Letter to the Editor

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Impact of stopping vitamin K antagonist therapy on concentrations of dephospho-uncarboxylated Matrix Gla protein

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To the Editor,

Several experimental and clinical studies suggest that vitamin K antagonist (VKA) therapy is a risk factor for the development of vascular calcifications and calciphylaxis in hemodialysis (HD) patients [1]. One major pathophysiological mechanism that could explain these observations involves both vitamin K and matrix Gla protein (MGP). Briefly, MGP is an 11 kDa protein secreted by vascular smooth muscle cells, acting as a potent local inhibitor of vascular calcification. In order to be fully active, MGP must be phosphorylated and carboxylated [2]. This carboxylation is highly dependent on availability of vitamin K. HD patients are per se prone to vitamin K deficiency which can be potentiated by VKA therapy. In this situation of vitamin K depletion, the MGP calcification inhibitor activity is decreased leading to vascular calcification [3]. The inactive form of MGP, namely the dephospho-uncarboxylated MGP (dp-ucMGP), is presented as a good biomarker of vitamin K status and vascular calcification. Indeed, higher dp-ucMGP levels have been associated with higher level of vascular calcifications in CKD and HD patients [4]. To sustain the hypothesis of dp-ucMGP as a marker of vitamin K status, several authors have shown in the general population, in CKD and in HD patients that VKA therapy was associated with higher dp-ucMGP levels [4–6]. Conversely, recent data in HD patients have shown that dp-ucMGP concentrations were decreasing after vitamin K supplementation [7–9]. Whether vitamin K therapy could be of interest in the prevention of vascular calcification in HD patients is currently under investigation [10]. Until recently, few alternative strategies were available to VKA in HD with atrial fibrillation or valve replacement. Recent data in HD suggested that fondaparinux, an indirect factor Xa inhibitor, could be safely used in these patients [11]. In the present study, we have measured dp-ucMGP in patients directly after switching from VKA to fondaparinux. Our goal was to confirm the influence of VKA on dp-ucMGP levels and study the kinetic of these potential changes in MGP concentrations.

We studied HD patients treated by VKA in our university center. These patients were all treated by acenocoumarol. Switching from VKA to fondaparinux was considered only in patients anticoagulated for atrial fibrillation. Seven patients, dialyzed three times a week were considered. Two measurements (T1 and T2) were obtained at the beginning of the two dialysis sessions before stopping VKA. The patients stopped VKA therapy the day before the first dialysis session of the next week (on Sunday or Monday). Five measurements were then obtained at the beginning of each of the next five dialysis sessions (T3–T7). dp-ucMGP was quantified with an automated assay (Ina*K*tif MGP iSYS kit, IDS, Boldon, UK). dp-ucMGP concentrations were compared using one-way repeated measures analysis of variance by ranks (Friedman test), followed by Dunn's test correcting the α-level for pairwise comparison between time-points. Mann-Whitney test was used for comparison between cohorts.

Main clinical and biological characteristics of the seven patients are described in Table 1. Before switching from VKA to fondaparinux, median concentrations of dp-ucMGP

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Table 1: Main clinical characteristics and biological data of the total population of the study and according to antivitamin K (VKA) therapy status.

n	7
Age, years	75 (68; 84)
Male gender	5
Body mass index, kg/m^2	23.7 (22.8; 25.8)
Dialysis vintage, months	59 (45; 80)
Hypertension, %	100
Diabetes, %	29
Smoking habit, %	29
Calcium, mmol/L	2.07 (2.00; 2.22)
Phosphate, mmol/L	1.46 (1.13; 1.77)
Albumin, g/L	41 (40; 42)
PTH, pg/mL	206 (158; 357)
dp-ucMGP, pmol/L	6312 (5485; 8693)

Data are expressed as median and interquartile range (percentile 25; percentile 75). dp-ucMGP, dephosphorylated and uncarboxylated; PTH, parathormone.

obtained at two different times (T1 and T2) were very high and not different: T1 6316 (5485; 8693) and T2 6150 (4911; 7325) pmol/L. We have indeed previously shown a median concentration of 1939 (1419; 2841) pmol/L in 137 HD patients not treated by VKA [4]. In the two first dialysis sessions following the switch (T3 and T4), the median concentration did still not change significantly: 5902 (4842; 9165) and 4505 (3295; 6791) pmol/L for T3 and T4, respectively. However, all measurements obtained after T4 significantly ($p<0.05$) decreased in comparison to T1: T5 3810 (2331; 4979), T6 3850 (2159; 4586), and T7 2948 (1644; 3721) pmol/L. After this initial decrease, a plateau was reached and concentrations at T4, T5, T6 and T7 were similar (Figure 1). Our sample is limited (n=7) but the decrease in dp-ucMGP is constant in every single patient and clinically relevant as values at T7 have decreased by at least 40% compared to values at T1.

