

## Abstracts

### Mycotoxins – General

GAREIS, M. and WOLFF, J. 2000. [Relevance of mycotoxin contaminated feed for farming animals and carry over of mycotoxins in food of animal origin]. *Mycoses* **43**: 79–83.

At the present time in Germany the only mycotoxins of practical relevance from the view of food hygiene and food safety are aflatoxins (aflatoxin M<sub>1</sub> in milk) and ochratoxin A (in blood, meat and edible tissues from swine). Other mycotoxins including trichothecenes, zearalenone and fumonisins, and ergot alkaloids are not important as possible food contaminants from animal origin although they could have a negative impact on animal production. (In German).

STRASSER, H., VEY, A. and BUTT, T.M. 2000. Are there any risks in using entomopathogenic fungi for pest control, with particular reference to the bioactive metabolites of *Metarhizium*, *Tolypocladium* and *Beauveria* species? *Biocontrol Science and Technology* **10**: 717–735.

Entomopathogenic fungi are promising alternatives to chemical insecticides. However, a major hurdle concerning the registration of these fungi as plant protection agents is the possible toxicity of secreted metabolites. This review with 139 references summarises data on specific secondary metabolites including destruxins, efrapeptins, oosporein, beauvericin and beauveriolides produced by the important genera *Beauveria*, *Metarhizium* and *Tolypocladium*. Methods and strategies are suggested which could standardise the risk assessment of fungal biological control agents.

KPODO, K., THRANE, U. and HALD, B. 2000. *Fusaria* and fumonisins in maize from Ghana and their co-occurrence with aflatoxins. *International Journal of Food Microbiology* **61**: 147–157.

Fifteen maize samples from four markets and processing sites in Accra, Ghana were analysed for fumonisins B<sub>1</sub>, B<sub>2</sub> and B<sub>3</sub>. All samples contained fumonisins. Total fumonisin levels for 14 samples ranged from 70 to 4222 mg/kg. Mycological examination of the samples showed *Aspergillus* as the dominant genus (76.4%) followed by *Penicillium* (19.9%). *Fusarium* formed 2.6%. Thirty-two *Fusarium* strains representing five species were isolated and tested for the production of fumonisins in maize substrates. From 21 of 22 *F. verticillioides* strains tested, all three types of fumonisins were produced. Additional studies on maize samples from 15 processing sites in Accra revealed a co-occurrence of both fumonisins and aflatoxins in 8/15 samples.

DASILVA, J.B., POZZI, C.R., MALLOZZI, M.A.B., ORTEGA, E.M. and CORREA, B. 2000. Mycoflora and occurrence of aflatoxin B<sub>1</sub> and fumonisin B<sub>2</sub> during storage of Brazilian sorghum. *Journal of Agricultural and Food Chemistry* **48**: 4352–4356.

In 140 samples of freshly harvested and stored sorghum in Brazil, the levels of aflatoxin and fumonisin contamination detected in the grains and the prevailing abiotic factors (grain moisture content, water activity, temperature, relative humidity and mean rainfall) at the time of sampling were investigated. The results show a predominance of the genera *Phoma* (57.1%), *Aspergillus* (42.7%), *Fusarium* (25.0%) and *Rhizopus* (21.4%). The species most frequently found were *Aspergillus flavus* and *Fusarium moniliforme*. Of the samples analysed, 12.8% were contaminated with aflatoxin B<sub>1</sub> (concentration mean 7–33 mg/kg) and 74.2% were contaminated with fumonisin

B<sub>1</sub> (concentration mean 0.11–0.15 mg/g). This paper is the first report of the natural occurrence of aflatoxins and fumonisins in sorghum grain from Brazil.

CANDLISH, A.A.G., AIDOO, K.E., SMITH, J.E. and PEARSON, S.M. 2000. A limited survey of aflatoxins and fumonisins in retail maize-based products in the UK using immunoassay detection. *Mycotoxin Research* **16**: 2–8.

A total of 27 maize based products destined for human consumption were collected from retail outlets in Glasgow, UK, and were analysed for the presence of aflatoxins using immunoaffinity column chromatography with fluorescence detection and for fumonisins by competitive ELISA. Aflatoxins were detected at trace levels (<4 mg/kg) in 8/27 samples and one sample of sweet corn contained aflatoxins at 5–10 mg/kg. Fumonisins were detected in 8/27 samples at levels from 1–8 mg/kg and a further 8 samples contained fumonisins at detectable levels (<1 mg/kg).

GUPTA, A.K., AHMAD, I., BORST, I. and SUMMERBELL, R.C. 2000. Detection of xanthomegnin in epidermal materials infected with *Trichophyton rubrum*. *Journal of Investigative Dermatology* **115**: 901–905.

Xanthomegnin is a mutagenic mycotoxin best known as an agent of nephropathy and death in farm animals. The production of xanthomegnin by the most common dermatophytic species, *Trichophyton rubrum*, both in dermatologic human nail specimens and in culture was investigated. In view of the labile nature of xanthomegnin, a chromatographic procedure was developed to allow HPLC analysis within 1 hr of sample extraction. Xanthomegnin was repeatedly extracted from human nail and skin material infected by *Trichophyton rubrum*. The level

of xanthomegnin present varied among the clinical samples studied but was not detected in uninfected nails.

MUBATANHEMA, W., MOSS, M.O., FRANK, M.J. and WILSON, D.M. 1999. **Prevalence of *Fusarium* species of the *Liseola* section on Zimbabwean corn and their ability to produce the mycotoxins zearalenone, moniliformin and fumonisin B<sub>1</sub>**. *Mycopathologia* **148**: 157–163.

Maize samples were collected from nine Grain Marketing Board (GMB) centres in Zimbabwe during the 1991 harvest season and further samples were collected directly from farmers and from GMB centres in Chinhoyi and Kwekwe during the 1992 harvest season and analysed mycologically. The predominant flora was *Fusarium*. The ability of these isolates to produce zearalenone (ZEA), moniliformin (MON) and fumonisin B<sub>1</sub> (FB<sub>1</sub>) was tested using a simplified TLC Agar plate method. Of 886 isolates tested, only one produced all three mycotoxins simultaneously whilst most produced FB<sub>1</sub> and/or MON. Only nine isolates produced ZEA.

FISCHER, G., MULLER, T., SCHWALBE, R., OSTROWSKI, R. and DOTT, W. 2000. **Exposure to airborne fungi, MVOC and mycotoxins in bio-waste-handling facilities**. *International Journal of Hygiene and Environmental Health* **203**: 97–104.

The fungal species composition in a compost facility was compared to the spectrum of microbial metabolites in the air with regard to the physiological properties of different fungal species. A number of relevant fungi was tested for the production of both volatile and non-volatile metabolites on different substrata. The profiles of mycotoxins and microbial volatile organic compounds (MVOC) were shown to be specific for certain species in pure culture.

FISCHER, G., MULLER, T., SCHWALBE, R., OSTROWSKI, R. and DOTT, W. 2000. **Species-specific profiles of mycotoxins produced in cultures and associated with conidia of airborne fungi derived from biowaste**. *International Journal of Hygiene and Environmental Health* **203**: 105–116.

The potential to produce mycotoxins and non-volatile secondary metabolites was investigated for approximately 250 freshly isolated fungal strains. Among the eleven most relevant species, *Aspergillus flavus*, *A. fumigatus*, *A. niger*, *A. parasiticus*, *A. versicolor*, *Emericella nidulans*, *Paecilomyces variotii*, *Penicillium brevicompactum*, *P. clavigerum*, *P. crustosum* and *P. polonicum*, a wide range of metabolites partly of toxico-

logical relevance was identified. The spectra of metabolites in conidial extracts and culture extracts (containing also mycelium and medium) were compared for a limited number of relevant fungi. Some mycotoxins, such as sterigmatocystin in *Emericella nidulans*, were not present in the conidial extracts, though produced by most strains. Fumigaclavine C, tryptoquivaline and trypacidin, characteristic for *A. fumigatus*, were found in conidial extracts, but highly toxic compounds such as gliotoxin and fumitremorgens were not present. Compounds such as cyclopenol, cyclopenin and penitrem A were found in conidial extracts and are therefore assumed to occur in native bio-aerosols.

SAKUDA, S., ONO, M., IKEDA, H., NAKAMURA, T., INAGAKI, Y., KAWACHI, R., NAKAYAMA, J., SUZUKI, A., ISOGAI, A. and NAGASAWA, H. 2000. **Blasticidin A as an inhibitor of aflatoxin production by *Aspergillus parasiticus***. *Journal of Antibiotics* **53**: 1265–1271.

Blasticidin A at a concentration of 0.5 mM almost completely inhibited aflatoxin production by *Aspergillus parasiticus*. The structure of blasticidin A was characterised by NMR and chemical degradation experiments.

REDDY, M.V.B., ANGERS, P., CASTAIGNE, F. and ARUL, J. 2000. **Chitosan effects on black mold rot and pathogenic factors produced by *Alternaria alternata* in postharvest tomatoes**. *Journal of the American Society for Horticultural Science* **125**: 742–747.

Stem scar application of chitosan inhibited growth of *Alternaria alternata* in challenged tomato (*Lycopersicon esculentum* Mill.) fruit stored at 20°C for 28 days. Lesions were visible within 4 days of inoculation in control fruit compared with more than 7 days in chitosan treated fruit. Chitosan also inhibited production of host specific toxins such as alternariol and alternariol monomethylether by the fungus. Such chitosan-pathogen-host interactions may be exploited in the control of postharvest pathogens of fresh fruit and vegetables.

TAG, A., HICKS, J., GARIFULLINA, G., AKE, C., PHILLIPS, T.D., BEREMAND, M. and KELLER, N. 2000. **G-protein signalling mediates differential production of toxic secondary metabolites**. *Molecular Microbiology* **38**: 658–665.

Previous work revealed that synthesis of sterigmatocystin (STG) and aflatoxin in *Aspergillus* species is negatively controlled by FadA, the alpha-subunit of a heterotrimeric G-protein. In contrast, it is shown here

that the dominant activating *fadA* allele, *fadA*(G42R), stimulates transcription of a gene from the *A. nidulans* penicillin gene cluster and elevates penicillin production. Thus, FadA has opposite roles in regulating the biosynthesis of penicillin and STG in *A. nidulans*. Furthermore, expression of *fadA*(G42R) in *Fusarium sporotrichioides* increases trichothecene production and alters trichothecene gene expression. These data provide evidence that targeting G-protein signal transduction pathways as a means of controlling or preventing the production of a single mycotoxin could have serious undesirable consequences with regard to the production of other secondary metabolites.

LAIB, T. and ZHU, J.P. 2000. **Synthesis of model of phomopsin-ustiloxin-type antimetabolic agents**. *Synlett* (9): 1363–1365.

An intramolecular SNAr reaction was applied to the synthesis of a 13-membered meta-cyclophane that is the structural core of phomopsin and ustiloxin type antimetabolites.

BURKE, L.T., DIXON, D.J., LEY, S.V. and RODRIGUEZ, F. 2000. **A short stereoselective total synthesis of the *Fusarium* toxin equisetin**. *Organic Letters* **2**: 3611–3613.

A short stereoselective synthesis of equisetin, an N-methylserine-derived acyl tetramic acid and potent inhibitor of HIV-1 integrase enzyme, is described.

WILLIAMS, D.R. and TURSKE, R.A. 2000. **Construction of 4-hydroxy-2-pyridinones. Total synthesis of (+)-sambutoxin**. *Organic Letters* **2**: 3217–3220.

The synthesis of (+)-sambutoxin has been achieved, establishing the relative and absolute stereochemistry of this mycotoxin.

## Mycotoxins – Methodology

LEWIS, C.W., SMITH, J.E., ANDERSON, J.G. and FRESHNEY, R.I. 1999. **Increased cytotoxicity of food-borne mycotoxins toward human cell lines *in vitro* via enhanced cytochrome P450 expression using the MTT bioassay**. *Mycopathologia* **148**: 97–102.

Eight foodborne mycotoxins were tested for their cytotoxic effects on human cells previously immortalised and transfected to introduce human cytochrome P450 (CYP450) genes. Such cells retain many characteristics of normal cell growth and differentiation while simultaneously having the potential of either increasing or decreasing the cytotoxicity of the challenging mycotoxins. The MTT assay was used to

indicate cytotoxicity. Of the nine CYP450s introduced, CYP1A2 was most effective, rendering the cells 540 times more sensitive to AFB<sub>1</sub> than the control cells, 28 times more sensitive to AFG<sub>1</sub> and 8 times more sensitive to ochratoxin A. CYP3A4 resulted in the cells being 211 times more toxic to AFB<sub>1</sub> and 8 times more toxic to AFG<sub>1</sub> while CYP 2A6, CYP 3A5 and CYP 2E1 also produced observable effects. No increase in metabolic activity was found using cyclopi-azonic acid, deoxynivalenol, FB<sub>1</sub>, patulin or T-2 toxin. *In vitro* biological assays thus provide an excellent system for quantifying the often low CD<sub>50</sub>s expressed by mycotoxins in foods.

GEISEN, R. 2000. [Molecular methods to ensure the microbiological quality of vegetable food]. *Ernährungs-Umschau* 47: 303.

A review with 11 references. The polymerase chain reaction (PCR) is a rapid molecular technique which can be used for the detection of microorganisms in food. Several diagnostic PCR systems based on genes of the biosynthetic pathways of aflatoxins and trichothecenes as target sequences have been described. (In German).

VIDEMANN, B., BONY, S. and BERNY, P. 2000. **Determination of ergovaline in endophyted seeds by high performance thin layer chromatography (HPTLC)**. *Journal of Liquid Chromatography & Related Technologies* 23: 2727–2738.

An HPTLC method for the rapid determination of ergovalin in seeds of *Graminaceae* infested with *Neotyphodium coenophialum* is described. Quinine is used as an internal standard. The mycotoxin is solubilised under acid conditions and extracted with chloroform under alkaline conditions. Samples are sprayed on silica gel plates and detected by fluorescence. The method developed is specific for endophyte infested seeds, linear from 3 to 12 mg/kg and is both repeatable and reproducible. The limit of detection is 0.5 mg/kg and the limit of determination is 3.0 mg/kg.

## Mycotoxins – Toxicology

ANTTILA, K. 2000. **Mycotoxins, fungus and ‘electrohypersensitivity’**. *Medical Hypotheses* 55: 208–214.

Many people displaying symptoms caused by electromagnetic fields have fungus infections or have been living in fungus contaminated environments for long periods. In animal studies mycotoxins have shown the same effects as those seen in the ‘electrohypersensitivity’ syndrome. Photo-

toxic reactions are well known in veterinary medicine and in medical science which raises the question of whether the ‘electrohypersensitivity’ syndrome is caused by ‘phototoxic’ reactions.

LIU, H.G., JACKMAN, S., DRISCOLL, H. and LARSEN, B. 2000. **Immunologic effects of gliotoxin in rats. Mechanisms for prevention of autoimmune diabetes mellitus**. *Annals of Clinical and Laboratory Science* 30: 366–378.

The immunologic effects of gliotoxin treatment of rats was studied. Splenocytes from 65-day-old prediabetic diabetes prone rats were phenotypically characterised after chronic treatment with gliotoxin. A parallel study examined the direct effects of gliotoxin on splenocyte preparations incubated with the mycotoxin. *In vitro* treatment of splenocytes with gliotoxin revealed relative decreases in CD4(+) and increases in CD8(+) T-cell subsets, whereas *in vivo* treatment with gliotoxin did not result in detectable alterations in relative CD4+ and CD8+ cell subsets. However, *in vitro* and *in vivo* gliotoxin treatments significantly enhanced the detectable RT6 surface marker, a key regulatory element in autoimmune diabetes pathogenesis.

MULLER, G., KIELSTEIN, P., ROSNER, H., KOHLER, H., BERNDT, A. and HELLER, M. 2000. **[Do mycotoxins impair immune and defence reactions in pigs?]** *Praktische Tierarzt* 81: 932.

The immunomodulating effects of the relatively low concentrations of mycotoxins which are found in feeds in Germany, were investigated in pigs. Experimental applications of ochratoxin A (OA) at 7–50 mg/kg body weight alone or together with deoxynivalenol at 25 mg/kg, T-2 toxin at 15 mg/kg and fumonisin at 12–15 mg/kg, induces clear immunomodulatory changes. OA doses between 20 and 50 mg/kg alone can cause an increase in the severity of artificially induced pneumonia and additional clinical symptoms. (In German).

BEVER, R.J., COUCH, L.H., SUTHERLAND, J.B., WILLIAMS, A.J., BEGER, R.D., CHURCHWELL, M.I., DOERGE, D.R. and HOWARD, P.C. 2000. **DNA adduct formation by *Fusarium* culture extracts: Lack of role of fusarin C**. *Chemico-Biological Interactions* 128: 141–157.

The incubation of methanol extracts of *Fusarium verticillioides* cultures with DNA in the presence of rat liver fractions (S9) resulted in the formation of a unique DNA adduct that was detected by [<sup>32</sup>P]-postlabelling. Fusarin C, purified from cultures of *F. verticillioides*, was not responsible for the formation of the DNA adduct. The unique

DNA adduct was formed following the incubation of several *F. verticillioides* isolates from the USA and South Africa, while extracts of cultures of *F. graminearum* and *F. sacchari* isolates formed very little of the DNA adduct when incubated with DNA and S9. These data suggest that neither fusarin C nor any of its metabolites is responsible for formation of the DNA adduct and that an unidentified compound is present in *F. verticillioides* cultures that forms a DNA adduct, and may be important in the aetiology of human oesophageal cancer.

HASAN, H.A.H. 1999. **Phytotoxicity of pathogenic fungi and their mycotoxins to cereal seedling viability**. *Mycopathologia* 148: 149–155.

The ability of AFB<sub>1</sub> and G<sub>1</sub>, diacetoxyscirpenol (DAS), kojic acid and tenuazonic acid to reduce cereal seedling viability was studied. The LD<sub>50</sub>s for barley, wheat and sorghum treated with aflatoxins were 0.83, 1.74 and 2.75 mg/L, respectively, for DAS 1.26, 3.98 and 10 mg/L, respectively, and for kojic acid 63, 105 and 251 mg/L, respectively. Tenuazonic acid was less toxic and toxicity ranged between 79–550 mg/L. The germination inhibition was more pronounced in barley followed by wheat and was negligible in sorghum for all tested mycotoxins. This inhibition was attributed to reduction in seedling amylase activity, as amylase was reduced in the same trend: barley > wheat > sorghum.

## Patulin

LEGGOTT, N.L. and SHEPHARD, G.S. 2001. **Patulin in South African commercial apple products**. *Food Control* 12: 73–76.

Locally produced commercial apple products purchased from retail outlets in South Africa were surveyed for the presence of patulin from 1996 to 1998. Twenty-three of the 31 fruit juice samples had no detectable patulin contamination. The eight contaminated juice samples had patulin concentrations in the range 5–45 mg/L with a mean of 10 mg/L. Among whole fruit products, 2/6 were contaminated with patulin at 10 mg/kg. Among infant fruit juices, 6/10 samples had patulin concentrations in the range 5–20 mg/L. The infant fruit purees showed no detectable patulin contamination.

GOKMEN, V. and ACAR, J. 2000. **Long-term survey of patulin in apple juice concentrates produced in Turkey**. *Food Additives and Contaminants* 17: 933–936.

A LC method for the determination of patulin was evaluated for a long term survey of patulin in apple juice concentrates. Patulin was separated on a reversed phase C<sub>18</sub> LC column with water-acetonitrile (99:1) as the mobile phase and quantified with a photodiode array (PDA) detector. Relatively low amounts of patulin (less than 5 mg/L for single strength juice at 11.2°Brix) were detected in apple juice concentrates and confirmed by PDA detector. In Turkey, 482 apple juice concentrates produced through 1996–99 were analysed for their patulin contents. Year-to-year variations in patulin levels were statistically significant. Contamination levels tended to decrease through the years 1996–99 and averaged 63, 43, 19 and 31 mg/L, respectively. Percentages of concentrates exceeding the maximum permitted concentration of 50 mg/L were 52, 34, 8 and 8%, respectively.

TSAO, R. and ZHOU, T. 2000. **Micellar electrokinetic capillary electrophoresis for rapid analysis of patulin in apple cider.** Journal of Agricultural and Food Chemistry **48**: 5231–5235.

A micellar electrokinetic capillary chromatography mode was applied to a capillary electrophoresis method, which was developed for detection and quantitation of patulin in apple ciders. This method used a small sample amount and consumed minimal organic solvent compared to the most commonly used HPLC methods. The sample preparation procedure of the CE method was also simpler than other chromatographic techniques developed for patulin analysis. Patulin was detected with a PDA detector at 273 nm. The limit of detection of the method was 3.8 mg/L. Patulin recoveries from 4 spiked samples ranged from 95.2 to 105.4%. Recoveries were around 98%.

MACDONALD, S., LONG, M., GILBERT, J. and FELGUEIRAS, I. 2000. **Liquid chromatographic method for determination of patulin in clear and cloudy apple juices and apple puree: Collaborative study.** Journal of AOAC International **83**: 1387–1394.

A collaborative trial was conducted to validate the effectiveness of a LC procedure for determination of patulin in both clear and cloudy apple juices and apple puree. The test portion of clear apple juice was directly extracted with ethyl acetate. Cloudy apple juice and apple puree were treated with pectinase enzyme before extraction. After back-extraction into sodium carbonate to remove interfering acidic compounds, the extract was dried and concentrated, and patulin was determined by LC with UV detection. For the three sample types spiked with patulin at 75 mg/kg, recoveries ranged from 80 to 92%. The relative standard deviations

for repeatability (RSD<sub>r</sub>) and reproducibility (RSD<sub>R</sub>) ranged from 8 to 35% and 11 to 36%, respectively.

SEWRAM, V., NAIR, J.J., NIEUWOUDT, T.W., LEGGOTT, N.L. and SHEPHARD, G.S. 2000. **Determination of patulin in apple juice by high-performance liquid chromatography-atmospheric pressure chemical ionization mass spectrometry.** Journal of Chromatography A **897**: 365–374.

An HPLC-MS-MS method with selected reaction monitoring for the determination of patulin in apple juice samples is described. MS detection was accomplished following atmospheric pressure chemical ionisation in both positive and negative ion modes. Detection in the negative ion mode proved superior and a linear response was observed over the injected range from 6 to 200 ng patulin. Apple juice samples spiked with patulin between 10 and 135 mg/L were analysed following liquid-liquid extraction with ethyl acetate and cleanup with sodium carbonate. Utilising reversed phase HPLC with acetonitrile-water (10:90), levels down to 10 mg/L were readily quantified and a detection limit of 4 mg/L was attainable. The MS data for the spiked samples compared well to the UV data.

### Citrinin

ZHELIFONOVA, V.P., VINOKUROVA, N.G. and OZER-SKAYA, S.M. 2000. **Effect of microelements on the biosynthesis of secondary metabolites by the fungus *Penicillium citrinum* Thom VKM F-1079.** Microbiology **69**: 536–540.

*Penicillium citrinum* VKM-1079 was found to produce clavine ergot alkaloids and citrinin. Citrinin was produced in the idiophase, whereas the production of ergot alkaloids paralleled fungal growth. The addition of manganese ions to the growth medium stimulated the biosynthesis of both citrinin and ergot alkaloids. Zinc ions stimulated only citrinin synthesis. The presence of these microelements in the growth medium influenced the proportion between the ergot alkaloids synthesised.

WILD, D. 2000. **[Red mould rice (angkak). Analysis and detection in meat products].** Fleischwirtschaft **80**: 91–93.

An HPLC technique was developed for the analysis of red mould rice. Laboratory cultures of *Monascus purpureus* DSM1379 on a rice medium contained citrinin at 600–800 mg/kg. Commercial samples of red mould rice contained lower amounts of citrinin, less than 50 mg/kg, and they differed

considerably with regard to colouring and other constituents. A simple assay for the monitoring of red mould rice in meat products is described. Preliminary data suggest that red mould rice is only sporadically used in the production of meat products but more frequently in vegetarian sausage-type products. Because of the potential formation of citrinin, high variability of commercial samples and insufficient data for these complex mixtures the safety of red mould rice as a food additive is considered problematic. (In German).

HAJJAJ, H., BLANC, P., GROUSSAC, E., URIBELARREA, J.L., GOMA, G. and LOUBIERE, P. 2000. **Kinetic analysis of red pigment and citrinin production by *Monascus ruber* as a function of organic acid accumulation.** Enzyme and Microbial Technology **27**: 619–625.

*Monascus ruber* was grown in submerged cultures and assayed for red pigment and citrinin. In oxygen limiting conditions, the production of the two metabolites was growth associated, as was the production of primary metabolites. In oxygen excess conditions, the profile of citrinin production was typical of a secondary metabolite, since it was produced mostly during the stationary phase. In contrast, the production of the pigment decreased rapidly throughout the culture, showing a profile characteristic of an inhibitory mechanism. The organic acids produced during the culture, L-malate and succinate, were slightly inhibitory against pigment production, while citrinin production was unaffected.

### Tremorgens

ZHAO, Y., KU, Y.L., HAO, X.J. and LEE, S.S. 2000. **Preparation of analogues of territrein B, a potent AChE inhibitor.** Tetrahedron **56**: 8901–8913.

The synthesis of analogues of the acetylcholinesterase inhibitor, territrein B, was carried out starting from the naturally occurring jujubogenin glycosides. The anti-acetylcholinesterase activity of the analogues was measured. The aromatic ring moiety seemed to be less important when compared with the 2-en-1-one pharmacophore.

SMITH, A.B., KANO, N., ISHIYAMA, H. and HARTZ, R.A. 2000. **Total synthesis of (–)-penitrein D.** Journal of the American Chemical Society **122**: 11254–11255.

The first total synthesis of (–)-penitrein D is described. Key elements of the synthesis include the stereocontrolled elaboration of the advanced eastern hemisphere, a highly efficient union of the hemispheres

exploiting a 2-substituted indole synthesis and the efficient construction of rings A and F via a Sc(OTf)<sub>3</sub> promoted cation cascade.

ZEHNDER, L.R., HSUNG, R.P., WANG, J.S. and GOLDING, G.M. 2000. **A concise stereoselective route to the pentacyclic frameworks of arisugacin A and territrein B.** *Angewandte Chemie – International Edition* **39**: 3876.

This paper describes a concise route to the pentacyclic frameworks of arisugacin A and territrein B using a stereoselective formal cycloaddition reaction.

LONGLAND, C.L., DYER, J.L. and MICHELANGELI, F. 2000. **The mycotoxin paxilline inhibits the cerebellar inositol 1,4,5-trisphosphate receptor.** *European Journal of Pharmacology* **408**: 219–225.

Paxilline is a reversible inhibitor of the cerebellar inositol 1,4,5-trisphosphate (InsP(3)) receptor. It inhibits the amount or extent of InsP(3) induced Ca<sup>2+</sup> release (ICR), at sub-maximal concentrations of InsP(3), in a biphasic manner consistent with two inhibition constants. The rate constants for Ca<sup>2+</sup> release from both the fast and slow populations are reduced by paxilline at 100 mM by about 70 and 60%, respectively.

## Ochratoxins – General

STUDER-ROHR, I., SCHLATTER, J. and DIETRICH, D.R. 2000. **Kinetic parameters and intraindividual fluctuations of ochratoxin A plasma levels in humans.** *Archives of Toxicology* **74**: 499–510.

The toxicokinetic profile of OA was studied in one human volunteer following ingestion of 395 ng [<sup>3</sup>H]-labelled OA. A two compartment open model best described the *in vivo* data and consisted of a fast elimination and distribution phase (T-1/2 about 20 hr) followed by a slow elimination phase (renal clearance about 0.11 mL/min) and a calculated plasma half-life of 35.55 days. This half-life was approximately eight times longer than that determined previously in rats. In addition, the intra-individual fluctuation of OA plasma levels was investigated in eight individuals over a period of 2 months. The plasma levels in some individuals remained nearly constant over time, while others varied considerably.

WOLFF, J., BRESCH, H., CHOLMAKOV-BODECHTEL, C., ENGEL, G., GAREIS, M., MAJERUS, P., ROSNER, H. and SCHEUER, R. 2000. **Ochratoxin**

**A: Contamination of foods and consumer exposure. General introduction.** *Archiv für Lebensmittelhygiene* **51**: 84.

The German Federal Ministry for Health has initiated a study aimed to determine the exposure of the consumer to and the contamination of foods with OA. Within a period of 2.5 years, almost 7000 food samples and blood samples from more than 1000 test persons were examined for OA. In the same period, the nutritional and consumption habits of more than 2500 persons were recorded by trained interviewers using questionnaires elaborated by nutritional scientists. With the help of these data, OA exposure of representative consumer collectives such as vegetarians, infants and adolescents as well as low level, normal and high level consumers could be established and compared with OA concentrations of the blood sera analysed.

WOLFF, J. 2000. **Ochratoxin A in cereals and cereal products.** *Archiv für Lebensmittelhygiene* **51**: 85–88.

OA was determined in cereals and in a large variety of cereal products including flour, bread, bread rolls, pasta and cereal based foods. A total of 2374 samples were analysed and 68.6% were found to contain OA. However, only 1.4% of samples contained OA at levels exceeding 3 mg/kg.

BRESCH, H., URBANEK, M. and HELL, K. 2000. **Ochratoxin A in coffee, tea and beer.** *Archiv für Lebensmittelhygiene* **51**: 89–94.

A total of 357 samples of coffee, 161 samples of tea and 318 samples of beer were assayed for OA. About 50% of the samples of roasted and ground coffee, with and without caffeine, contained OA at concentrations of 0.3–6.3 mg/kg. Of instant coffee samples with caffeine, about 90% contained OA in the range 0.3–9.47 mg/kg. A lower level was found in decaffeinated instant coffee. No toxin was found in black or fruit tea, but OA was found in 3% of green tea, 9% of herb tea and 42% of childrens tea samples on a herb basis. Of Pils, Export, Weizen and Starkbeer samples analysed, 70–80% contained OA at concentrations of 0.01–0.29 mg/L.

MAJERUS, P., BRESCH, H. and OTTENEDER, H. 2000. **Ochratoxin A in wines, fruit juices and seasonings.** *Archiv für Lebensmittelhygiene* **51**: 95–97.

A total of 281 wine samples from 22 countries were analysed for OA. Forty percent of samples were contaminated with OA with a 90th percentile value of 0.4 mg/kg. The contamination rates of rosé and red wines and products originating from warmer regions were considerably higher. Fifty per-

cent of wine vinegar samples contained OA with a 90th percentile value of 0.22 mg/kg. Of 91 grape juice samples analysed, 85% contained OA and the 90th percentile value was 2.32 mg/kg.

ENGEL, G. 2000. **Ochratoxin A in sweets, oil seeds and dairy products.** *Archiv für Lebensmittelhygiene* **51**: 98–101.

Among 1933 samples of dairy products, sweets (including cocoa, chocolate, snacks and crisps), nuts, dried fruits, puddings and sandwich spreads, and oil and oil seeds, OA was detected in 59% of samples. All of the analysed cocoa samples contained OA with cocoa powder having a 90th percentile value of 0.934 mg/kg. Among raisins and currants samples, 95% contained OA with a 90th percentile value of 3.33 mg/kg. Individual high concentrations of OA were found in muesli, biscuits, nut bars and dried figs.

GAREIS, M. and SCHEUER, R. 2000. **Ochratoxin A in meat and meat products.** *Archiv für Lebensmittelhygiene* **51**: 102–104.

OA was assayed in 620 samples of beef, pork and poultry meat, pig liver and kidney, as well as a range of meat products on the German market. No OA was found in poultry and only one sample of beef contained trace amounts. The incidences of OA in pork meat, kidneys and liver were 17, 44 and 17%, respectively, however the median values were less than 0.01 mg/kg. Among meat products, blood sausages and liver sausages were the most frequently contaminated and were 77 and 68%, respectively, with maximum OA values of 3.2 and 4.6 mg/kg, respectively.

ROSNER, H., ROHRMANN, B. and PEIKER, G. 2000. **Ochratoxin A in human serum.** *Archiv für Lebensmittelhygiene* **51**: 104–107.

The OA exposure of the German population was determined by examining 927 blood serum specimens collected from representative groups. Among the 98.1% positive results, 65.5% contained serum OA in the range 0.11–0.20 mg/L and 22.1% contained serum OA in the range 0.21–0.5 mg/L.

GAREIS, M., ROSNER, H. and EHRHARDT, S. 2000. **Blood serum levels of ochratoxin A and nutrition habits.** *Archiv für Lebensmittelhygiene* **51**: 108–110.

A statistical evaluation was carried out of the OA analyses of 785 serum samples from healthy blood donors from different parts of Germany and their data on nutritional habits. Analyses showed a significant effect due to location in Germany but no effect due to sex, age, weight, body-mass

index and vegetarian diet. The statistical comparison of the persons with low, medium and high OA serum levels showed significant effects due to the consumption of bread, rye bread, muesli, blood sausages, salami type sausages, chocolate with nuts and coffee. However, OA levels in blood were not the result of a single food item or food group alone.

CHOLMAKOV-BODECHTEL, C., WOLFF, J., GAREIS, M., BRESCH, H., ENGEL, G., MAJERUS, P., ROSNER, H. and SCHNEIDER, R. 2000. **Ochratoxin A: Representative food consumption survey and epidemiological analysis.** *Archiv für Lebensmittelhygiene* **51**: 111–115.

The intake of OA with food was studied by means of representative OA analyses of food and an epidemiological food consumption survey. Included in the survey were 2005 adults and 574 children. The total daily intake of OA by adults and children was calculated to be 39.9 and 27.9 ng, respectively. The relative intake by adults and children was 0.58 and 0.97 ng/kg body weight, respectively. The highest intake was found for children of age 4–6 years and was 1.3 ng/kg body weight. Because of their high consumption frequency, cereal products are the main contributors (40–50%) of OA for all groups. Furthermore, coffee and beer are an important source of OA for adults, while red grape juice and sweets are important sources for children.

WOLFF, J., BRESCH, H., CHOLMAKOV-BODECHTEL, C., ENGEL, G., GAREIS, M., MAJERUS, P., ROSNER, H. and SCHEUER, R. 2000. **Ochratoxin A: Contamination of foods and consumer exposure final evaluation.** *Archiv für Lebensmittelhygiene* **51**: 115–117.

This paper gives a final evaluation of the results reported for the chemical-analytical examinations of foods and human sera for OA in Germany. These results give a comprehensive overview of the exposure to OA of the German consumer from foods available on the market.

LEONI, L.A.B., SOARES, L.M.V. and OLIVEIRA, P.L.C. 2000. **Ochratoxin A in Brazilian roasted and instant coffees.** *Food Additives and Contaminants* **17**: 867–870.

Samples of roast and ground coffee, instant coffee and decaffeinated instant coffee were collected in markets and supermarkets in the city of Campinas, Brazil, and analysed for OA. Twenty-three of 34 samples of ground and roast coffee were contaminated with OA at levels ranging between 0.3 and 6.5 mg/kg. All 14 samples

of instant coffee contained OA at levels ranging from 0.5 to 5.1 mg/kg. To study the transfer of OA into coffee brew, the beverage was prepared by one of two methods: the drip method and the Brazilian country style method. No significant difference was observed between the two methods in terms of extraction of OA. The drip method extracted 86 ± 15 % and the Brazilian country style 74 ± 20 % of the OA initially present in the coffee.

LEE, H.B. and MAGAN, N. 2000. **Impact of environment and interspecific interactions between spoilage fungi and *Aspergillus ochraceus* on growth and ochratoxin production in maize grain.** *International Journal of Food Microbiology* **61**: 11–16.

Using layers of irradiated but still fertile maize grain, the effects of water activity ( $a_w$ ) and temperature on interspecific interactions between *Aspergillus ochraceus* and five other spoilage fungi were examined. At 18°C, ochratoxin production by *A. ochraceus* was inhibited significantly by *A. candidus* (0.995 and 0.95  $a_w$ ) and *A. niger* (0.995  $a_w$ ). When grown on maize grain at 30°C, ochratoxin production by *A. ochraceus* was significantly inhibited by other spoilage fungi when both were grown on maize grain, especially by *A. niger* and *Eurotium amstelodami* at 0.995  $a_w$  and *A. flavus* at 0.95  $a_w$ .

STANDER, M.A., BORNSCHEUER, U.T., HENKE, E. and STEYN, P.S. 2000. **Screening of commercial hydrolases for the degradation of ochratoxin A.** *Journal of Agricultural and Food Chemistry* **48**: 5736–5739.

A number of commercial hydrolases were screened for the ability to degrade OA to nontoxic compounds. A crude lipase from *Aspergillus niger* (Amano A) proved to substantially hydrolyse OA to the nontoxic alphaO and phenylalanine, as confirmed by HPLC with fluorescence detection. The enzyme was purified by anion exchange chromatography to homogeneity. Activity staining of the purified enzyme with *n*-naphthyl acetate/Fast Red revealed only one band exhibiting hydrolytic activity. The specific activity of the purified enzyme toward OA was 2.32 units/mg.

## Ochratoxins – Methodology

ENTWISLE, A.C., WILLIAMS, A.C., MANN, P.J., SLACK, P.T. and GILBERT, J. 2000. **Liquid chromatographic method with immunoaffinity column cleanup for determination of ochratoxin A in barley: Collaborative**

**study.** *Journal of AOAC International* **83**: 1377–1383.

A collaborative study was conducted to evaluate a LC method with immunoaffinity column cleanup for determination of OA. The test portion was extracted with acetonitrile-water by blending at high speed. The extract was filtered, diluted with phosphate-buffered saline and applied to an OA immunoaffinity column. The column was washed with water and the OA eluted with methanol. The solvent was then evaporated and the residue redissolved in injection solvent. After injection of this solution onto reversed phase LC column, OA was measured by fluorescence detection. Test portions were spiked with OA at 4 mg/kg and recoveries ranged from 65 to 113%. The relative standard deviation for repeatability (RSDr) ranged from 4 to 24%, and the relative standard deviation for reproducibility (RSDR) ranged from 12 to 33%.

THIRUMALA-DEVI, K., MAYO, M.A., REDDY, G., REDDY, S.V., DELFOSSE, P. and REDDY, D.V.R. 2000. **Production of polyclonal antibodies against ochratoxin A and its detection in chillies by ELISA.** *Journal of Agricultural and Food Chemistry* **48**: 5079–5082.

Polyclonal antibodies were produced for OA by injecting OA-bovine serum albumin conjugate sc into a New Zealand White inbred rabbit. Antiserum could be used at a dilution exceeding 1:100 000 in an indirect competitive ELISA, and detected OA concentrations up to 0.1 mg/L. Antibodies did not react with OB, coumarin, 4-hydroxycoumarin, L-phenylalanine or AFB<sub>1</sub>. OA contamination in chillies (*Capsicum annum* L.) collected from commercial markets and cold storage units was determined. Of 100 chilli samples tested, 26 were found to contain over 10 mg/kg of OA. This is the first record in India of OA in chillies, a major component of cooked foods in this country, and it is noteworthy that OA contamination exceeded the permissible limit for human consumption (< 20 mg/kg) in over 26% of the market samples tested.

## Ochratoxins – Toxicology

HONG, J.T., PARK, K.L., HAN, S.Y., PARK, K.S., KIM, H.S., OH, S.D., LEE, R.D. and JANG, S.J. 2000. **Effects of ochratoxin A on cytotoxicity and cell differentiation in cultured rat embryonic cells.** *Journal of Toxicology and Environmental Health – Part A* **61**: 609–621.

In the rat embryo the effects of OA on cytotoxicity, cell differentiation and other cell functions in the embryonic midbrain cells which are dopaminergic, were com-

pared to those in the limb bud cells which are nondopaminergic, to assess the selectivity of OA central action. OA significantly reduced the levels of protein, DNA and glutathione, and [<sup>3</sup>H] thymidine incorporation into DNA in both embryonic midbrain and limb bud cells in a similar concentration dependent manner. The IC<sub>50</sub> values for cytotoxicity in midbrain cells and limb bud cells were 1.10 and 1.05 mM, respectively, and for cell differentiation were 1.10 and 1.0 mM, respectively.

ZANIC-GRUBISIC, T., ZRINSKI, R., CEPELAK, I., PETRIK, J., RADIC, B. and PEPELJNJAK, S. 2000. **Studies of ochratoxin A-induced inhibition of phenylalanine hydroxylase and its reversal by phenylalanine.** *Toxicology and Applied Pharmacology* **167**: 132–139.

The effects of low doses of OA on the activity of phenylalanine hydroxylase in kidney and in liver was studied in rats. Daily administration of OA at 50 mg/kg body weight for 10 and 35 days caused a significant reduction in the phenylalanine hydroxylase activity. Inhibition was more pronounced in liver than in kidney, although actual OA concentration was higher in the kidney tissue. Simultaneous application of OA with phenylalanine could reduce inhibition of phenylalanine hydroxylase, in particular in liver. Enzyme activity was almost completely preserved after 35 days of combined treatment.

SUJANI, G. and SRIRAMAN, P.K. 2000. **Pathology of ochratoxicosis in quails.** *Indian Journal of Animal Sciences* **70**: 1120–1122.

Four-week-old quails were fed OA at 2 or 4 mg/kg for 2 months. Total leukocyte count, packed cell volume and haemoglobin values were significantly reduced in OA treated groups. Significant and consistent lesions were noticed in kidney and liver indicating the nephrotoxic and hepatotoxic potentiality of OA. The toxin also had a potential to cause anaemia and deleterious effects on the lymphoid organs.

## Fumonisin – General

MACHINSKI, M. and SOARES, L.M.V. 2000. **Fumonisin B<sub>1</sub> and B<sub>2</sub> in Brazilian corn-based food products.** *Additives and Contaminants* **17**: 875–879.

Corn products were acquired from markets and supermarkets in the city of Campinas, SP, Brazil, and were analysed for FB<sub>1</sub> and FB<sub>2</sub>. Among 81 samples, 40 were positive for FB<sub>1</sub> (0.03–4.93 mg/kg) and 44 were positive for FB<sub>2</sub> (0.02–1.38 mg/kg). The samples, in order of decreasing contamina-

tion were corn meal followed by degerminated corn, corn flour, precooked corn flour, corn grits and popcorn. Relatively lower incidences and levels of contamination were found in corn flakes and corn flour baby cereal. The samples of corn on the cob and of the typical foods curau and pamonha, both prepared with corn in the milky stage, did not show any detectable contamination. Canned sweet corn, also harvested in the milky stage, exhibited a very low incidence of and level of contamination. This is the first report on fumonisins in Brazilian corn based food products.

JINDAL, N., MAHIPAL, S.K. and ROTTINGHAUS, G.E. 1999. **Occurrence of fumonisin B<sub>1</sub> in maize and poultry feeds in Haryana, India.** *Mycopathologia* **148**: 37–40.

Samples of maize and poultry feed collected from poultry farms, feed manufacturers and markets in Haryana in 1998 were analysed for FB<sub>1</sub>. Ninety-one percent of maize samples and 84% of poultry feed samples contained FB<sub>1</sub> in the ranges 0.1–87.0 and 0.02–28.0 mg/kg, respectively.

DESJARDINS, A.E. and PLATTNER, R.D. 2000. **Fumonisin B<sub>1</sub>-nonproducing strains of *Fusarium verticillioides* cause maize (*Zea mays*) ear infection and ear rot.** *Journal of Agricultural and Food Chemistry* **48**: 5773–5780.

The role of various fumonisins in pathogenesis on maize under field conditions was tested using strains of *Fusarium verticillioides*. One strain producing FB<sub>1</sub>, FB<sub>2</sub> and FB<sub>3</sub>, one strain producing only FB<sub>2</sub>, one strain producing only FB<sub>3</sub> and one fumonisin nonproducing strain were applied to ears via the silk channel and on seeds at planting. Disease severity on the harvested ears was evaluated by visible symptoms and by weight percent symptomatic kernels. All three FB<sub>1</sub> nonproducing strains were able to infect ears following either silk channel application or seed application at planting and were as effective as the FB<sub>1</sub> producing strain in causing ear rot following silk channel application.

DOMBRINK-KURTZMAN, M.A., DVORAK, T.J., BARRON, M.E. and ROONEY, L.W. 2000. **Effect of nixtamalization (alkaline cooking) on fumonisin-contaminated corn for production of masa and tortillas.** *Journal of Agricultural and Food Chemistry* **48**: 5781–5786.

The fate of fumonisins during the process of alkaline cooking (nixtamalisation) was investigated using normal appearing corn that was naturally contaminated with FB<sub>1</sub> at 8.79 mg/kg. Corn was processed into tortillas, starting with raw corn that was

cooked with lime and allowed to steep overnight. The steeped corn (nixtamal) was washed and ground into masa, which was used to make tortillas. Tortillas contained approximately 0.50 mg/kg of FB<sub>1</sub> plus 0.36 mg/kg of hydrolysed FB<sub>1</sub>, which represented 18.5% of the initial FB<sub>1</sub> concentration. Seventy-five percent of the original amount of fumonisin was present in the liquid fractions, primarily as hydrolysed FB<sub>1</sub>. Nixtamalisation significantly reduced the amount of fumonisin in maize.

## Fumonisin – Methodology

HARTL, M. and HUMPF, H.U. 2000. **Toxicity assessment of fumonisins using the brine shrimp (*Artemia salina*) bioassay.** *Food and Chemical Toxicology* **38**: 1097–1102.

FB<sub>1</sub> and FB<sub>2</sub>, their hydrolysed analogues HFB<sub>1</sub> and HFB<sub>2</sub> and the recently discovered fumonisin derivatives N-palmitoyl-HFB<sub>1</sub> and N-carboxymethyl-FB<sub>1</sub> were compared for their toxicity in a short term bioassay using brine shrimp (*Artemia salina*). The brine shrimp were exposed to the fumonisins in microwell plates with a mortality endpoint after 48 hr. FB<sub>1</sub> was the most toxic whereas its N-carboxymethyl analogue was 100-fold less effective. The hydrolysed fumonisins showed a 4- to 6-fold reduced toxicity compared to FB<sub>1</sub>. N-Palmitoyl-HFB<sub>1</sub> had a higher LC<sub>50</sub> value than its precursor HFB<sub>1</sub>. The brine shrimp assay proved to be a convenient and rapid system for toxicity assessment of this group of mycotoxins.

## Fumonisin – Toxicology

ZOMBORSZKY-KOVACS, M., VETESI, F.F., KOVACS, F., BATA, A., TOTH, A. and TORNOS, G. 2000. **Preliminary communication: Examination of the harmful effect to fetuses of fumonisin B<sub>1</sub> in pregnant sows.** *Teratogenesis Carcinogenesis and Mutagenesis* **20**: 293–299.

Three sows were fed a diet mixed with *Fusarium moniliforme* culture material to give FB<sub>1</sub> at 300 mg/sow/day from day 107 of pregnancy. FB<sub>1</sub> was administered to two sows for an additional 7 days subsequent to parturition (total 1–16 days). The third sow was given the toxin only until parturition (total 7 days). There were no symptoms observed in any of the sows. Piglets were sacrificed either immediately after parturition and prior to the first suckling, after 24 hr or on the 7th day postparturition. Feeding FB<sub>1</sub> to sows resulted in damage to the foetuses *in utero*. Changes indicating toxic effect were intraalveolar, subpleural and

interstitial pulmonary oedema of various degrees of severity, as well as changes in the histopathological sections of the liver, increases in the activities of liver enzymes and changes in the sphinganine/sphingosine ratio. In the piglets of the sows fed the toxin for an additional 7 days subsequent to parturition, mild pulmonary oedema could be detected after colostrum suckling 24 hr and 7 days after parturition.

BONDY, G.S., BARKER, M.G., LOMBAERT, G.A., ARMSTRONG, C.L., FERNIE, S.M., GUROFSKY, S., HUZZEL, V., SAVARD, M.E. and CURRAN, I.H.A. 2000. **A comparison of clinical, histopathological and cell-cycle markers in rats receiving the fungal toxins fumonisin B<sub>1</sub> or fumonisin B<sub>2</sub> by intraperitoneal injection.** Food and Chemical Toxicology **38**: 873–886.

The comparative toxicity of FB<sub>1</sub> and FB<sub>2</sub> was examined in male rats dosed ip at 0.75 mg/kg body weight for 2, 4 or 6 consecutive days. In the liver, mRNA expression for the cyclin kinase inhibitor p21 gene was significantly increased. Expression of mRNA for the cyclin D1 gene was significantly depressed in FB<sub>2</sub> treated rats. Hepatic cyclin E mRNA was elevated in response to FB<sub>1</sub> and FB<sub>2</sub>. In FB<sub>2</sub> treated animals this corresponded with decreased liver p27 mRNA expression. Hepatic proliferating cell nuclear antigen (PCNA) transcription was elevated in FB<sub>1</sub> but not FB<sub>2</sub> treated rats. Changes in liver microsomal protein levels of p27, cyclin E and PCNA were similar to changes in gene expression. In contrast, cyclin D1 protein levels were elevated in rats treated with FB<sub>1</sub> and, to a lesser extent, FB<sub>2</sub>.

DOMBRINK-KURTZMAN, M.A., GOMEZ-FLORES, R. and WEBER, R.J. 2000. **Activation of rat splenic macrophage and lymphocyte functions by fumonisin B<sub>1</sub>.** Immunopharmacology **49**: 401–409.

The effects of FB<sub>1</sub> on rat splenic macrophage and lymphocyte functions were investigated. Pretreatment (24 hr) of resident macrophages with FB<sub>1</sub> at 1, 10 or 100 mg/L significantly stimulated nitric oxide production after 72 hr of culture. FB<sub>1</sub> and IFN-gamma acted in an additive manner to activate nitric oxide production. The response of IFN-gamma activated macrophages was potentiated by FB<sub>1</sub>. In addition, FB<sub>1</sub> significantly potentiated Con A and antiTCR, IL-2 or antiTCR + IL-2 induced proliferation of splenic cells in the presence of the nitric oxide synthase inhibitor N-G-monomethyl-L-arginine. These results show two distinct and separate effects of FB<sub>1</sub>: it induces nitric oxide production by macrophages and it stimulates T-cell proliferation.

MELI, R., FERRANTE, M.C., RASO, G.M., CAVALIERE, M., DICARLO, R. and LUCISANO, A. 2000. **Effect of fumonisin B<sub>1</sub> on inducible nitric oxide synthase and cyclooxygenase-2 in LPS-stimulated J774A.1 cells.** Life Sciences **67**: 2845–2853.

The cytotoxic effect of FB<sub>1</sub> at 1–100 mM was evaluated using a murine macrophage cell line (J774A.1) as model system. The effect of FB<sub>1</sub> on nitric oxide (NO) and prostaglandin E-2 (PGE(2)) production induced by lipopolysaccharide (LPS) was also investigated. Macrophages were pretreated with FB<sub>1</sub> for 72 hr and then stimulated with LPS for 24 hr. The increase of LPS induced production of these inflammatory mediators was observed at increasing concentrations of FB<sub>1</sub> and was concentration dependent. Western blot analysis demonstrated that the observed increase of NO and PGE(2) production by FB<sub>1</sub> was related to an enhancement of iNOS and COX-2 expression.

VANDERWESTHUIZEN, L., SHEPHARD, G.S. and VANSCHALKWYK, D.J. 2001. **The effect of a single gavage dose of fumonisin B<sub>1</sub> on the sphinganine and sphingosine levels in vervet monkeys.** Toxicology **39**: 273–281.

Vervet monkeys were given a single gavage dose of FB<sub>1</sub> at 1 or 10 mg/kg body weight. In the high dose monkeys the serum sphinganine(Sa)/sphingosine(So) ratio, as well as levels of serum cholesterol and liver function enzymes, increased during the first week after dosing and remained elevated for several weeks thereafter. The urinary Sa/So ratio and the serum renal function indicators showed a more rapid response and a correspondingly more rapid return to predosing levels. In the low dose monkeys, serum Sa and the Sa/So ratio were the only parameters to increase above the control levels. The serum Sa/So ratio was exclusively elevated above the control levels in the low and high dose monkeys and seems more relevant as a marker for fumonisin exposure than any of the other indicators.

HENRY, M.H., WYATT, R.D. and FLETCHER, O.J. 2000. **The toxicity of purified fumonisin B<sub>1</sub> in broiler chicks.** Poultry Science **79**: 1378–1384.

Broiler chicks were fed FB<sub>1</sub> at 0, 20, 40 or 80 mg/kg from 0 to 21 days of age. FB<sub>1</sub> at concentrations of 80 mg/kg or less did not adversely affect body weight, feed efficiency or water consumption. Total liver lipids of chicks fed FB<sub>1</sub> at 40 or 80 mg/kg were significantly lower than those of the chicks fed either 0 or 20 mg/kg of feed. Liver Sa concentration and the Sa:So ratio were increased significantly in all treated groups. Chicks fed FB<sub>1</sub> at 80 mg/kg had significantly higher serum glutamate oxaloace-

tate aminotransaminase:aspartate aminotransferase ratios and levels of free sphinganine in the serum.

## Trichothecenes – General

CLEAR, R.M., PATRICK, S.K. and GABA, D. 2000. **Prevalence of fungi and fusariotoxins on oat seed from western Canada, 1995–1997.** Canadian Journal of Plant Pathology **22**: 310–314.

The mycoflora and levels of deoxynivalenol (DON) on oat seed (*Avena sativa*) was determined in grain samples collected from 39 crop districts in Alberta, Saskatchewan and Manitoba, Canada, during 1995–1997. *Alternaria alternata* was the most frequently isolated species from each province. DON levels greater than or equal to 0.10 mg/kg were found in seeds from two Manitoba crop districts in 1996 and three in 1997, with a maximum level of 0.34 mg/kg. In 1997, DON was also detected in composite samples of seed from three Saskatchewan and two Alberta crop district composites.

CONTRERAS, M.D.M., YEPEZ, A.J.M. and MARTINEZ, R.R. 2000. **[Determination of deoxynivalenol (DON) in wheat, barley and corn and its relationship with the levels of total molds, *Fusarium* spp., infestation percentage, and water activity].** Archivos Latinoamericanos de Nutricion **50**: 183–186.

Fifty samples of cereals including wheat, barley and corn were analysed for DON. Water activity (a<sub>w</sub>) and mycoflora levels were also investigated. The highest level of infection (12–80%) and the highest count of total moulds were detected in wheat samples, while the highest levels of *Fusarium* species were detected in white corn. DON was found in wheat and barley samples but not in corn. The wheat red winter soft samples showed the highest levels of DON (3.2 mg/kg). There was no correlation between mould count, *Fusarium* species, infestation grade, a<sub>w</sub> or DON levels. (In Spanish).

OMURTAG, G.Z. and YAZICIOGLU, D. 2000. **Determination of T-2 toxin in grain and grain products by HPLC and TLC.** Journal of Environmental Science and Health Part B – Pesticides Food Contaminants and Agricultural Wastes **35**: 797–807.

A total of 30 commercially available grain and grain product samples from Turkey were analysed for T-2 toxin. T-2 toxin was determined using HPLC with UV detection and TLC. Two corn flour samples contained T-2 toxin at levels of 1.60 and 4.08 mg/kg.

RAYMOND, S.L., HEISKANEN, M., SMITH, T.K., REIMAN, M., LAITINEN, S. and CLARKE, A.F. 2000.

**An investigation of the concentrations of selected *Fusarium* mycotoxins and the degree of mold contamination of field-dried hay.** Journal of Equine Veterinary Science **20**: 616.

The levels of DON, T-2 toxin and ZEA and mould contamination of Ontario field-dried hay from 10 performance horse farms were examined. DON was present in the highest amounts and the levels of DON found in this study could potentially have an influence on the health of horses consuming such hay.

SAVARD, M.E., SINHA, R.C., SEAMAN, W.L. and FEDAK, G. 2000. **Sequential distribution of the mycotoxin deoxynivalenol in wheat spikes after inoculation with *Fusarium graminearum*.** Canadian Journal of Plant Pathology **22**: 280–285.

One central spikelet of spring wheat (*Triticum aestivum*) cv. Roblin spikes was inoculated with macroconidia of *Fusarium graminearum* and the entire spikes were harvested at 2 to 4 day intervals from 2 to 25 days after inoculation. The amount of DON in each spikelet and in each internode of the rachis was measured by ELISA. High concentrations of DON were first detected in the inoculated spikelets, 4 days after inoculation. DON concentrations in the spikelets below the inoculation point eventually reached 500–600 mg/kg while the corresponding internodes of the rachis contained 1000–1200 mg/kg. Much lower amounts of DON were found in spikelets and rachis above the inoculation point.

ITTU, M., GRABARKIEWICZ-SZCZESNA, J., KOSTECKI, M. and GOLINSKI, P. 2000. **Deoxynivalenol accumulation and other scab symptoms in six Romanian wheat genotypes inoculated with *Fusarium graminearum*.** Mycotoxin Research **16**: 15–22.

Six Romanian winter wheat genotypes were inoculated at anthesis with three *Fusarium graminearum* isolates. There were important differences between the six examined wheat genotypes in terms of DON accumulation, *Fusarium* head blight development, yield reduction and models of host-pathogen interactions. *Fusarium* damaged kernels was highly correlated with DON content. Area under the disease progress curve was also significantly correlated with DON content.

LAMPER, C., TEREN, J., BARTOK, T., KOMOROCZY, R., MESTERHAZY, A. and SAGI, F. 2000. **Predicting DON contamination in *Fusarium*-infected**

**wheat grains via determination of the ergosterol content.** Cereal Research Communications **28**: 337–344.

Four winter wheat varieties of different *Fusarium* resistance were artificially infected with various isolates of *Fusarium graminearum* and *F. culmorum*. Kernel infection and DON and ergosterol contents were assessed by visual inspection, reversed phase HPLC and GC/MS, respectively. Both DON and ergosterol contents correlated with kernel infection strongly and positively. Similarly, there was a strong, positive correlation between DON and ergosterol contents in the infected grains.

KESHRI, G. and MAGAN, N. 2000. **Detection and differentiation between mycotoxigenic and non-mycotoxigenic strains of two *Fusarium* spp. using volatile production profiles and hydrolytic enzymes.** Journal of Applied Microbiology **89**: 825–833.

Volatile profiles and hydrolytic enzyme production by one non-mycotoxigenic and three mycotoxigenic strains of *Fusarium moniliforme* and *F. proliferatum* were examined for differentiation of isolates. After spore lawn inoculation, measurements were made after 48, 72 and 96 hr by sampling the head space above cultures with an electronic nose system using a 14 sensor surface polymer array, and by extraction and quantification of hydrolytic enzymes. Principal component analysis indicated that discrimination could be achieved between the non-mycotoxigenic strain and the mycotoxin producing strains for both species after 48 hr. The total and specific activity of 3/7 enzymes were found to increase significantly in the non-mycotoxigenic when compared with the toxigenic strains of both species after 72 hr. The study has shown for the first time that it is possible to differentiate between mycotoxigenic and non-mycotoxigenic strains of such spoilage fungi based on their volatile production patterns using an electronic nose system.

CHELKOWSKI, J., WISNIEWSKA, H., ADAMSKI, T., GOLINSKI, P., KACZMAREK, Z., KOSTECKI, M., PERKOWSKI, J. and SURMA, M. 2000. **Effects of *Fusarium culmorum* head blight on mycotoxin accumulation and yield traits in barley doubled haploids.** Journal of Phytopathology - Phytopathologische Zeitschrift **148**: 541–545.

The susceptibility of barley doubled haploids to *Fusarium* head blight was investigated. Heads of 24 doubled haploids lines derived from F-1 Maresi (two-rowed) x Pomo (six-rowed) hybrids were inoculated with a conidial suspension of *Fusarium culmorum*. The average nivalenol concentration in kernels of inoculated lines ranged

from 0.15 mg/kg in the two-rowed line MP7 to 6.36 mg/kg in the six-rowed line MP113. A low accumulation of DON was observed (from 0.01 to 0.20 mg/kg). Generally, no significant differences in mycotoxin content were found between two-rowed and six-rowed genotypes.

MORIN, L., GIANOTTI, A.F. and LAUREN, D.R. 2000. **Trichothecene production and pathogenicity of *Fusarium tumidum*, a candidate bioherbicide for gorse and broom in New Zealand.** Mycological Research **104**: 993–999.

The relationship between trichothecene production and pathogenicity was investigated for 29 isolates of *Fusarium tumidum*, a potential bioherbicide for gorse (*Ulex europaeus*) and broom (*Cytisus scoparius*) in New Zealand. All isolates originally derived from broom produced high levels of T-2 tetraol derivatives when grown on ground maize kernels and pearl barley grains, compared with isolates from gorse. Low amounts of scirpentriol derivatives were also produced by both groups of isolates. No nivalenol and DON derivatives were detected in any of the culture extracts. Two isolates offer prospects for the development of a safe bioherbicide that could target two major weeds in New Zealand, as trichothecenes were not detected from them at the higher concentrations.

PINEDA-VALDES, G. and BULLERMAN, L.B. 2000. **Thermal stability of moniliformin at varying temperature, pH, and time in an aqueous environment.** Journal of Food Protection **63**: 1598–1601.

The effects of temperature (100, 125 and 150°C) and pH (4, 7 and 9) on the stability of moniliformin (MON) were determined in aqueous buffer solutions at processing times ranging from 10 to 60 min. The percentage of MON reduction was positively related to increasing temperature and pH. After 60 min at pH 4 and 150°C, MON was reduced by only 5%. After 60 min at pH 10 and 100, 125 or 150°C, MON was reduced by 56, 72 and 83%, respectively.

RAZZAZI, E., BOHM, J., AHMED, K.E., CECON, B. and RABUS, B. 2000. **Investigation on the biodegradability of mycotoxins nivalenol (NIV) and deoxynivalenol (DON) in a RUSITEC fermentor and their monitoring by HPLC/MS.** Mycotoxin Research **16**: 9–14.

The biodegradation of nivalenol (NIV) and DON were studied in a RUSITEC (rumen simulation technique) system. NIV at 1 mg/kg and DON at 2 mg/kg were added to vessels containing the rumen fluid. The kinetics of NIV and DON biodegradation during the fermentation process were moni-

tored by HPLC combined with MS and an atmospheric pressure chemical ionisation interface.

### Tricothecenes – Methodology

WHITAKER, T.B., HAGLER, W.M., GIESBRECHT, F.G. and JOHANSSON, A.S. 2000. **Sampling, sample preparation, and analytical variability associated with testing wheat for deoxynivalenol.** Journal of AOAC International **83**: 1285–1292.

The variability associated with testing wheat for DON was measured using a 0.454 kg sample, Romer mill, 25 g comminuted subsample and the Romer Fluoroquant analytical method. The total variability was partitioned into sampling, sample preparation, and analytical variability components. Each variance component was a function of the DON concentration and equations were developed to predict each variance component using regression techniques. For the test procedure, the coefficient of variation (CV) associated with testing wheat at 5 mg/kg was 13.4%. The CVs associated with sampling, sample preparation, and analysis were 6.3, 10.0 and 5.3%, respectively. The CVs associated with testing wheat are relatively small compared to the CV associated with testing other commodities for other mycotoxins.

FREESE, L., FRIEDRICH, R., KENDALL, D. and TANNER, S. 2000. **Variability of deoxynivalenol measurements in barley.** Journal of AOAC International **83**: 1259–1263.

In assaying barley for DON contamination, a study to measure the variability among measurements of different sample sizes found no detectable differences in variability attributable to sample size. It was concluded that the variability among DON concentrations in samples from the lot was small relative to that introduced by the measurement process (combined sample preparation and analysis). A separate experiment investigated variation among samples taken from a lot, variation among subsamples taken from ground samples, and variation among multiple replicated measurements of an extract. On 10 lots, all three sources were significant contributors to variation. Stratification of DON within lots was hypothesised as a source of variation of DON measurements. Tests indicated that some stratification might exist.

MARAGOS, C.M. and MCCORMICK, S.P. 2000. **Monoclonal antibodies for the mycotoxins deoxynivalenol and 3-acetyl-deoxynivalenol.** Food and Agricultural Immunology **12**: 181–192.

Three monoclonal antibodies were developed following the immunisation of mice with a conjugate of DON and ovalbumin. One of these antibodies was selected for the development of a competitive direct ELISA (CD-ELISA). The IC<sub>50</sub> for colour development in the assay for DON was 18 mg/L. The antibody from this clone showed strong cross-reactivity to 3-acetylDON, with an IC<sub>50</sub> of 2.9 mg/L. Cross-reactivity to 19 other tricothecene mycotoxins was low. The CD-ELISA was applied to wheat spiked with DON over the range 0.01–10 mg/kg. Recoveries over the range 0.05–5 mg/kg averaged 88.7% with a coefficient of variation of 10.9%.

### Tricothecenes – Toxicology

HASSANANE, M.S., ABDALLA, E.S.A., EL-FIKY, S., AMER, M.A. and HAMDY, A. 2000. **Mutagenicity of the mycotoxin diacetoxyscirpenol on somatic and germ cells of mice.** Mycotoxin Research **16**: 53–59.

Mice were injected ip with single or repeated doses of diacetoxyscirpenol (DAS) at 0.5, 0.75 or 1.0 mg/kg body weight. DAS treatment resulted in a significant reduction in mitotic activity at all levels of doses tested. In somatic cells (bone marrow) and in germ cells (testicles), both structural and numerical chromosome abnormalities were also observed. In general, the frequencies of scored abnormalities in germ cells were lower than that in somatic cells. Abnormalities of sperm shape were also observed.

MEYER, K., USLEBER, E., MARTLBAUER, E. and BAUER, J. 2000. **[Occurrence of zearalenone, alpha- and beta-zearalenol in biles of breeding sows in relation to the reproductive performance].** Berliner und Munchener Tierarztliche Wochenschrift **113**: 374–379.

The correlation of ZEA and ZEA derivatives in bile and feed with fertility problems of unknown origin in sows was investigated. ZEA and ZEA derivatives were detected in 96.2% of bile samples analysed. The contamination rate of feeding stuffs was 25.9%. However, a correlation between the occurrence of ZEA, alpha- and beta-zearalenol in bile of sows and noninfectious reproductive disorders could not be established at the loading level found. (In German).

RAFAI, P., PETERSSON, H., BATA, A., PAPP, Z., GLAVITS, R., TUBOLY, S., VANYI, A. and SOOS, P. 2000. **Effect of dietary T-2 fusariotoxin concentrations on the health and production of white Pekin duck broilers.** Poultry Science **79**: 1548–1556.

White Pekin ducks were given feed containing T-2 toxin at 0.2–4.0 mg/kg from day 1 until 49 days of age. Dermatotoxic oral lesions developed in most experimental ducks within 2 days. The gradual disappearance of macroscopic signs indicated the development of tolerance in ducks treated with the lower T-2 toxin content. No repair was found in the 3 and 4 mg/kg groups. Dietary concentrations of T-2 toxin below 0.4 mg/kg had no effect on the average weekly weight gain in the first 6 weeks, but a severe decrease was found in the last week of the experiment. T-2 toxin concentrations of 1 mg/kg and greater uniformly depressed growth rate. From week 3 on, the feed intakes of the 0.6 to 4 mg/kg groups were usually less than that of the control group. Serum and plasma chemical values and haematological parameters failed to show dose dependent effects.

LEITGEB, R., LEW, H., KHIDR, R., BOHM, J., ZOLLITSCH, W. and WAGNER, E. 2000. **Influence of Fusarium toxins on growth and carcass characteristics of turkeys.** Bodenkultur **51**: 171–178.

Turkeys were fed diets containing maize contaminated with MON (4.94 mg/kg), beauvericin (3.24 mg/kg), DON (2.02 mg/kg) and FB<sub>1</sub> (0.35 mg/kg). Maize accounted for 0, 36.8, 48.9 or 59.3 % of the total diet. At the end of the growing period (77 days) live weight of the turkeys were 6.71, 6.26, 6.33 and 6.27 kg, respectively, and feed conversion rates were 2.07, 2.16, 2.23 and 2.19, respectively. The dressing percentages of eviscerated carcass and roast carcass, the weight of heart, liver Bursa fabricii, spleen and the valuable parts of carcass showed no significant differences between the feeding groups.

CURTUI, V.G. 2000. **Effects of feeding a Fusarium poae extract and a natural zeolite to broiler chickens.** Mycotoxin Research **16**: 43–52.

One-day-old broiler chicks were fed diets containing *Fusarium poae* culture material, with or without the addition of 0.5% zeolite, for 28 days. *F. poae* culture material, administered singly or in combination with zeolite, caused reductions in body weight gains, feed intake, feed utilisation and water consumption. In addition, there were significant decreases in leukocyte counts, serum total protein and serum albumin. For some parameters the addition of zeolite increased the adverse effects of the *F. poae* extract.

EUDES, F., COMEAU, A., RIOUX, S. and COLLIN, J. 2000. **[Phytotoxicity of eight mycotoxins associated with**

**Fusarium in wheat head blight].** Canadian Journal of Plant Pathology **22**: 286–292.

Eight mycotoxins from *Fusarium* species including DON, 3-acetylDON, NIV, T-2 toxin, HT-2 toxin, DAS and ZEA, were shown to be phytotoxic to wheat. The inhibition of coleoptile elongation by each trichothecene was well described by a linear equation. DON and 3-acetylDON were 2.5 times more toxic than T-2 toxin, 8 times more toxic than HT-2 toxin and 13 times more toxic than DAS. Moreover, reduction of coleoptile elongation of these cultivars was correlated with scab susceptibility. (In French).

## Aflatoxins – General

BAKIRCI, I. 2001. **A study on the occurrence of aflatoxin M<sub>1</sub> in milk and milk products produced in Van province of Turkey.** Food Control **12**: 47–51.

The levels of AFM<sub>1</sub> in raw milk samples and the carry-over and fate of AFM<sub>1</sub> in milk products during manufacture in the dairy plant of Yuzuncu Yil University, Turkey, were studied. There were no statistical differences between AFM<sub>1</sub> contents of bulk milk and pasteurised milk, skim milk, yoghurt, buttermilk and whey. AFM<sub>1</sub> contents of white-pickled cheese and Kashar cheese samples were higher than those of bulk milk samples, whereas those of cream and butter samples were lower.

MARTINS, M.L. and MARTINS, H.M. 2000. **Aflatoxin M<sub>1</sub> in raw and ultra high temperature-treated milk commercialized in Portugal.** Food Additives and Contaminants **17**: 871–874.

Thirty-one samples of raw milk and 70 samples of ultrahigh temperature treated (UHT) milk including whole milk, semi-skimmed milk and skimmed milk, were collected in supermarkets in Lisbon, Portugal. AFM<sub>1</sub> was detected in 80.6% of raw milk and 84.2% of UHT milk. Among raw milk samples, 54.8% contained levels of AFM<sub>1</sub> between 5–10 mg/L and 19.3% had levels between 21 and 50 mg/L. Among UHT milk samples analysed, 44.4% of whole milk, 4.5% of semi-skimmed milk and 53.4% of skimmed milk had AFM<sub>1</sub> contamination levels between 11–20 mg/L, while 33.3% of whole milk and 81.9% of semi-skimmed milk had levels between 21–50 mg/L.

BLUTHGEN, A. and SCHWERT-FEGER, M. 2000. **[Elimination of aflatoxin M<sub>1</sub> with the milk of lactating cows after simultaneous feeding of adsorptive additives and experiments with**

**aflatoxin B<sub>1</sub> – in vivo and in vitro].** Kieler Milchwirtschaftliche Forschungsberichte **52**: 145–164.

Lactating cows were fed AFB<sub>1</sub> at 50 mg/day with or without the addition of a sorbent (five clay minerals and two yeast extracts) at 40–400 g/day for 4 days. The concentration of AFM<sub>1</sub> in milk was measured by ELISA. The mineral products showed moderate effects leading on average to a reduced elimination of AFM<sub>1</sub> in milk of approximately 14%. The esterified glucomanoses of yeast had nearly no effect on the aflatoxin content of milk. Bentonites demonstrated a relatively high adsorption of AFB<sub>1</sub>. Adding epoxidases to the adsorbent decreased the adsorption capacity whereas the adsorption of clinoptilolith was more or less low. (In German).

BILOTTI, L.G., PINTO, V.E.F. and VAAMONDE, G. 2000. **Aflatoxin production in three selected samples of triticale, wheat and rye grown in Argentina.** Journal of the Science of Food and Agriculture **80**: 1981–1984.

Triticale, a hybrid resulting from crossing wheat and rye, was evaluated as a substrate for aflatoxin accumulation in comparison with its parents. Aflatoxin (B<sub>1</sub>, B<sub>2</sub>, G<sub>1</sub> and G<sub>2</sub>) accumulation curves were obtained for the three substrates inoculated with *Aspergillus parasiticus* and incubated at 25°C and a<sub>w</sub> 0.925 for 10 weeks. Wheat and triticale were poor substrates for aflatoxin production. Rye was more prone than the other substrates to rapid colonisation by *A. parasiticus* and accumulated larger aflatoxin quantities. The maximum aflatoxin concentration in rye (11.84 mg/kg) was significantly larger than those obtained in wheat (2.15 mg/kg) and triticale (2.85 mg/kg).

SHARMA, Y.P. and SUMBALI, G. 1999. **Incidence of aflatoxin producing strains and aflatoxin contamination in dry fruit slices of quinces (*Cydonia oblonga* Mill.) from the Indian State of Jammu and Kashmir.** Mycopathologia **148**: 103–107.

The occurrence of aflatoxins and the aflatoxin producing potential of *Aspergillus flavus* strains isolated from dry fruit slices of quinces produced in Jammu and Kashmir, India, were investigated. Of 147 *Aspergillus flavus* strains isolated from the dried fruit slices 23.14% were aflatoxigenic, producing AFB<sub>1</sub> and B<sub>2</sub> in varying amounts. Aflatoxins G<sub>1</sub> and G<sub>2</sub> were not detected. All 25 of the investigated market samples contained AFB<sub>1</sub> with the level of contamination ranging from 96 to 8164 mg/kg of the dry fruit.

HASKARD, C., BINNION, C. and AHOKAS, J. 2000. **Factors affecting the sequestration of aflatoxin by *Lactobacillus rhamnosus* strain GG.** Chemico-Biological Interactions **128**: 39–49.

The interaction of AFB<sub>1</sub> with a probiotic strain of lactic acid bacteria *Lactobacillus rhamnosus* strain GG (GG) has been investigated. The binding of AFB<sub>1</sub> to GG in the late exponential-early stationary phase was studied for viable, heat-killed and acid-killed bacteria. In general, viable, heat-killed and acid-killed GG responded in a similar manner. The effects of pronase E, lipase and m-periodate on AFB<sub>1</sub> binding and release were consistent with AFB<sub>1</sub> binding predominantly to carbohydrate components of the bacteria. The effect of urea suggested hydrophobic interactions play a major role in binding.

PELTONEN, K.D., EL-NEZAMI, H.S., SALMINEN, S.J. and AHOKAS, J.T. 2000. **Binding of aflatoxin B<sub>1</sub> by probiotic bacteria.** Journal of the Science of Food and Agriculture **80**: 1942–1945.

The abilities of six probiotic bacteria including six *Lactobacillus* strains and one *Bifidobacterium* strain to bind AFB<sub>1</sub> was assessed. The strains were incubated *in vitro* with AFB<sub>1</sub> and the toxin residue in the supernatant was measured using HPLC. The aflatoxin binding capacity of the strains ranged from 5.8 to 31.3%.

JAYASHREE, T. and SUBRAMANYAM, C. 2000. **Oxidative stress as a prerequisite for aflatoxin production by *Aspergillus parasiticus*.** Free Radical Biology and Medicine **29**: 981–985.

The relevance of free radical generation and oxidative stress to aflatoxin production was examined by comparing the oxygen requirement and antioxidant status of a toxigenic strain of *Aspergillus parasiticus* with that of a nontoxigenic strain at early (trophophase) and late logarithmic (idiophase) growth phases. In the nontoxigenic strain, oxygen requirements were relatively unaltered at various growth phases, whereas the toxigenic strain exhibited greater oxygen requirements at trophophase coinciding with onset of aflatoxin production. The activities of antioxidant enzymes were all enhanced during the progression of the toxigenic strain from trophophase to idiophase. The combined results suggest that aflatoxin production by the toxigenic strain may be a consequence of increased oxidative stress leading to enhanced lipid peroxidation and free radical generation.

AZIZ, N.H., SHAHIN, A.A.M., ABOUZEID, A.A.M. and EL-ZEANY, S.A. 2000. **Correlation of growth and aflatoxin production by *Aspergillus flavus***

with some essential metals in gamma irradiated crushed corn. *Nahrung* **44**: 354–359.

The effects of gamma irradiation and some essential metals on growth and AFB<sub>1</sub> production by *Aspergillus flavus* in crushed corn were investigated. AFB<sub>1</sub> production was influenced by the addition of zinc, copper or iron and the effect gradually decreased with increasing metal concentration from 0 to 300 mg/kg. The presence of 100 mg/kg zinc, copper or iron plus gamma irradiation (0.5, 1.0, 2.0 kGy) enhanced the growth of *A. flavus* and the production of aflatoxin in contrast with irradiated samples alone. *A. flavus* was able to metabolise and deplete elements in all gamma irradiated samples.

MAHONEY, N., MOLYNEUX, R.J. and CAMPBELL, B.C. 2000. **Regulation of aflatoxin production by naphthoquinones of walnut (*Juglans regia*)**. *Journal of Agricultural and Food Chemistry* **48**: 4418–4421.

The effects of four naphthoquinones which occur in walnut husks on fungal viability and aflatoxigenesis was studied. The quinones delayed germination of the fungus and completely inhibited growth at higher concentrations. Their effect on aflatoxin levels was highly dependent on the concentration of individual naphthoquinones in the media. At higher concentrations, aflatoxin production was decreased or completely inhibited, but at lower concentrations there was a stimulatory effect on aflatoxin biosynthesis. Structural features of the naphthoquinones associated with decreased fungal viability and greatest effect on aflatoxigenesis were the presence of a 5-hydroxyl or 2-methyl substituent, but there was no significant additive effect when both of these substituents were present.

MARNEWICK, J.L., GELDERBLUM, W.C.A. and JOUBERT, E. 2000. **An investigation on the antimutagenic properties of South African herbal teas**. *Mutation Research – Genetic Toxicology and Environmental Mutagenesis* **471**: 157–166.

The antimutagenic properties of South African herbal teas were investigated using the *Salmonella typhimurium* mutagenicity assay. Aqueous extracts of fermented and unfermented rooibos tea (*Aspalathus linearis*) and honeybush tea (*Cyclopia intermedia*) both possess antimutagenic activity against AFB<sub>1</sub> induced mutagenesis using tester strains TA98 and TA100 in the presence of metabolic activation. The data suggest that two mechanisms are involved in the antimutagenicity of the tea extracts towards carcinogens that require metabolic activation: the tea components may interfere

with cytochrome P350-mediated metabolism of these mutagens, and the direct interaction between the tea constituents, presumably the polyphenolic compounds, with the promutagens and/or the active mutagenic metabolites.

ZHOU, R., RASOOLY, R. and LINZ, J.E. 2000. **Isolation and analysis of *fluP*, a gene associated with hyphal growth and sporulation in *Aspergillus parasiticus***. *Molecular and General Genetics* **264**: 514–520.

The *fluP* gene was cloned by screening an *Aspergillus parasiticus* genomic DNA library with a cDNA probe encoding part of a polyketide synthase (PKS), the 6-methylsalicylic acid synthase from *Penicillium patulum*. FluP was hypothesised to function as a PKS in aflatoxin biosynthesis. Disruption of *fluP* in *A. parasiticus* resulted in the loss of *fluP* transcript, a 3- to 4-fold reduction in hyphal growth rate, the appearance of a fluffy, cotton-like hyphal morphology, reduction or elimination of asexual spores and spore bearing structures, and a 2-fold reduction in aflatoxin accumulation. The data suggest that *fluP* encodes a novel PKS associated with hyphal growth and cell development (sporulation), whose activity indirectly influences aflatoxin accumulation in *A. parasiticus*.

CHANG, P.K., YU, J.J., EHRlich, K.C., BOUE, S.M., MONTALBANO, B.G., BHATNAGAR, D. and CLEVELAND, T.E. 2000. ***adhA* in *Aspergillus parasiticus* is involved in conversion of 5'-hydroxyaverantin to averufin**. *Applied and Environmental Microbiology* **66**: 4715–4719.

The *adhA* gene was isolated from the aflatoxin gene cluster of *Aspergillus parasiticus* SU-1. *A. parasiticus* SU-1, which produces aflatoxins, has two copies of *adhA* (*adhA1*), whereas *A. parasiticus* SRRC 2043, a strain that accumulates *O*-methylsterigmatocystin, has only one copy. Disruption of *adhA* in SRRC 2043 resulted in a strain that accumulates predominantly 5'-hydroxyaverantin. This result suggests that ADHA is involved in the dehydrogenation of 5'-hydroxyaverantin to averufin.

ANNIS, S.L., VELASQUEZ, L., XU, H.X., HAMMERSCHMIDT, R., LINZ, J. and TRAIL, F. 2000. **Novel procedure for identification of compounds inhibitory to transcription of genes involved in mycotoxin biosynthesis**. *Journal of Agricultural and Food Chemistry* **48**: 4656–4660.

A novel assay is described for the identification and isolation of compounds that inhibit the transcription of genes involved in mycotoxin biosynthesis. The TLC based

assay was used to screen plant extracts for compounds that would inhibit the expression of the beta-glucuronidase reporter gene under the control of an aflatoxin biosynthesis gene promoter in *Aspergillus parasiticus*. The assay was used to track purification of an inhibitory compound, cp2, from extracts of black pepper (*Piper nigrum*). Cp2 inhibited aflatoxin biosynthesis at the transcriptional level.

YU, J.J., CHANG, P.K., BHATNAGAR, D. and CLEVELAND, T.E. 2000. **Cloning of a sugar utilization gene cluster in *Aspergillus parasiticus***. *Biochimica et Biophysica Acta – Gene Structure and Expression* **1493**: 211–214.

At one end of the 70 kb aflatoxin biosynthetic pathway gene cluster in *Aspergillus parasiticus* and *Aspergillus flavus*, a group of four genes that constitute a well-defined gene cluster related to sugar utilisation in *A. parasiticus* has been cloned. The expression of the *hxtA* gene encoding a hexose transporter protein, was found to be concurrent with the aflatoxin pathway cluster genes, in aflatoxin conducive medium. This is significant since a close linkage between the two gene clusters could potentially explain the induction of aflatoxin biosynthesis by simple sugars such as glucose or sucrose.

## Aflatoxins – Methodology

STROKA, J., PETZ, M and ANKLAM, E. 2000. **Analytical methods for the determination of aflatoxins in various food matrices at concentrations regarding the limits set in European Regulations: Development, characteristics, limits**. *Mycotoxin Research* **16**: 23–42.

Methods for the determination of aflatoxins in paprika, peanut butter, pistachio paste and baby food were developed. The methods employ an immunoaffinity cleanup step and reversed phase LC. All steps of the analysis were tested for their suitability for all matrices with focus on method robustness, simplicity, toxicology, environment and user friendliness. Extraction procedures, chromatographic separation and postcolumn derivatisation techniques were elaborated for this purpose.

DALY, S.J., KEATING, G.J., DILLON, P.P., MANNING, B.M., O'KENNEDY, R., LEE, H.A. and MORGAN, M.R.A. 2000. **Development of surface plasmon resonance-based immunoassay for aflatoxin B<sub>1</sub>**. *Journal of Agricultural and Food Chemistry* **48**: 5097–5104.

Two different anti-AFB<sub>1</sub> antibodies were examined to develop a surface plasmon resonance based immunoassay to AFB<sub>1</sub>. A conjugate consisting of AFB<sub>1</sub>-bovine serum

albumin was immobilised on the dextran gel surface. Competition between immobilised AFB<sub>1</sub> conjugate and free AFB<sub>1</sub> in solution for binding to antibody injected over the surface formed the basis for the assay. Regeneration of the antibody from the immobilised conjugate surface is essential for the development of such an inhibitive immunoassay. Conventional regeneration solutions worked to a degree, but regeneration was at the expense of the integrity of the immobilised conjugate. A polyclonal anti-AFB<sub>1</sub> antibody was produced and was found to be regenerable using an organic solution consisting of 1M ethanolamine with 20% (v/v) acetonitrile, pH 12.0. This combined high ionic strength and extreme pH, as well as chaotropic properties and allowed the development of an inhibitive immunoassay. The assay had a linear range of 3.0–98.0 mg/L with good reproducibility.

PARK, D.L., WHITAKER, T.B., GIESBRECHT, F.G. and NJAPAU, H. 2000. **Performance of three pneumatic probe samplers and four analytical methods used to estimate aflatoxins in bulk cottonseed.** Journal of AOAC International **83**: 1247–1251.

In Arizona, USA, cottonseed samples for aflatoxin analysis are collected from 100 ton piles by an accumulation of three or more probings with a pneumatic probe. When sampling compacted cottonseed piles, the large official pneumatic probe (7.6 ¥ 127 cm) decreases in efficiency. Two smaller probes (1.9 ¥ 127 cm and 1.9 ¥ 254 cm) were therefore developed and tested for their suitability for sampling cottonseed piles. Three rapid analytical methods (one TLC and two immunochemical) were tested for suitability as on-site assay systems. An analysis of variance of the analytical test results showed no differences between the various probes tested. Of the rapid methods, only the AflaTest-P immunoaffinity column gave results similar to those of the official AOAC TLC method.

COKER, R.D., NAGLER, M.J., DEFIZE, P.R., DERKSEN, G.B., BUCHHOLZ, H., PUTZKA, H.A., HOOGLAND, H.P., ROOS, A.H. and BOENKE, A. 2000. **Sampling plans for the determination of aflatoxin B<sub>1</sub> in large shipments of animal feedstuffs.** Journal of AOAC International **83**: 1252–1258.

Incremental samples (50, 100, and 500 g) were systematically collected from large shipments of copra meal pellets, copra cake and palm kernel cake to study the distribution of AFB<sub>1</sub> and evaluate adherence of distribution to the model,  $CV_{is}^2(EQ) = A + B/M_{is}$  (where  $CV_{is}$  = coefficient of variation of the true concentration of AFB<sub>1</sub> within the incremental samples;  $M_{is}$  = mass of the

incremental samples; and A and B are constants). The coefficient of variation (CV) among incremental samples varied from 0 to 38%, and was independent of incremental sample size. No significant difference was found between the efficacy of four sample preparation (grinding and subdivision) methods when these methods were applied to copra meal pellets and cottonseed cake. Various sampling plans were evaluated with estimated CVs from 4.0 to 12.5%, for the AFB<sub>1</sub> content of the composite samples.

