

## RETENTION AND RECOVERY DETERMINATIONS FOR THE MYCOTOXIN, OCHRATOXIN A, USING VARIOUS FILTER MEDIA

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### ABSTRACT

The fungal mycotoxin ochratoxin A (OA) has received wide attention as a result of its prevalence in grains, cereals, and other foodstuffs, as well as the negative health effects it can have in humans and animals from ingestion. Research has shown that inhalation of OA contributes to the overall body burden of the toxin and that significant exposures can occur in the work environment. Currently there is no standardized method for the sampling of airborne OA that could be used to develop exposure based risk assessments. This study evaluated the recovery and stability of OA on three filter media: glass fiber, polycarbonate, and polyvinyl chloride under laboratory conditions. The mycotoxin, in a methanol solution, was spiked onto the surface of each filter using a micro-syringe; one set of samples were attached to air pumps operating at 2 lpm for 8-hours, the remaining samples received no additional treatment. The percent recovery of OA from the filters that did not have air drawn through them was similar for all three filter media, however there was high intra-variation in the recovery between samples in each filter group. The samples that had air pulled showed a definite trend in percent recovery with PVC (72.5 %) and polycarbonate (46.9%) performing significantly better than glass fiber filters (19.1%).

### INTRODUCTION

The mycotoxin ochratoxin A (OA) is a toxin produced as a by-product of metabolism in *Penicillium verrucosum* in temperate climates, and by several species of *Aspergillus* in tropical and subtropical climates. It has been largely studied as a contaminant in foods and has been detected in samples of cereals, (10) wheat and barley grains (20,31), coffee, cocoa, spices (15), pig-liver pate (17), wine (25), and dried vine fruits (1). Based on the adverse human health effects associated with ingestion of the mycotoxin, tolerable intakes of 100 ng/kg of body weight/week of OA have been suggested by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) (11).

Chronic exposures to OA have been shown to lead to nephrotoxic, genotoxic, teratogenic, carcinogenic, and immunosuppressive effects in mammals (14). Ingestion of the mycotoxin in cereals and other foodstuffs has been linked to a chronic kidney disease (Balkan endemic

nephropathy (BEN)) that affects wide populations in the Balkan region (29). OA has a half-life in the human body of 35 days (32), and has been found in samples of human blood and breast milk (27).

DiPaolo attributed OA found in *Aspergillus ochraceus* contaminated wheat to be the cause of respiratory symptoms in a farmer and acute renal failure in his wife; both had worked for 8-hours sieving grain (6). OA is very efficiently absorbed from the lung as seen in experiments where rats have received intratracheal administration of the mycotoxin (2). Though the inhaled intake of OA is typically small as compared to dietary intake, inhalation can contribute to the total toxic load of the body. This is especially true for activities that create high airborne concentrations in a work or indoor environment. OA levels of 1500 µg/kg were found in dust collected from the inside of the ducts of a home heating system (22); disturbances of this dust could result in significant exposure to the toxin. Because of respiratory toxicity and inhalation as a route of systemic absorption, it is necessary to evaluate and quantify not only ingested OA, but also airborne exposures that can occur in contaminated environments.

Methods of analysis for OA and other mycotoxins were first developed and standardized for bulk food samples (5, 12,16,28). It has only been recently that efforts have been made to translate bulk methods into effective airborne sampling methods. The creation of an effective, standardized method for sampling airborne OA is the necessary first step in creating an occupational exposure limit. By measuring a specific component of bioaerosols rather than surrogates, such as total dust levels, it allows for an evaluation of the relevant etiological agent and a more accurate determination of the exposure-effect relationship (9).

Filter sampling is the preferred method for collecting airborne bioaerosols in the work environment because it is convenient and efficient, sampling time can be varied, and analysis can be performed using a large range of methods. A potential disadvantage to filter sampling is that some bacteria, fungal cells, and spores lose culturability as a result of desiccation (3). Constituents of microorganisms, such as mycotoxins, are more stable and therefore can be collected with a filtration method and measured using non-culture-based methods (9).

In one study, airborne OA was collected using 25-mm glass fiber filters in coffee, cocoa, and spice processing plants. The subsequent analysis by HPLC showed levels ranging from <0.003 ng/m<sup>3</sup> for an office worker to 8.15 ng/m<sup>3</sup> for a packer (15). In another study, HPLC analysis positively identified OA in samples of dust collected from dairy farms using 25-mm polycarbonate filters (pore size, 0.4 µm) (26).

Studies comparing various filter media for collection and analysis of OA have not been performed, however there have been comparative media studies for endotoxins and other mycotoxins. Airborne endotoxin was analyzed from dust in a potato processing plant using glass fiber (25-mm), Teflon (25-mm; pore size, 1.0 µm), polycarbonate (25-mm; pore size, 1.2 µm), and mixed cellulose ester (MCE) (25-mm; pore size, 1.2 µm). The amount of endotoxin extracted from the glass fiber, Teflon, and polycarbonate filters was twice that extracted from the mixed cellulose ester filters. It was suggested that endotoxin irreversibly binds with MCE more than the other filter materials (7). Side-by-side airborne endotoxin measurements were performed using the All-Glass-Impinger (AGI) and polyvinyl chloride (PVC) (pore size, 0.8 µm)

in swine barns and sawmills. In sawmills, impinger samples gave significantly higher endotoxin concentrations and lower variance than the PVC samples. In swine barns, where the endotoxin concentrations were 10-fold higher on average than in the sawmills, sample results showed that impinger and filter methods yielded comparable endotoxin concentrations, however the variability was lower using the impinger method (8). Impingers can be effectively used in occupational environments for area sampling, however they are often impractical for personal sampling.

A laboratory study comparing various filter media was performed using a chamber filled with airborne *Stachybotrys Atra Corda* contaminated materials. Cellulose-nitrate filters (37-mm; pore size, 0.8  $\mu\text{m}$ ), and polycarbonate filters (37-mm; pore size, 0.4  $\mu\text{m}$  and 0.2  $\mu\text{m}$ ) were evaluated and it was determined that the 0.2  $\mu\text{m}$  pore size polycarbonate filters gave the best results in this experimental design. It was hypothesized that this may have been a consequence of pore size (21).

OA can be effectively collected as airborne particle samples, however no standardized method for sample collection and analysis exists. A fundamental step in creating a standardized method is determining which filter media is the most effective for the specific analyte. This study evaluated percent recovery and analyte stability for OA using three different filter types: glass fiber, polyvinyl chloride, and polycarbonate.

## METHODS AND MATERIALS

110 ng of OA in a methanol solution was spiked onto samples of three different filter media randomly divided into two groups. Group 1 received a simple spiking and was then analyzed for percent OA recovery. In Group 2, air was pulled through spiked samples for 8-hours and percent recovery was calculated to determine analyte stability. The criterion for successful recovery of analyte for each filter type was based on protocols established by the National Institute for Occupational Safety and Health (NIOSH), which states that recovery should differ by no more than  $\pm 25\%$  from the target concentration 95 times of 100 (4). The target concentration of OA was based on the JECFA's weekly tolerable intake recommendations.

For the experiment, three types of 37-mm diameter filters: glass fiber filters (Gelman filters, pore size 1.0  $\mu\text{m}$ , Pall Corporation, Ann Arbor, MI), polycarbonate filters (Gelman filters, pore size 1.0  $\mu\text{m}$ , Pall Corporation, Ann Arbor, MI), and polyvinyl chloride filters (pore size 0.5  $\mu\text{m}$ , Mine Safety Appliances Company, Pittsburgh, PA), were mounted in 2-piece closed-face cassettes. All cassettes had 37-mm cellulose back-up pads. A solution of methanol and 98% certified pure ochratoxin A, as determined by thin layer chromatography, was prepared by Trilogy Analytical Laboratory to a concentration of 2.0 ng/ml. (Trilogy Analytical Laboratory, Inc., Washington, MO.) The solution was stored at 5 °C for three weeks before use.

