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Correspondence

Total keratin 18 measurements in patients following orthotopic liver transplantation: What is the most suitable diagnostic assay?

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Dear Editor,

In patients following orthotopic liver transplantation from deceased donors, total keratin 18 (K18) measurement was recently shown to be favourable in terms of its prognostic value for early identification of a complicated course in comparison to routine markers of liver impairment (e.g., aspartate-amino-transferase, alanine-amino-transferase, lactate-dehydrogenase).[1] In this investigation, the M65 enzyme linked immunosorbent assay (ELISA) kit (#10 020; Peviva AB, Bromma, Sweden) was used for quantitative determination of total K18. In the meantime, the manufacturer (Peviva AB, Bromma, Sweden) launched the M65 EpiDeath ELISA (#10 040) as a new tool for total K18 monitoring, which was intended for the same applications as for the M65 ELISA. The M65 EpiDeath ELISA provides improved measuring characteristics with regard to dilution linearity and spiking recovery. Moreover, it is suspected to allow for a better discrimination between patients with very low and very high levels of K18 due to an extended measuring range of 5000 U/l, compared to 2000 U/l of the M65 ELISA and lower baseline values in healthy controls.

However, up to now the use of M65 and M65 EpiDeath ELISA has not been compared in patients following orthotopic liver transplantation, why it has been unclear, if one of the two assays may represent a more suitable method for determination of total K18 in these patients. Therefore, all patients participating in the CaspAct-LTPL (Caspase Activation in Liver Transplantation)-Trial (German Clinical Trials Register (DRKS)-ID: DRKS00003434/Ethics Committee of the Medical Faculty of Heidelberg; Trial-Code no.: S055-2009/$n=150$) were evaluated for short-term complications within 10 days after orthotopic liver transplantation from deceased donors as described previously.[1,2] Furthermore, plasma samples were collected prior to surgery (Pre), immediately following the end of the surgical procedure (T0), as well as 1 day (T1), 3 days (T3), 5 days (T5) and 7 days (T7) later.[1,2] Plasma levels of total K18 were measured using the M65 and the M65 EpiDeath ELISA-kits according to the manufacturer's instructions at different timepoints. As a result, the M65 and the M65 EpiDeath ELISA revealed a highly similar pattern of total K18 plasma values. Both ELISAs showed increasing levels of total K18 after the end of the surgical procedure reaching peak concentrations at T1, followed by continuously decreasing levels until T7 (Figure 1). Accordingly, total K18 results of both ELISAs revealed a high positive correlation (Pearson's product-moment correlation coefficient: $r=0.882$/Spearman's

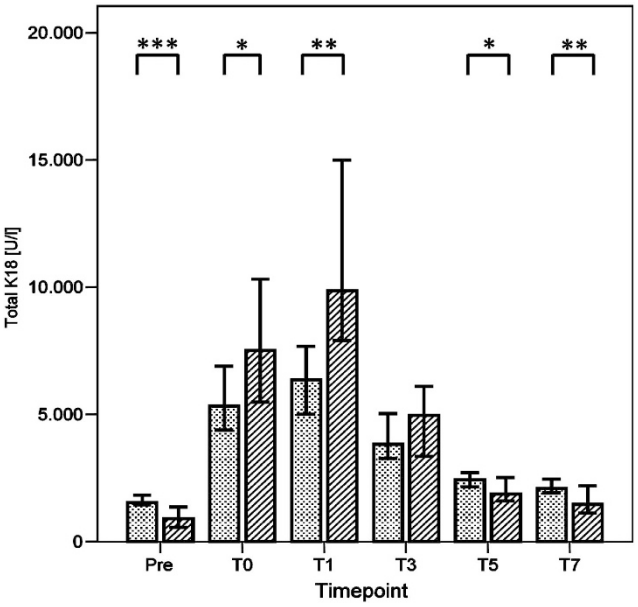

Figure 1 Comparison of total keratin 18 (total K18) measurements using the M65 and the M65 EpiDeath ELISA in patients following orthotopic liver transplantation at baseline, immediately after the end of the surgical procedure as well as 1 day, 3 days, 5 days and 7 days later. Plasma levels of total K18 were measured using the M65 (black spotted bars) and the M65 EpiDeath (black striped bars) ELISA from plasma samples of patients following orthotopic liver transplantation from deceased donors ($n=150$) at Pre (baseline level prior to transplantation), as well as at 5 other times, T0 for immediately after the end of the surgical procedure, T1 for 1 day, T3 for 3 days, T5 for 5 days and T7 for 7 days after liver transplantation. Data in bar charts are given as median and the 95% confidence interval. Concerning symbolism and higher orders of significance: $^{*}P<0.05$, $^{**}P<0.01$, $^{***}P<0.001$

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rank correlation coefficient: $\rho = 0.906$). However, probably as a result of different performance characteristics, the M65 and the M65 EpiDeath ELISA revealed significant differences depending on the amount of circulating total K18 in human plasma. In the low concentration K18 range (e.g., at Pre, T5, T7), the M65 ELISA revealed significantly increased levels in comparison to the M65 EpiDeath ELISA. In contrast, in the high concentration range (e.g., T0, T1, T3) results of the M65 EpiDeath ELISA were shown to be significantly elevated compared with the M65 ELISA (Figure 1). Concerning their ability to identify patients with a high risk for a complicated course, the M65 and the M65 EpiDeath ELISA-based measurements of total K18 revealed a comparable prognostic value, as assessed by comparisons of the areas under the two correlated receiver operating characteristic (ROC) curves (as described by DeLong *et al.*[3]; complications at all → M65 at T1: ROC-area under the curve (AUC): 0.661/M65 EpiDeath at T1: ROC-AUC: 0.683/$P = 0.106$).

In conclusion, the M65 EpiDeath ELISA is at least equivalent to the M65 ELISA in terms of its prognostic value for early identification of a complicated course in patients following orthotopic liver transplantation from deceased donors.

Conflict of Interest

The authors declare no conflict of interest.

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