FACTORS INFLUENCING DRYING PROCEDURE OF SAMPLES FOR TRACE METALS ANALYSES.

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Introduction

In biomonitoring and ecotoxicology, clean and accurate sample preparation is required, in respect of potential low quantity of the analyte and non-homogeneity of material (Kebbekus and Mitra, 1998). In the case of trace metals analyses, the dry weight of tissues (d.w.) should also be obtained, as levels of those metals are more accurately assessed in samples of the same hydration state.

Many scientists dry samples at 105°C or 100°C, while the others at 80°C or 60°C. The discrepancy in temperature used may lead to different d.w. obtained, thus the final concentrations may be different. During the drying procedure adequate temperature and time are crucial. Other factors, like the storage, laboratory materials used and detailed type of the sample, may also influence the drying protocol. Frozen samples are being used by some scientists, while the others use fresh samples. Due to the fact of ice formation during storage at -18° and subsequent destruction of many structures (Bancroft et al., 2008) there is a possibility to observe differences between d.w. obtained after different storage. Additionally, the differences in samples' dry weight obtained drying on dishes made of different materials, e.g. glass and plastic (Binkowski et al.,2012) may be suspected, as well as, the influence of the detailed tissue type (e.g. healthy and cancerous lung tissue) (Błaszczyk et al., 2016).

This work investigates subsequent issues 1) the rate of water loss from samples which defines the minimal oven drying time of the tissues, which is required to obtained the constant d.w. 2) the differences in water loss dynamics between different tissues 3) the influence of the sort of weighing plate (glass or plastic one) on the obtained d.w. This brings us the information how to handle drying protocol to avoid potential sources of errors, as well as, to what time we can shorten the drying procedure to obtain reliable results.

Methods

The research was conducted on birds' and human tissues (0.5-1.0 g of wet weight). Mallard's (*Anas platyrhynchos* L.) samples (brain, pectoral muscle, lung, liver, kidney, intestine, spleen and bone) were taken from individuals (n=24). The samples were separated into 3 experiments: effect of temperature (60°C and 105°C), plate and storage. The d.w. was measured in consecutive controls: 1, 2, 4, 7, 11, 18 and 28 days after starting the drying.

Human samples were collected from 77 patients during a surgery. From each patient two samples of cancerous and seemingly unaffected tissue fragments have been collected. Before analysis, the samples were put into dryer at 60°C. For the first three days the samples had been weighed every 12 hours and further (4th -13th day) every 24 hours until the weight was stable. All the results were shown as means of d.w. percentage. The factorial ANOVA was used to determine the differences between consecutive measurements.

Results

We observed the fastest water loss between 1st and 4th day of the drying. The frozen samples (liver, spleen and intestine) drying in glass plates lost nearly the same amount of water in both temperatures (60°C and 105°C) (Fig. 1). Our results indicated that due to plate factor (glass or plastic) the d.w. differed in the case of kidney (p=0.0007) and spleen samples (p=0.0334). Moreover, only in the case of liver samples the significant variability according to the freezing (p=0.0028) and its interaction with the plate factor (p=0.0118) were observed.

The percent of the d.w. was higher in lung unaffected samples than in the lung cancerous samples (Fig.1). Human samples were visibly losing their weight up to 72 hours and the highest decrease in weight loss was found in the first 24 hours of drying in both tested tissues.

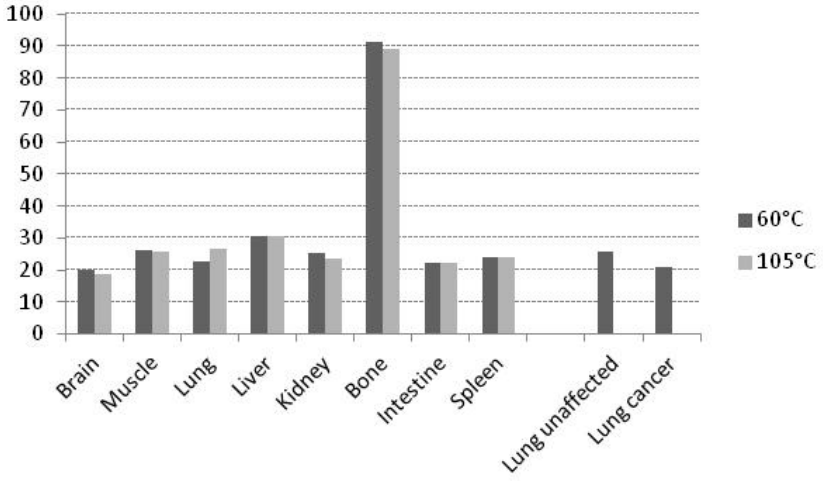

Figure 1. % of the d.w obtained at different temperatures (60°C, 105°C) from bird' and human (unaffected and cancer) lung tissue .

Conclusion

The drying temperature, as well as the drying time, the material of which the weighing dishes are made and freezing the material significantly influence the d.w. Our results revealed that the final d.w. percentage is influenced by the tissue type. We also suggest to shorten the drying period to 72 hours (longer drying does not cause a significant water loss). This allows us to save the money and the time, which is of special importance for the medical patients for whom both the duration of the analyses and the costs are crucial.

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