### Percent Recovery

Group 1 consisted of six samples of each of the three filter types. 55  $\mu\text{l}$  aliquots of the 2.0 ug/ml solution of OA in methanol were applied directly to the surface of the filter via the inlet hole of the cassette holder using a 200  $\mu\text{l}$  glass micro-syringe (Gas Tight #1725, Hamilton Company,

Reno, NV) for a total weight of 110 ng of OA. Inlet and outlet plugs were replaced once the filters were spiked and samples were stored in a resealable plastic bag at 5 °C before being shipped by overnight mail to the laboratory for analysis. Ochratoxin A was extracted from the filters and back-up pads, and analyzed with HPLC with fluorescence detection by Trilogy Analytical Laboratory.

### **Analyte Stability**

Group 2 consisted of six samples of each of the three filter types. Each filter was spiked with 110 ng of OA in the manner described above. Ten of the eighteen cassettes were randomly selected and attached to one of two manifolds, each with five ports, fabricated with standard brass fittings. The cassettes were attached to a high-volume pump (GAST Model 1531, Gast Manufacturing, Benton Harbor, MI) with Tygon tubing (Saint-Gobain Performance Plastics Corporation, Akron, OH). The eight remaining samples were stored in a resealable plastic bag at 5 °C for 8.5-hours until use.

Airflow into each manifold port was calibrated for the specific type of filter media using a DryCal DC-Lite primary flow meter (Bios International Corporation, Butler, NJ). Air (70 °F, 12.5 % RH) was pulled through each of the cassettes for 8-hours at an average flow of 2.08 l/min (range 1.960-2.079). Flow rates were calibrated at the start, middle, and end of each run. At the end of the 8-hour period the cassettes were removed from the manifold, inlet and outlet plugs were replaced, and samples were stored at 5 °C. The process was repeated with the remaining eight cassettes. Samples were transported to Trilogy Analytical Laboratory and OA was extracted from the filters and back-up pads and analyzed with HPLC with fluorescence detection.

### **Analysis of Ochratoxin A by HPLC**

Each filter and paper support was extracted in 50ml of methanol/water (70/30) by shaking for 1 hour. The sample extract was filtered and 4.0 ml of the sample extract was taken to dryness; approximately 5.0 ml of acetonitrile was added to each tube to aid in the evaporation.

Each sample and spike was reconstituted with 1.0 ml of ochratoxin HPLC mobile phase and 150 µl was injected in to the HPLC. The HPLC conditions were: mobile phase of acetonitrile/water/acetic acid (45/54/1) at a flow rate of 1.0 ml/min through a Brown-Lee RP-18, 5 µm, 10 cm x 4.6 cm column with a fluorescence detector at 460 nm emission and 333 nm excitation. Analysis was performed by Trilogy Analytical Laboratory.

### **Control**

For quality assurance, an OA standard was injected into the HPLC and an average 97% recovery was obtained. Blanks of each filter type were analyzed and blanks did not show any fluorescence interference at the OA retention time.

### **Statistics**

Statistical analysis was carried out using STATA 7.0 statistical software (Statistics/Data Analysis, Stata Corporation, College Station, TX). To determine the success of each filter type

at recovering the analyte, both with and without pulling air, a one sample t-test was performed. The significance between the means for each filter, both with and without pulling air, was evaluated using a Wilcoxon Rank Sum test due to the non-normal distribution of data. The coefficient of variation was calculated to determine which filter that presents the best precision/least variation. A Kruskal-Wallis one-way analysis of variance was performed to compare the differences in recovery between each filter type in Groups 1 and 2, with *post hoc* paired analyses using Wilcoxon Rank Sum tests with a Bonferroni correction.

