High-resolution glycosylation profiling of prostate-specific antigen for evaluating its diagnostic potential of prostate cancer

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As an early detection method for prostate cancer (PCa) the concentration of PSA in the circulation is determined. However, the concentration determination lacks specificity for PCa, as increased PSA concentrations can also be found with benign hyperplasia. The lack of specificity indicates the necessity of a more specific test to prevent unnecessary biopsies. Previous studies have reported that PCa can be distinguished from benign hyperplasia by investigating the post translational modification of PSA, namely glycosylation.[1-3] To date, the used methods for determining the glycosylation of PSA suffer from interferences from other glycoproteins. This study addresses the mentioned shortcoming by measuring specifically the glycosylation of PSA by using a high resolution mass spectrometry platform. By studying the glycosylation of PSA of PCa patients and controls with benign hyperplasia we expect to observe altered glycosylation patterns characteristic of PCa which will hopefully help to improve the diagnosis of PCa.

Figure 1. MALDI-TOF-MS profile of the PSA N69-K70 tryptic dipeptides carrying the heterogeneous PSA N-glycans with derivatized sialic acid moieties. Commercially acquired PSA was reduced, alkylated and digested with trypsin, sialic acids were stabilized enabling us to distinguish between α2,3- and α2,6-linked sialic acids. Mass spectra were acquired by matrix-assisted laser desorption/ionization (MALDI)-time-of-flight (TOF)-mass spectrometry (MS). We were able to identify 40 distinct glycoforms, containing mono- and disialylated species, with or without core fucosylation (not all displayed in the Figure).

We used an analytical platform with high sensitivity for the analysis of N-glycosylation of PSA (Figure 1). Commercially acquired PSA was derivatized, allowing to distinguish between the isomers of the differentially linked sialic acids. This differentiation could be of importance as α2,3-linked sialic acids seems to be a hallmark of malignant PCa. This can be explained by the fact that α2,3-linked sialic acids are needed for the formation of sialyl-LewisX structures which are implicated in cellular motility[4].
In summary, with our current method we were able to distinguish 40 different \(N\)-glycans from a single \(N\)-glycosylation site (N\(_{69}\)) of PSA. The used method will be applied on PSA derived from urine of patients. For this analysis, the glycosylation of PSA will be determined after affinity capturing next to the determination of the PSA concentration. By studying the glycosylation of PSA in PCa we have a promising tool to identify disease related alterations of glycosylation, which will be evaluated for its diagnostic and prognostic potential.

References: