:30—7.
[11] Freedman JE, Fabian A, Loscalzo J. Impaired EDRF production by endothelial cells exposed to fibrin monomer and FDP. *Am J Physiol* 1995;**268**:C520—6.
[12] Jaeger BR, Goehring P, Schirmer J, Uhrig S, Lohse P, Kreuzer E, et al. Consistent lowering of clotting factors for the treatment of acute cardiovascular syndromes and hypercoagulability: a different pathophysiological approach. *Therap Apher* 2001;**5**:252—9.
[13] Wang Y, Blessing F, Walli AK, Überfuhr P, Fraunberger P, Seidel D. Effects of heparin-mediated extracorporeal low-density lipoprotein precipitation beyond lowering proatherogenic lipoproteins—reduction of circulating proinflammatory and procoagulatory markers. *Atherosclerosis* 2004; **175**:145—50.
[14] Armstrong VW, Harrach B, Robenek H, Helmhold M, Walli D, Seidel D. Heterogeneity of human lipoprotein Lp[a]: cytochemical and biochemical studies on the interaction of two Lp[a] species with the LDL receptor. *J Lipid Res* 1990; **31**:429—41.
[15] Blessing F, Jaeger BR, Oberhoffer M, Reichart B, Seidel D. Prevention of early graft occlusion after coronary bypass grafting by post-operative reduction of plasma fibrinogen by H.E.L.P. apheresis. First evaluation of patients treated during our study (44 bypasses). *Z Kardiol* 2003;**92**(Suppl. 3):III42—7.
[16] Thiery J, Ivandic B, Bahlmann G, Walli AK, Seidel D. Hyperlipoprotein(a) anemia in nephrotic syndrome. *Eur J Clin Invest* 1996;**26**:316—21.
[17] Mellwig KP, Baller D, Gleichmann U, Moll D, Betker S, Weise R, et al. Improvement of coronary vasodilation capacity through single LDL apheresis. *Atherosclerosis* 1998;**139**: 173—8.
[18] Sampram ES, Lindblad B. The impact of factor V mutation on the risk for occlusion in patients undergoing peripheral vascular reconstructions. *Eur J Vasc Endovasc Surg* 2001; **22**:134—8.