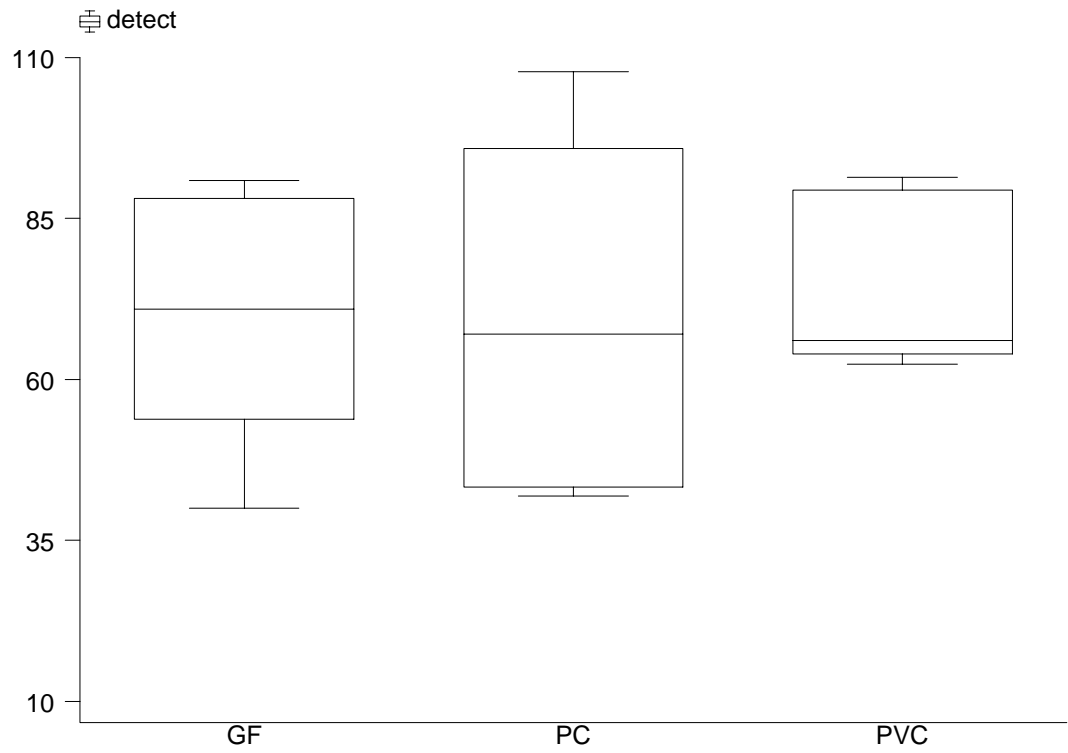
## RESULTS

The results in Table 1 show that there is no significant difference in the percent recovery of OA from the filters in Group 1. The average recovery of OA from the glass fiber filters was 62.8%, polycarbonate 64.1%, and PVC 66.6%. Though the average desorption efficiency was similar for the three filter type, there were large differences in the variability of OA recovered by samples within the same filter type. PVC showed both the best average recovery and the least variability between runs with a coefficient of variance (CV) of 18.3%. Polycarbonate filters returned the second best recovery, however the variability of OA recovered within runs of the polycarbonate filters (CV = 40.4%) was more than double that of PVC. The NIOSH criteria for acceptable analyte desorption requires  $\pm 25\%$  of the target concentration, in this case a range of 87.5-137.5 ng of OA, to be recovered in 95% of trials. Individual runs produced results that met the NIOSH criteria, however the mean recovery for each of the three filter types in Group 1 not pass the criteria when evaluated using a one sample t-test.

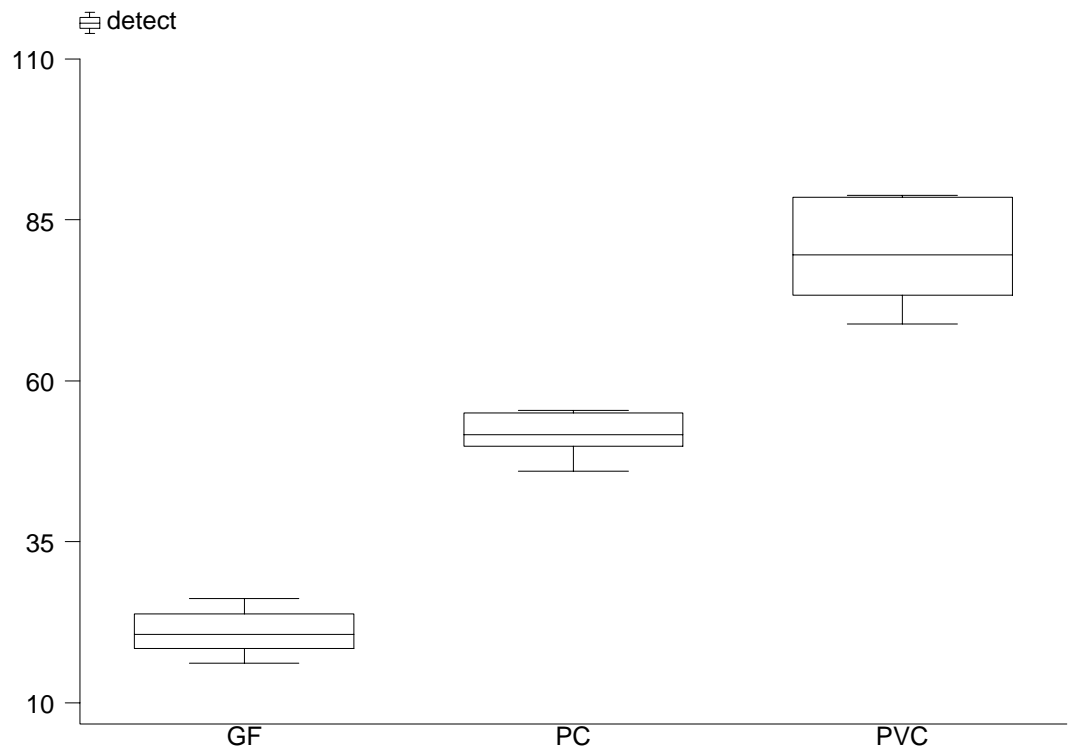
Table 1. OA recovery results.

Filter condition	No. samples	Results in nanograms			CV	% Recovery	% Recovery range
		Mean recovery	Recovery range	SD			
Group 1 – w/o air							
Glass fiber	6	69.1	40.0-90.9	20.7	30.0	62.8	36.4-82.6
Polycarbonate	6	70.5	41.9-107.8	28.5	40.4	64.1	38.1-87.2
PVC	6	73.2	62.4-91.4	13.4	18.3	66.6	56.7-83.1
Group 2 – with air							
Glass fiber	6	21.0	16.1-26.2	3.8	18.1	19.1	14.6-23.8
Polycarbonate	6	51.6	46.0-55.4	3.6	7.0	46.9	41.8-50.4
PVC	6	79.8	68.8-88.8	8.2	10.3	72.5	62.6-80.7

Graph 1. Detection of OA (ng) in samples from Group 1.



Graph 2. Detection of OA (ng) in samples from Group 2.



A comparison of results in Table 1 for Group 1 & 2 show analyte stability after air has been pulled through the samples for 8-hours. A definite trend was identified in the desorption efficiencies of each filter type in Group 2. The largest mean percent recovery was 72.5% found in PVC filters, followed by polycarbonate at 46.9%, and glass fiber with only 19.1% recovery. Variances in recovery between runs for each filter type were markedly reduced when air was pulled through the sampling cassettes. This was seen most dramatically in polycarbonate filters ( $CV_{\text{with air}} = 7.0\%$ ,  $CV_{\text{without air}} = 40.4\%$ ); the two other media showed almost two-fold reductions.

Table 2. Comparison of OA recovery between and among each filter type.

Group 1-without air		Group 2-with air		
Filter type	Mean recovery (ng)	Filter type	Mean recovery (ng)	p-value **
Glass Fiber	69.1	Glass Fiber	21.0	0.0039
Polycarbonate	70.5	Polycarbonate	51.6	0.6310
PVC	73.2	PVC	79.8	0.3367
<b>p-value*</b>	0.778		0.005***	

\*Comparison of recovery for between the filter types in Group 1 using a Kruskal-Wallis one-way analysis of variance test.

\*\*Comparison of recovery, with and without air, for each filter type using a Wilcoxon Rank Sum test.

\*\*\**Post hoc* analyses using Wilcoxon Rank Sum, Glass fiber v. Polycarbonate ( $p= 0.0039$ ), Glass Fiber v. PVC ( $p=0.0039$ ), Polycarbonate v. PVC ( $p=0.0039$ ); Bonferroni correction –  $\alpha^*=0.05/3 = 0/0167$ .

The mean analyte recovery for PVC ( $p=0.3367$ ) and polycarbonate filters ( $p= 0.6310$ ) was not statistically different when comparing samples in Groups 1 and 2 using a Wilcoxon Rank Sum test. The percent recovery for glass fiber filters drops greatly between the series without pulled air (62.8%) and with pulled air (19.1%). When evaluating the desorption efficiency of each sampling media based on the NIOSH criteria, the mean recovery for each of the three filter types did not perform satisfactorily when assessed using a one-sample t-test.

A Kruskal-Wallis one-way analysis of variance test showed that there were no significant differences ( $p=0.778$ ) in the amount of ochratoxin recovered from each media when samples were only spiked with OA. When air was pulled through samples for 8-hours, statistically different recoveries ( $p=0.0005$ ) were obtained for the three media types. Using a Wilcoxon Rank Sum *post hoc* analysis with a Bonferroni correction, no two filter types gave the same recovery (see Table 2).

## DISCUSSION

The data shows that approximately one-third of the analyte could not be recovered from the filters in Group 1. In addition, there was a large variability in the recovery ranges between runs for each filter type. For the series of samples that had air pulled for 8-hours, there is a distinct trend in the recovery patterns between the different filter types as seen in Graph 2. PVC gave the best ochratoxin recovery (72.5%) and glass fiber the worst (19.1%).

In Group 1, the average recoveries of OA from each filter type are very similar. Once air was pulled through the filters, the recovery for glass fiber dropped significantly, polycarbonate filters

showed a moderate reduction, and the change in PVC was minimal. This suggests that OA is most stable on the PVC filters.

The protocol employed in this experiment used methanol as a solvent to extract OA from the filters and support pads. The mycotoxin has been successfully extracted from bulk samples with a methanol solvent (31), however pulling air through the filters may have changed the manner in which the mycotoxin binds to or passes through the surface of the filter. The use of a stronger solvent, such as methylene chloride, may increase the desorption of ochratoxin A.

Analyte loss due to deposition on the inside walls of polystyrene cassettes has been reported in sampling for airborne endotoxins in a fiberglass insulation-manufacturing plant during spray cleaning operations using recycled wash water. Sampling was performed using 37-mm polycarbonate membrane filters in closed-face polystyrene cassettes (33). An evaluation of the cassettes found a large portion of endotoxin deposited on the inside walls. This effect was most pronounced in the dry areas, however it appeared that high humidity levels might have greatly reduced the quantity of endotoxin deposited on the walls. In the case of this experiment, the loss of analyte as a result of deposition on the cassette walls is not a likely explanation because the mycotoxin in solution was deposited directly onto the surface of the filters.

In evaluating the patterns of desorption for samples with pulled air, it can be assumed that the physical characteristics the three media types played a role in analyte recovery efficiency. Polycarbonate filters have a smooth filtering surface and pores that are cylindrical and almost uniform in diameter created by fission track-etching. The collection efficiency is more dependent on particle size than other filter types. Glass fiber filters are created by randomly orienting fibers within the plane of the filter and this creating a highly tortuous airflow path. The collection efficiency of these fibrous filters is highly dependent on face velocity. PVC filters are membrane filters made from gel sheets with uniform pores, and though the production is different than fibrous filters, the tortuosity is similar. Membrane filters differ from fiber filters in that a larger proportion of the analyte deposit is concentrated at or close to the front surface (19).

Ochratoxin A may have expressed an affinity for hydrophobic materials ( $K_{ow}=4.74$ ). This could help to explain why the recovery was dramatically higher for polycarbonate and PVC, both highly hydrophobic polymers, than for glass fiber filters. Current research in the field of mycotoxin, and specifically ochratoxin adsorbents (13), has shown several resins and molecularly imprinted polymers (MIPs) (18) to be very successful in binding the mycotoxin. This technology is based on electrostatic interactions between the molecule and the polymer.

