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Re: Florian Jentzmik, Carsten Stephan, Kurt Miller, et al. Sarcosine in Urine after Digital Rectal Examination Fails as a Marker in Prostate Cancer Detection and Identification of Aggressive Tumours. *Eur Urol* 2010;58:12–8

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Conflicts of interest: Christopher Beecher was previously an employee of Metabolon. Christopher Beecher, Danny C. Alexander, Jeffrey R. Shuster, and Alvin Berger own equity in Metabolon. Arul M. Chinnaiyan joined the scientific advisory board of Metabolon in July 2008. The University of Michigan has licensed the diagnostic field of use of the metabolomic biomarkers discussed in this manuscript to Metabolon (Arul M. Chinnaiyan and Arun Sreekumar are named as inventors).

In the paper published by Jentzmik et al. [1], the authors address the importance of urine-derived sarcosine based on public interest in our report [2], which describes elevated levels of the metabolite in urine of biopsy-proven prostate cancer (PCa) patients. We found especially elevated sarcosine levels in tumor specimens from patients with metastatic PCa, compared with organ-confined tumors. In their paper [1], the authors have examined urine supernatants collected after digital rectal examination (DRE) from 139 patients with prostate-specific antigen (PSA) levels between 2 and 20 ng/ml. These patients included 106 patients with PCa and 33 individuals with no evidence of malignancy (NEM), as assessed by biopsy. A total of 99 patients in this cohort had PSA levels between 0 and 10 ng/ml.

Jentzmik et al. [1] report that ratio of free to total PSA (%fPSA) performed significantly better than sarcosine in delineating PCa from NEM, with an area under the curve (AUC) of 0.81 versus 0.63 ($p = 0.012$), whereas PSA performed similarly to sarcosine (AUC: 0.64 vs 0.63; $p = 0.933$). When analysis was restricted to patients with a total PSA range of 0–10 ng/ml, %fPSA performed better than sarcosine and PSA (AUC: 0.79 vs 0.67 vs 0.59). Based on this information, Jentzmik et al conclude that measurement of sarcosine in urine after DRE is hardly suitable to improve the diagnostic performance in comparison with routine markers like %fPSA.

Our studies, reported in *Nature* [2], did not aim to establish sarcosine as a clinical test in urine to detect PCa. Rather, we reported results of a proof-of-concept study in which unbiased metabolomic profiling was used to delineate tissue-derived biomarkers for PCa aggressiveness and prognosis, of which sarcosine also could be detected in urine and, additionally, had a role in PCa progression. The intent of the initial study was to demonstrate the power of metabolomics in the identification of potential biomarkers of cancer. In this study [1], using post-DRE urine-derived sediments and supernatants, sarcosine levels were observed to be significantly elevated in biopsy-positive patients *versus* negative controls, indicative of its *potential* to serve as a biomarker.

Notably, using an independent set of 40 urine sediments ($n = 20$; each from biopsy-positive patients and negative individuals) (unpubl. data), we have recently confirmed our earlier findings on elevated levels of sarcosine in biopsy-positive PCa patients (Fig. 1). It is recognized that the use of urinary sarcosine in clinical implementation is in its early stages, and its diagnostic value can be ascertained only after a series of comprehensive blinded validation studies.

The AUCs of 0.67 and 0.59 that Jentzmik et al. [1] report for sarcosine and PSA, respectively, in patients having PSA levels between 0 and 10 ng/ml are similar to the ones reported in the *Nature* paper [2] from an independent set of 110 samples (ie, 0.69 and 0.53, respectively). These studies comparing sarcosine to PSA are independent validations of the initially reported observation. Furthermore, in our report [2], we used caution when describing the biomarker potential of urine-derived sarcosine, as is evident by the following statement: “The overall receiver operating characteristic curves for sarcosine indicate that its predictive value is modest, with an area under the curve (AUC) of 0.71 for urine sediments and 0.67 for supernatants (Supplementary Fig. 14b, c).”

In summary, we maintain that sarcosine, when multiplexed with other metabolomic markers and perhaps with other diagnostic modalities, will have the potential to increase the accuracy of detecting PCa in future. We agree that %fPSA could be a valuable parameter to include when developing such multiplex clinical tests. Moreover, tumor and urinary sarcosine may prove to be highly predictive of prognosis, which would be very helpful for clinical decision making.

References

1. Jentzmik F, Stephan C, Miller K, et al. Sarcosine in urine after digital rectal examination fails as a marker in prostate cancer detection and identification of aggressive tumors. *Eur Urol.* 2010; 58:12–8. [PubMed: 20117878]
2. Sreekumar A, Poisson LM, Rajendiran TM, et al. Metabolomic profiles delineate potential role for sarcosine in prostate cancer progression. *Nature.* 2009; 457:910–4. [PubMed: 19212411]

Fig. 1. Box plot of sarcosine levels based on isotope-dilution gas chromatography–mass spectrometry analysis showing median sarcosine-to-alanine levels in urine sediments from an independent group of biopsy-positive and -negative individuals.