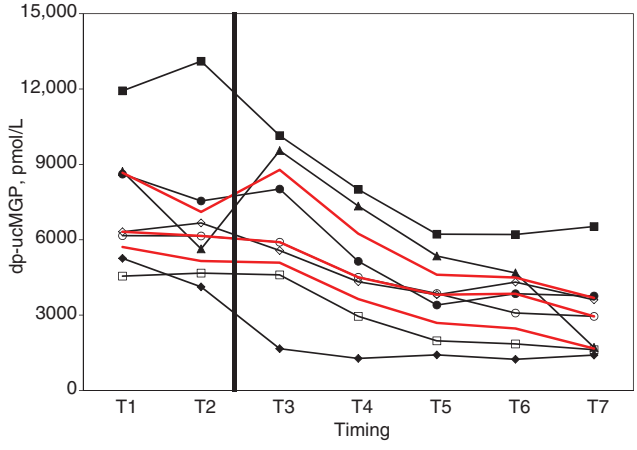

Figure 1: Evolution of dp-ucMGP concentrations after stopping VKA in 7 HD patients.
In red, median and P25-P75 values. The thick dark line corresponds to the time when AVK therapy is stopped.

Also, the median concentration of dp-ucMGP in the seven patients with VKA at T1 was significantly higher than the concentration of the 137 HD patients not treated by VKA ($p<0.0001$). However, the median concentrations between these 137 patients and the seven patients at T7 were not significantly different anymore [4].

Stopping VKA in HD patients is associated with a rapid reduction of dp-ucMGP concentrations. Within 5 days after stopping the VKA, the median concentrations of dp-ucMGP decreased to usual levels observed in HD patients not treated by VKA. This time-effect could however be different with other types of VKA than acenocoumarol. It remains to be proven if this reduction of dp-ucMGP is associated with a better evolution of vascular calcifications.

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References

1. Brandenburg VM, Kramann R, Specht P, Ketteler M. Calciphylaxis in CKD and beyond. Nephrol Dial Transplant 2012;27:1314–8.
2. Schurgers LJ, Spronk HM, Skepper JN, Hackeng TM, Shanahan CM, Vermeer C, et al. Post-translational modifications regulate matrix Gla protein function: importance for inhibition of vascular smooth muscle cell calcification. J Thromb Haemost 2007;5:2503–11.
3. Tantisattamo E, Han KH, O'Neill WC. Increased vascular calcification in patients receiving warfarin. Arterioscler Thromb Vasc Biol 2015;35:237–42.

4. Delanaye P, Krzesinski JM, Warling X, Moonen M, Smelten N, Medart L, et al. Dephosphorylated-uncarboxylated Matrix Gla protein concentration is predictive of vitamin K status and is correlated with vascular calcification in a cohort of hemodialysis patients. BMC Nephrol 2014;15:145.
5. Cranenburg EC, Koos R, Schurgers LJ, Magdeleyns EJ, Schoonbrood TH, Landewe RB, et al. Characterisation and potential diagnostic value of circulating matrix Gla protein (MGP) species. Thromb Haemost 2010;104:811–22.
6. Boxma PY, van den Berg E, Geleijnse JM, Laverman GD, Schurgers LJ, Vermeer C, et al. Vitamin K intake and plasma desphospho-uncarboxylated matrix Gla-protein levels in kidney transplant recipients. PLoS One 2012;7:e47991.
7. Schlieper G, Westenfeld R, Kruger T, Cranenburg EC, Magdeleyns EJ, Brandenburg VM, et al. Circulating nonphosphorylated carboxylated matrix gla protein predicts survival in ESRD. J Am Soc Nephrol 2011;22:387–95.
8. Westenfeld R, Krueger T, Schlieper G, Cranenburg EC, Magdeleyns EJ, Heidenreich S, et al. Effect of vitamin K2 supplementation on functional vitamin K deficiency in hemodialysis patients: a randomized trial. Am J Kidney Dis 2012;59:186–95.
9. Caluwe R, Vandecasteele S, Van VB, Vermeer C, De Vriese AS. Vitamin K2 supplementation in haemodialysis patients: a randomized dose-finding study. Nephrol Dial Transplant 2014;29:1385–90.
10. Krueger T, Schlieper G, Schurgers L, Cornelis T, Cozzolino M, Jacobi J, et al. Vitamin K1 to slow vascular calcification in haemodialysis patients (VitaVasK trial): a rationale and study protocol. Nephrol Dial Transplant 2014;29:1633–8.
11. Speeckaert MM, Devreese KM, Vanholder RC, Dhondt A. Fondaparinux as an alternative to vitamin K antagonists in haemodialysis patients. Nephrol Dial Transplant 2013;28:3090–5.