Impact of Blood Sampling on Circulating Tissue Inhibitors of Metalloproteinases

To the Editor: I read with great interest the article by Ruokolainen et al. (1) reporting on the clinical validity of tissue inhibitor of matrix metalloproteinase (MMP)-1 (TIMP-1) in head and neck squamous cell carcinoma. Comparing the preoperative serum TIMP-1 concentrations and the immunohistochemical staining of TIMP-1 in tumor sections, they showed that increased TIMP-1 in both compartments is a suitable indicator in predicting the clinical outcome of these tumor patients. The determination of circulating TIMP-1 as reflection in the tissue could be a promising biomarker to assess the preoperative situation and the follow-up of patients. However, more attention to the interference effect of blood collection on the circulating TIMPs should be given before further studies with more patients as suggested by the authors. The issue whether serum or a special plasma should be used for TIMP determination was discussed in analytic journals (2–4). I recommend considering the possible preanalytic pitfalls in the measurement of circulating TIMPs depending on the blood sample used to improve the interpretation of data in future.

To make the readership aware of that problem, I refer to my own experiments summarized in Fig. 1 (3, 4). Briefly, blood samples from 10 healthy volunteers were simultaneously collected either in plastic tubes supplied with kaolin-coated plastic granulate (S-Monovette tubes 01.1601, Sarstedt, Nümbrecht, Germany) to obtain serum or in devices coated with heparin or EDTA to obtain plasma. The tubes were centrifuged within 30 minutes after venipuncture (1,600 × g, 15 minutes). TIMP-1 and TIMP-2 were measured using ELISA kits from Amersham (Little Chalfont, Buckinghamshire, United Kingdom). The TIMP-1 concentrations were higher in serum than in plasma, probably due to the release of TIMP during the platelet activation. The inhibitory effect of heparin on the determination of TIMP-1 was excluded by recovery studies. In contrast, TIMP-2 values were higher in heparin plasma than in serum or EDTA plasma and increased depending on the heparin concentration (data not shown), probably because of the interaction of heparin with proMMP-2 in the pro-MMP-2/TIMP-2 complex (3).

In conclusion, serum does not seem the appropriate sample for determining circulating TIMP-1 whereas the TIMP-2 determination is interfered by heparin. Citrate plasma was suggested as sample of choice for determining circulating MMP-2 and MMP-9 (5) whereas similar recommendations about TIMPs are lacking thus far. Detailed analytic studies are advisable to elaborate practical guidelines for blood sampling to avoid pitfalls in the measurement of circulating TIMPs as biomarkers.

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References
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