Testing of a new sampling procedure for airborne allergens in the workplace atmosphere

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Introduction

The standard procedure for sampling of airborne allergens at workplaces is the filtration method. Allergen samples from workplace air are normally taken with a sampling volume of 10 L · h^{-1} for at least four hours on Teflon-filters (PTFE). For the sampling of allergens in indoor air a new device has been developed (AS 100, Holbach Umweltanalytik, Wadern, Germany). The measuring principle of this device is separating of allergenic particles from the air with a sampling volume of 100 L · min^{-1} directly into the test receptacle which is used for the allergen-test using an Enzyme-linked-immunosorbent assay (ELISA).

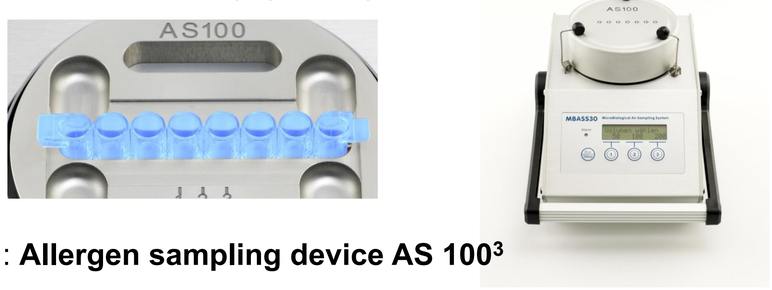

Fig. 1: **Allergen sampling device AS 100[3]**

In 2013 IFA initiated a research project focused on the suitability of the new sampling device for the determination of airborne allergens at workplaces.

Materials and Methods

Reduction of sampling time because of the higher sampling volume may be advantageous to describe the allergen concentration e.g. at indoor-workplaces or for short-time measurements. Furthermore it was the idea to save the extraction step by direct sampling of the air into the test receptacle for the ELISA. This procedure might be an advantage in quantitative sample analysis, especially for the proof of small allergen concentrations. For this reason, the AS 100 is operated with single stripes of microtiter plates containing 8 wells as sampling units (MTS). In 2013 three measurements were done in different areas of cow stables accompanying activities like feeding or dividing of straw material at three different cattle farms. Both sampling systems were used in parallel (Fig. 2).

Fig. 2: Field sampling of Bos d2 in a cowshed

Sampling on PTFE-filters (Ø 37 mm) was performed with 10 L · min^{-1} for 60 min using GSP-Sampling-heads and pumps, SG 10, GSA, Germany. In MTS allergen-containing air was sampled for 3 x 20 min, using AS 100 and MBASS 30, Holbach Umweltanalytik, Germany. Filter samples were extracted with 5 ml PBS + 0.05 % Tween 20 in Falcon-tubes (15 ml, PP) at room-temperature for 60 min on a shaking device. Solution without filter was centrifuged (30 min, 3,000 g) and the supernatant was used for analysis (storage at -20 to -70 °C). The major cow hair and dander allergen was determined with the Bos d2 ELISA-Kit from Indoor Biotechnologies Ltd., UK. Allergen analysis in MTS was carried out as direct ELISA.

Field sampling

Within three measurements in different areas of the stables accompanying activities like feeding or dividing of straw material at three different cattle farms, Bos d 2 was detectable with both measuring systems.

Fig. 3: Typical activities in a cowshed

Single results varied within a wide range: 57.0 to 1,700 ng Bos d 2 · m^{-3} for filter samples (n=28) and 0.38 to 429 ng Bos d2 · m^{-3} for samples taken with the AS 100 (n=30). Most data received with the AS 100 were lower than results measured by standard filtration. This might partially be owed to the fact, that Bos d2 analysis of the MTS was carried out as direct ELISA without coating antibody and not as sandwich ELISA. To prove this assumption, a recovery experiment was performed with an extract of a material sample (mixture of silage and stable dust). Investigating five dilutions with three replicates each, recovering of Bos d2 was 51 % in direct ELISA compared to sandwich ELISA.

Recovery experiments

To optimize the protocol for the analysis of the MTS as sample holders from direct sampling of airborne allergens with the AS 100, another recovery experiment was carried out with the known material sample (MS), with a standardized test dust (STD) too and with six different mixing ratios of both materials: 1:10, 1:20, 1:40, 1:80, 1:160, 1:320). 1 or 2 mg of every sample (pure MS, mixtures, pure STD) were weighed in seven of eight wells of a MTS, (one well of each MTS was left empty as blank value) with three replicates each for every sample.

