Enoxaparin decreases serum MCP-1 concentration during haemodialysis—preliminary report

Sir,

Monocyte chemoattractant protein (MCP-1) belongs to the large family of chemokines. It is a CC chemokine; therefore, it attracts mononuclear cells and can be found at sites of chronic inflammation. Evidence points to its major role in the early development of an atherosclerotic lesion, vasculogenesis and thrombosis. It plays a leading role in atherosclerosis, causing plaque instability through accumulation of monocytes/macrophages, chronic inflammation and smooth muscle cell proliferation. Furthermore, it induces the expression of other chemokines, tissue factor, matrix metalloproteinases, cell adhesion molecules and proinflammatory cytokines [1]. All in all, it might be a novel therapeutic target in myocardial infarction, ischaemic cardiomyopathy [2] or progressive renal injury in diabetic nephropathy [3].

Enoxaparin is one of the low-molecular-weight heparins (LMWHs). It differs from unfractionated heparin mostly due to its reduced tendency to non-specific binding to plasma proteins, platelets, endothelial and blood cells. It has pleiotropic effects and (as all other LMWHs) is believed to be a wonder drug [4].

We aimed to determine the effect of enoxaparin on MCP-1 levels in a specific population with highly accelerated atherosclerosis such as maintenance haemodialysis (HD) patients. We speculated that heparin probably releases MCP-1, as do many other cytokines and growth factors (our group proved before that it releases and modifies hepatocyte growth factor/activin A/follistatin system in haemodialyzed patients [5]).

Seventeen patients (7 men; mean age 71 years) who had been undergoing maintenance HD for a mean period of 62 months (range 15.5–177 months) were enrolled into this study. Exclusion criteria were malignancy, severe liver disease (alanine aminotransferase > 50 U/l), recent acute inflammatory or infectious diseases (C-reactive protein > 10 mg/l), recent surgery, immunosuppressive therapy, insulin-dependent diabetes mellitus, treatment with vitamin K antagonists, heparin (except for HD) or regularly with non-steroidal anti-inflammatory drugs, HD vintage < 3 months prior to the study, other HD access than native arteriovenous fistula and Kt/V < 1.2. The local ethics committee approved the study, and both oral and written information was given before signed consent was obtained from all patients prior to participation. The protocol also abided by the tenets of the Helsinki protocol.

The patients were treated with bicarbonate HD three times weekly, they were dialyzed for 4–5 h using the double-needle technique, native arteriovenous fistulas and low-flux dialysers, and 16 patients received erythropoietin. The dialysers were primed with saline, and then LMWH was administered as a single bolus via the first access needle. The effective dose of enoxaparin was 0.75 ± 0.35 mg/kg.

Five millilitres of fasting blood was drawn into ethylenediaminetetraacetic acid (EDTA)-coated vacutainers during a midweek morning HD. At T0 it was drawn from the access (before heparinization) and at T10 and T180 from the pre-dialyzer port after slowing the blood flow to 100 ml/min for 1 min. They were chilled in ice water and plasma was obtained by centrifugation at 3800 g for 10 min within 30 min of collection. Afterwards the samples were aliquoted and stored at −70 °C until further needed.

Plasma MCP-1 levels were determined by enzyme-linked immunosorbent assay (ELISA) kits purchased from R&D Systems Inc. (Minneapolis, MN, USA) (catalogue number DCP00) according to the manufacturer’s instructions. All samples were measured in duplicate. The within- and between-assay coefficients were < 8%. All data were normally distributed as provided by the Shapiro–Wilk W test, and expressed as means ± 1 SD. For statistical analysis, ANOVA for repeated measures with the post hoc Newman–Keuls test were used. Two-sided P-values < 0.05 were considered significant.

Pre-dialysis plasma MCP-1 levels in HD patients were 523.4 ± 229.4 pg/ml and comparable to values found in the literature [6–9]. After 10 min of HD, there was a significant decrease in MCP-1 concentration (P < 0.05) to 329.9 ± 158.1 pg/ml, which returned to baseline levels after 180 min of procedure (606.2 ± 318.2 pg/ml, Figure 1). There was no correlation between heparin dose and the percentage decrease in plasma MCP-1 T0 versus T10 (r = 0.1178, P = 0.6525).

This is, to our knowledge, the first report showing a decrease in MCP-1 concentration probably caused by enoxaparin and it needs further confirmation. It has been shown before that heparin inhibits INF-γ-dependent up-regulation of MCP-1 production and can also antagonize the tyrosine phosphorylation of phosphatidylinositol 3-kinase produced by MCP-1 [10]. This may partly explain the decreased concentration of MCP-1 immediately after enoxaparin administration. There are of course many limitations to our study: the lack of a healthy volunteer group receiving enoxaparin (which would be a completely different population) and no dialyzed control group (partly because of uncertain safety and operative efficacy of haemodialysis without anticoagulation), that is why it is hard to prove whether in fact enoxaparin and not haemodialysis per se reduced the circulating MCP-1 levels, although it is thought provoking. The few studies examining the effect of haemodialysis on MCP-1 are conflicting: one shows an increased MCP-1 level after haemodialysis [6] and the other decreased

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concentration 8 h later (in that study heparin was used although the authors did not elaborate on that fact) [7]. The phenomenon that enoxaparin might reduce the circulating levels of MCP-1 may be very interesting and potentially useful. It suggests an anti-atherosclerotic effect of this LMWH and yet another use of it.

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**Brain renin–angiotensin system: Is it important in dialysis patients?**

Sir,

Excessive inter dialytic weight gain (IDWG) is an important clinical problem and portends a poor outcome in haemodialysis patients [1]. Treatment is based on the restriction of salt and water intake. However, the compliance is usually poor and one-third of patients treated with maintenance haemodialysis are chronically overhydrated.

The cardiovascular and haemodynamic effects of the peripheral renin–angiotensin system (RAS) are well documented. Conversely, the current knowledge regarding the effects of the brain RAS on blood pressure and thirst is mainly based on experimental studies. It was shown in experimental models that the stimulation of centrally located angiotensin 1 receptors (AT1-R) causes a rise in blood pressure, a release of vasopressin [2], thirst and thereby an increase in salt [3] and water [4] intake. In this respect, the effects of the RAS system on the brain could be important in haemodialysed patients. The brain RAS system upregulation is likely to increase the patients’ drinking behaviour, along with an increase in salt and water intake causing excessive IDWG. Therefore, the brain RAS represents a potential target for RAS inhibitors to counteract these centrally related effects.

Dipsogenic activity is mediated only by AT-1 receptors. Brain tissue, with the exception of the circumventricular organs, is separated from the circulation by the blood–brain barrier. AT-1 receptors are widespread in the brain, and their stimulation in circumventricular structures causes drinking and pressor responses. In addition to circumventricular organs, AT1 receptors are also located in the inside compartment of the blood–brain barrier, and the activation of these AT1 receptors also causes drinking and pressor responses [5].

One is allowed to speculate that inhibiting centrally related effects of brain RAS with lipophilic angiotensin-converting enzyme and/or AT1-R blockers exerts a favourable effect on the IDWG. In a prospective, self-controlled and interventional study that included 30 anuric haemodialysed patients, we compared the IDWG in a period of 3 consecutive months without and with administration of telmisartan. We showed that the IDWG significantly decreased following telmisartan at a dosage of 40 mg/day [6]. These results suggest that the treatment of hypertension in haemodialysed patients with AT1-R inhibitors does not only aim at reducing the blood pressure—a short-term effect. In the long run, it also diminishes thirst, salt and...