The development of a standardized air sampling method for various mycotoxins is highly analyte dependent. This study has shown that polycarbonate and PVC filters have better analyte recovery than glass fiber filters in samples that have been spiked with OA and have had air pulled through them for 8-hours. The recovery from the glass fiber filters drops significantly when comparing the desorption efficiencies with and without air. The recovery from the PVC filters remains relatively unchanged suggesting greater analyte stability. Additional research will be needed to determine what combination of extraction techniques and filter media will result in a sufficient recovery and low variability for the sampling of airborne OA.

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## REFERENCES

1. Abarca, M. L., Accensi, F., Bragulat, M. R., Castella, C., Cabanes, F. J. 2003. *Aspergillus carbonarius* as the main source of ochratoxin A contamination in dried vine fruits from the Spanish market. *J. Food Prot.* 66:504-506.
2. Breitholtz-Emanuelsson, A., Fuchs, R., Hult, K. 1995. Toxicokinetics of ochratoxin A in rat following intratracheal administration. *Nat. Toxins* 3:101-103.
3. Burge, H. A., Ammann, H. M. 1999. Fungal Toxins and  $\beta$ -(1 $\rightarrow$ 3(-D-Glucans, p. 24.1-24.8. *In* Macher, J. (ed.), *Bioerosols: Assessment and Control*, American Conference of Governmental Industrial Hygienists, Cincinnati, OH.
4. Cassinelli, M.E., O'Connor, P.F. 1994. *NIOSH Manual of Analytical Methods*, 4<sup>th</sup> ed. National Institute for Occupational Safety and Health, Cincinnati, OH.
5. DeSaeger, S., Sibanda, L., Desmet, A., Van Peteghem, C. 2002. A collaborative study to validate novel field immunoassay kits for rapid mycotoxin detection. *Int. J. Food Microbiol.* 75:135-142.
6. Di Paolo, N., Guarnieri, A., Garosi, G., Sacchi, G., Mangiarotti, M. A., Di Paolo, M. 1994. Inhaled mycotoxins lead to acute renal failure. *Nephrol. Dial. Transplant.* 9:116-120.
7. Douwes, J., Versloot, P., Hollander, A., Heederik, D., Doekes, G. 1995. Influence of various dust sampling and extraction methods on the measurement of airborne endotoxin. *App. Environ. Microbiol.* 6: 1763-1769.
8. Duchaine, C., Thorne, P. S., Meriaux, A., Grimard, Y., Whillen, P., Cormier, Y. 2001. Comparison of endotoxin assessment by bioaerosol impinger and filter-sampling methods. *App. Environ. Microbiol.* 67: 2775-2780.
9. Eduard, W., Heederik, D. 1998. Methods for quantitative assessment of airborne levels of noninfectious microorganisms in highly contaminated work environments. *Am. Ind. Hyg. Assoc. J.* 59:113-127.
10. Eskola, M., Parikka, P., Rizzo, A. 2001. Trichothecenes, ochratoxin A and zearalenone contamination and fusarium infection in Finnish cereal samples in 1998. *Food. Addit. Contam.* 18:707-718.
11. FAO/WHO/UNEP. 1999. Conference proceedings from the Third Joint FAO/WHO/UNEP International Conference on Mycotoxins in Tunis, Tunisia. Food and Agricultural Organization of the United Nations, Rome, Italy.
12. Hald, B., Krogh, P. 1975. Detection of ochratoxin A in barley, using silica gel minicolumns. *J. Assoc. Off. Anal. Chem.* 58:156.
13. Huwig, A., Freimend, S., Kappeli, O., Dutler, H. 2001. Mycotoxin detoxication of animal feed by different absorbents. *Toxicol. Lett.* 122:179-188.