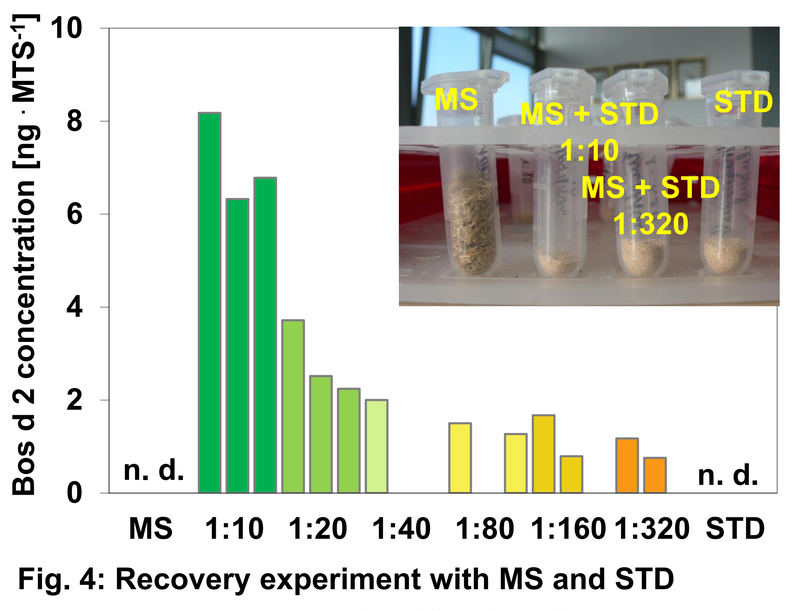

Fig. 4: Recovery experiment with MS and STD

Best results were received for the dilution 1:10. Rough particles restrained analysis of Bos d 2.

Sampling efficiency

To answer the question to what extent the antigen/ allergen containing airborne particle fraction is precipitated in sample units of the AS 100 (= MTS) compared to their collection on filters, sampling devices were operated simultaneously in a measuring channel of the IFA, using Jeloxyl haho 120 f (particles 10-200 µm) as standardized test dust. Flow rate in the channel was 0.5 m · s^{-1}, dust concentration was 60 mg · m^{-3}.Sampling time was 10 minutes, mass of STD in the wells was determined gravimetically.

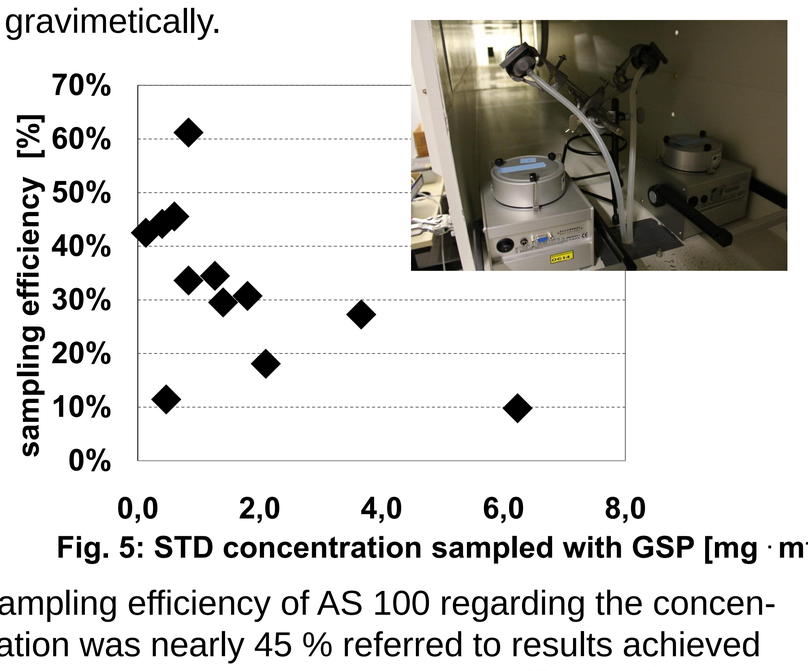

Fig. 5: STD concentration sampled with GSP [mg · m^{-3}]

Sampling efficiency of AS 100 regarding the concentration was nearly 45 % referred to results achieved with the GSP-system. It was more efficient for the smaller particles and lower concentrations.

Summary and Outlook

Previous results of the experiments in the measuring channel with an aerosol from standardized test dust showed, that it is possible to sample particles from the air using the AS 100 sampling device for airborne allergens. The protocol for the analysis of the MTS has been optimized. Sampling efficiency and analytical efficiency were better for small particles and lower concentrations. Further experiments concerning the sampling efficiency are planned within a bioaerosol chamber at least with mite allergens (Der p1, Der f1) but hopefully also with standardized cowshed dust.

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