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EXTENDED ABSTRACT

Analysis of selenium status from dried blood spots by total-reflection X-ray fluorescence analysis ☆

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Selenium (Se) takes a prime position for human health among the essential trace elements (Rayman, 2012). Se is not only essential for male fertility (Michaelis et al., 2014), but also for growth, differentiation and thyroid hormone metabolism (Dumitrescu et al., 2005). A severe Se deficit has been associated with subfertility, increased risk of chronic diseases including cancer, cardiovascular disease, type 2 diabetes, mostly related with oxidative stress. However, it is difficult to determine the Se status of a given individual due to the need for specialized methods and expensive instrumentations. In the clinics, congenital diseases are detected from small amounts of full blood taken shortly after birth, subsequently dried and stored on a filter paper as dried blot spots (DBS) (Vacchina et al., 2014). This technique has been meanwhile successfully adopted to different medical disciplines, from epidemiology via therapy monitoring to toxicology (Stove et al., 2012) Figure 1.

The principal advantages of the DBS technology are an easy collection of samples, storage as well as transport at room temperature, together with well-characterized handling procedures. In order to test whether the Se status can also be reliably determined from DBS by total reflection X ray fluorescence (TXRF) analysis various analytical parameters were compared.

DBS samples are a problematic and challenging matrix, as the dried blood constituents may have changed structure, and potentially undergone stable associations with the DBS material. For this reason additional digestion or other homogenization steps prior to TXRF analysis may be necessary. In general, the concentrations of essential elements like Cu, Zn, Co and Se are in linear fit when comparing human serum and full blood samples (Barany et al., 2002).

Comparing different desorption techniques, an incubation of isolated DBS in concentrated HCl (37%) as compared to H_2O, concentrated HNO_3 (65%), or a combination of HNO_3 (65%) plus H_2O_2 (30%), yielded the best recovery of Se. The application of sonication during the desorption process or an addition of polyvinylic alcohol was not improving the recovery rate. The comparison of glass and acryl-plastic carriers available for TXRF analyses showed a similar performance for the majority of trace elements analyzed with serum or tissue homogenates; only the measured Ca concentrations differed by >10% between these carriers. Investigating blood samples from ten healthy adults, the Se concentrations determined from undried blood and DBS samples showed a high degree of concordance ranging from 88% to 99%, while recovery of other trace elements was less reliable and differed significantly from the reference. Under the optimized conditions, the detection limit of Se from DBS was determined at below 5 μg Se/L.

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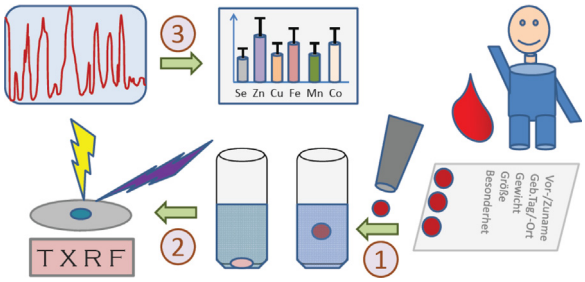

Figure 1 Workflow for dry blood spot (DBS) analysis. Probands apply a single drop onto a predefined area of a membrane documenting person- and purpose-specific details. A defined circle is punched out and transferred into a desorption solution (1). The reaction mix is incubated and an aliqout is taken (2), supplemented with an internal standard and used for total-reflection X-ray (TXRF) analysis. The resulting spectrum is converted into absolute concentrations of the different elements (3).

We conclude that Se analyses from DBS by TXRF analysis is a versatile option, given that appropriate desorption solvents and incubation methods are chosen. The results from these measurements are compatible with the data obtained from undried samples and thus open the perspective of systematic evaluation of the Se status during neonatal screening processes or from stored epidemiological DBS samples. Moreover, especially with respect to the field studies in less developed countries or regions, the herein described technique may be of substantial benefit, as costly collection procedures and fast sending of refrigerated samples become unnecessary.

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Conflict of interest

The authors declare that there is no conflict of interest.

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