14. IARC. 1991. IARC monographs of the evaluation of carcinogenic risks to humans: ochratoxin A. p. 489-521. International Agency for Research on Cancer Scientific Publication No. 56, Lyon, France.
15. Iavicoli, I., Brera, C., Carelli, G., Caputi, R., Marinaccio, A., Miraglia, M. 2002. External and internal dose in subjects occupationally exposed to ochratoxin A. *Int. Arch. Occup. Environ. Health* 75:381-386.
16. Jiao, Y., Blaas, W., Ruhl, C., Weber, R. 1992. Identification of ochratoxin A in food samples by chemical derivatization and gas chromatography-mass spectrometry. *J. Chromatogr.* 595:364-367.
17. Jimenez, A. M., Lopex de Cerain, A., Gonzales-Penas, E., Bello, J. 2001. Determination of ochratoxin A in pig liver-derived pates by high-performance liquid chromatography. *Food Addit. Contam.* 18:559-63.
18. Jodlbauser, J., Maier, N. M., Lindner, W. 2002. Towards ochratoxin A selective molecularly imprinted polymers for solid-phase extraction. *J. Chromatogr. A* 945:45-63.
19. Lippmann, M. 1995. Filters and Filter Holders, p. 247-278. *In Air Sampling Instruments for Evaluation of Atmospheric Contaminants*, 8<sup>th</sup> ed. American Conference of Governmental Industrial Hygienists, Cincinnati, OH.
20. Olsson, J., Borjesson, T. Lundstedt, T., Schnurer, J. 2002. Detection and quantification of ochratoxin A and deoxynivalenol in barley grains by GC-MS and electronic nose. *Int. J. Food Microbiol.* 72:203-214.
21. Pasanen, A. L., Nikulin, M., Tuomainen, M., Berg, S., Parikka, P., Hintikka, E. L. 1993. Laboratory experiments on membrane filter sampling of airborne mycotoxins produced by *Stachybotrys atra Corda*. *Atmos. Environ.* 27A:9-13.
22. Richard, J. L., Plattner, R. D., May, J., Liska, S. L. 1999. .: The occurrence of ochratoxin A in dust collected from a problem household. *Mycopathologia* 146:99-103.
23. Robbins, C.A., Swenson, L. J., Neally, M. L., Gots, R. E., Kelman, B. J. 2000. Health effects of mycotoxins in indoor air: a critical review. *Appl. Occup. Environ. Hyg.* 15:773-84.
24. Scott, P.M. 1995. Mycotoxin methodology. *Food. Addit. Contam.* 12:395-403.
25. Shepard, G. S., Fabiani, A., Stockenstrom, S., Mshicileli, N., Sewram, V. 2003. Quantitation of ochratoxin A in South African wines. *J. Agric. Food Chem.* 51:1102-1106.
26. Skaug, M. A., Eduard, W., Stormer, F. C. 2000. Ochratoxin A in airborne dust and fungal conidia. *Mycopathologia* 151:93-98.
27. Skaug, M. A., Helland, I., Solvoll, K., Saugstad, O. A. 2001. Presence of ochratoxin A in human milk in relation to dietary intake. *Food Addit. Contam.* 18:321-327.
28. Takeda, Y., Isohata, E., Amano, R., Uchiyama, M. 1979. Simultaneous extraction and fractionation and thin layer chromatographic determination of 14 mycotoxins in grains. *J. Assoc. Off. Anal. Chem.* 62:573-578.
29. Tatu, C. A., Orem, W. H., Finkelman, R. B., Feder, G. L. 1998. The etiology of Balkan endemic nephropathy: still more questions than answers. *Environ. Health Perspect.* 106:689-700.
30. Todd, B. E., Buchan R. M. 2002. Total dust, respirable dust and microflora toxin concentrations in Colorado corn storage facilities. *Appl. Occup. Environ. Hyg.* 17:411-415.
31. Trucksess, M., Giler, J., Young, K., White, K. D., Page, S. W. 1999. Determination and survey of ochratoxin A in wheat, barley, and coffee—1997. *J. AOAC Int.* 82:85-89.

32. Walker, R. 2002. Risk assessment of ochratoxin: current views of the European Scientific Committee on Food, the JECFA and the Codex Committee on food additives and contaminants. *Adv. Exp. Med. Biol.* 504:249-255.
33. Walters, M., Milton, D., Larsson, L., Ford, T. 1994. Airborne environmental endotoxin: a cross-validation of sampling and analysis techniques. *App. Environ. Microbiol.* 60:996-1005.