JOHANSSON, A.S., WHITAKER, T.B., HAGLER, W.M., GIESBRECHT, F.G., YOUNG, J.H. and BOWMAN, D.T. 2000. **Testing shelled corn for aflatoxin. Part I: Estimation of variance components.** Journal of AOAC International **83**: 1264–1269.

The variability associated with testing lots of shelled corn for aflatoxin was investigated. The total variance was estimated and partitioned into sampling, sample preparation, and analytical variances. All variances increased as aflatoxin concentration increased. With the use of regression analysis, mathematical expressions were developed to model the relationship between aflatoxin concentration and the total, sampling, sample preparation and analytical variances. The expressions for these relationships were used to estimate the variance for any sample size, subsample size and number of analyses for a specific aflatoxin concentration.

JOHANSSON, A.S., WHITAKER, T.B., GIESBRECHT, F.G., HAGLER, W.R. and YOUNG, J.H. 2000. **Testing shelled corn for aflatoxin. Part II: Modeling the observed distribution of aflatoxin test results.** Journal of AOAC International **83**: 1270–1278.

The suitability of several theoretical distributions to predict the observed distribution of aflatoxin test results in shelled corn was investigated. Fifteen positively skewed theoretical distributions were each fitted to 18 empirical distributions of aflatoxin test results for shelled corn. The compound gamma distribution was selected to model aflatoxin test results for shelled corn. The method of moments technique was chosen to estimate the parameters of the compound gamma distribution. Mathematical expressions were developed to calculate the parameters of the compound gamma distribution for any lot aflatoxin concentration and test procedure. Observed acceptance probabilities were compared to operating characteristic curves predicted from the compound gamma distribution, and all 18 observed acceptance probabilities were found to lie within a 95% confidence band.

JOHANSSON, A.S., WHITAKER, T.B., GIESBRECHT, F.G., HAGLER, W.M. and YOUNG, J.H. 2000. **Testing shelled corn for aflatoxin. Part III: Evaluating the performance of aflatoxin sampling plans.** Journal of AOAC International **83**: 1279–1284.

The effects of changes in sample size and/or sample acceptance level on the performance of aflatoxin sampling plans for shelled corn were investigated. Six sampling plans were evaluated for a range of sample sizes and sample acceptance levels. For a given sample size, decreasing the sample acceptance level decreases the percentage of lots accepted while increasing the percentage of lots rejected at all aflatoxin concentrations, and decreases the average aflatoxin concentration in lots accepted and lots rejected. For a given sample acceptance level that equals the regulatory guideline, increasing the sample size decreases misclassification of lots, both false positives and false negatives.

ANTOLINI, F., NERI, A., FLORIDI, A., FLORIDI, A. and FRANCIOSINI, S. 2000. **Aflatoxin M<sub>1</sub> in milk: Development of analytical and sampling strategies for the quality control in the dairy factories.** Industrie Alimentari **39**: 685–690.

An analytical strategy for a rapid and reliable screening of AFM<sub>1</sub> in milk at the points of milk production on farms was studied. The AFM<sub>1</sub> evaluation was performed by ELISA on 13 milk harvesting points for three days. The average level of AFM<sub>1</sub> in the harvesting points during the three days was 21.7 ng/L. The CV was high for both the different harvesting points on the same day and for the same harvesting point on the three different days. (In Italian).

SCHATZKI, T.F. 2000. Distribution of aflatoxin in pistachios. 7. **Sequential sampling.** Journal of Agricultural and Food Chemistry **48**: 4365–4368.

Sequential sampling for aflatoxin testing in pistachios was evaluated using the aflatoxin distribution and Monte Carlo results previously obtained. The sequential protocol was modelled on the current EU test protocol by applying a three-step sampling, using 10, 20 and 30 kg sample averages. An acceptance level of 15 mg/kg of total aflatoxin, under consideration for USA standards, was applied. Optimisation led to indifference regions of 2–30 mg/kg for the first two steps. The resulting operational characteristic curve approximates that for a single 50 kg sample. The sequential protocol was applied to the results for a set of 1293 lots of the 1998 crop year, each tested with a single 10 kg sample. Ninety-five percent of the lots would have been accepted on

the basis of the single test and 1.5% would have been rejected, whereas 3.5% of the lots would have required retesting.

KOMAROVA, N.V. 2000. **Determination of aflatoxin M<sub>1</sub> in milk using solid-phase extraction and high-performance liquid chromatography with fluorescence detection.** Journal of Analytical Chemistry **55**: 929–932.

Solid-phase extraction of AFM<sub>1</sub> from milk using Diapak S16M and Diapak S solid-phase extraction cartridges followed by HPLC determination with a Fluorot-02 fluorescence detector was studied.

## Aflatoxicoses

HENRY, S.H., BOSCH, F.X., TROXELL, T.C. and BOLGER, P.M. 1999. **Public health – Reducing liver cancer – Global control of aflatoxin.** Science **286(5449)**: 2453–2454.

This paper discusses the Joint FAO/WHO Expert Committee on Food Additives approach to the problem of reducing liver cancer incidence worldwide in terms of hepatitis B virus infection and aflatoxin levels in the diet.

WONG, N., LAI, P., PANG, E., FUNG, L.F., SHENG, Z., WONG, V., WANG, W.P., HAYASHI, Y., PERLMAN, E., YUNA, S., LAU, J.W.Y. and JOHNSON, P.J. 2000. **Genomic aberrations in human hepatocellular carcinomas of differing etiologies.** Clinical Cancer Research **6**: 4000–4009.

Comparative genomic hybridisation (CGH) was applied to the genome-wide chromosomal analysis in 83 hepatocellular carcinomas (HCC) tumour samples from four different geographic origins, Hong Kong, Shanghai, Japan and the USA. Cases from Hong Kong and Shanghai were all hepatitis B virus (HBV) related, those from Japan were hepatitis C virus (HCV) related and those from the USA were HBV negative. CGH revealed a complex pattern of chromosomal gains and losses, with the commonest aberration in each geographic location being chromosome 1q copy number gain. Shanghai cases displayed the highest number of total aberrations per sample, with significant copy losses on 4q, 8p and 16q. HCV related samples from Japan had a characteristically high incidence of 11q13 gain. p53 mutation was detected in 23% of Hong Kong cases, 40% of Shanghai, 31% of Japan, but only 6% of USA cases. The 'aflatoxin-associated' codon 249 mutation was, however, identified only in samples from China. This finding, together with the highly aberrant pattern of genetic changes detected in the Shanghai series, is suggestive of the

genotoxic effects of aflatoxin being more broadly based. It is also likely that there is a synergistic effect of HBV infection and high aflatoxin exposure in promoting hepatocellular carcinoma development. It appears from these studies that individual risk factors are indeed associated with distinct genetic aberrations, although changes in 1q gain appear common to all.

NWANKWO, J.O., TAHNTENG, J.G. and EMEROLE, G.O. 2000. **Inhibition of aflatoxin B<sub>1</sub> genotoxicity in human liver-derived HepG2 cells by kolaviron biflavonoids and molecular mechanisms of action.** European Journal of Cancer Prevention **9**: 351–361.

The possible chemopreventive potential of kolaviron in AFB<sub>1</sub> genotoxicity in HepG2 cells was studied. Kolaviron significantly and dose dependently inhibited the cytotoxicity (by 71.6%) and genotoxicity (47.1%) of AFB<sub>1</sub> in HepG2 cells. It induced about 3-fold increases in the messages for cytochrome P450 3A4 and glutathione S-transferases (GST) A1-1/ A2/2 and M1B, including a 2-fold increase in GST alpha protein.

BARTON, C.C., GANEY, P.E. and ROTH, R.A. 2000. **Lipopolysaccharide augments aflatoxin B<sub>1</sub>-induced liver injury through neutrophil-dependent and -independent mechanisms.** Toxicological Sciences **58**: 208–215.

The role of neutrophils (PMNs) in the pathogenesis of hepatic injury after AFB<sub>1</sub>/liposaccharide (LPS) cotreatment was investigated. Male rats were dosed ip with AFB<sub>1</sub> at 1 mg/kg, and 4 hr later with *Escherichia coli* LPS. Over a course of 6 to 96 hr after AFB<sub>1</sub> administration, rats were killed. LPS resulted in an increase in PMN accumulation in the liver that preceded the onset of liver injury. To assess if PMNs contributed to the pathogenesis, an anti-PMN antibody was administered to reduce PMN numbers in blood and liver, and injury was evaluated. Neutrophil depletion protected against hepatic parenchymal cell injury caused by AFB<sub>1</sub>/LPS cotreatment but not against markers of biliary tract injury. This suggests that LPS augments AFB<sub>1</sub> hepatotoxicity through two mechanisms: one of which is PMN dependent, and another that is not.

LIN, Y.L., HSU, J.D., CHOU, F.P., LEE, M.J., SHIOW, S.J. and WANG, C.J. 2000. **Suppressive effect of penta-acetyl geniposide on the development of gamma-glutamyl transpeptidase foci-induced by aflatoxin B<sub>1</sub> in rats.** Chemico-Biological Interactions **128**: 115–126.

The suppressive effects of penta-acetyl geniposide, (Ac)(5)-GP, on the hepatotoxic lesions induced by AFB<sub>1</sub> were investigated in male Wistar rats. Rats were given AFB<sub>1</sub> at 2 mg/kg body weight alone or with the addition of (Ac)(5)-GP at 2 mg/kg. Rats received treatments for 8 weeks, then were maintained on basal diet for 32 weeks. Although no differences in the total number of gamma -glutamyl transpeptidase (GGT)-positive foci was observed between the groups treated with AFB<sub>1</sub>, with or without (Ac)(5)-GP, treatment with (Ac)(5)-GP significantly reduced the number of AFB<sub>1</sub> induced GGT positive foci (with diameter larger than 0.3 mm). These results indicated that the protective effect of (Ac)(5)-GP on early hepatocarcinogenesis induced by AFB<sub>1</sub> was associated with the inhibition of GGT foci development.

SHEPHERD, A.G., MANSON, M.M., BALL, H.W.L. and MCLELLAN, L.I. 2000. **Regulation of rat glutamate-cysteine ligase (gamma-glutamylcysteine synthetase) subunits by chemopreventive agents and in aflatoxin B<sub>1</sub>-induced preneoplasia.** Carcinogenesis **21**: 1827–1834.

Dietary administration of the naturally occurring chemopreventive agents, ellagic acid, coumarin or alpha -angelicalactone caused an increase in glutamate cysteine ligase (GLCL) activity of between 3- and 5-fold in rat liver. The increases in GLCL activity were accompanied by increases (between 2- and 4-fold) in levels of both the catalytic heavy subunit (GLCLC) and regulatory light subunit (GLCLR). No substantial induction of GLCL was observed in rat kidney. The GST subunits A1, A3, A4, A5, P1 and M1 were all found to be inducible in rat liver by most of the agents. The expression of glutathione related enzymes was also examined in preneoplastic lesions induced in rat liver by AFB<sub>1</sub>. The majority of GGT-positive preneoplastic foci contained increased levels of GLCLC relative to the surrounding tissue. This was usually accompanied by an increase in GLCLR. Cells in the inner cortex of rat kidney were found to contain the highest levels of both GLCLC and GLCLR. The same cells showed the strongest staining for GGT activity.

CELIK, I., OGUZ, H., DEMET, O., BOYDAK, M., DONMEZ, H.H., SUR, E. and NIZAMLIOGLU, F. 2000. **Embryotoxicity assay of aflatoxin produced by *Aspergillus parasiticus* NRRL 2999.** British Poultry Science **41**: 401–409.

The embryotoxicity of mixed aflatoxins (AF) and AFB<sub>1</sub> were evaluated by a modified chick embryotoxicity screening test. AF consisted of 83.06% AFB<sub>1</sub>, 12.98% AFB<sub>2</sub>,

2.84% AFG<sub>1</sub> and 1.12% AFG<sub>2</sub>. AF and AFB<sub>1</sub> were injected into the eggs at doses of 10, 100 or 1000 ng/egg. AFB<sub>1</sub> at 10 ng/egg had a significantly greater embryotoxic effect than AF given at a similar dose. The higher doses of both AF and AFB<sub>1</sub> caused higher embryonic mortality and also an increase in early deaths. In the groups receiving AF and AFB<sub>1</sub> at 100 ng/egg an abnormal development was seen, with a protruded central region, corresponding to the area pellucida of the blastoderm.

CELIK, I., OGUZ, H., DEMET, O., DONMEZ, H.H., BOYDAK, M. and SUR, E. 2000. **Efficacy of polyvinylpyrrolidone in reducing the immunotoxicity of aflatoxin in growing broilers.** *British Poultry Science* **41**: 430–439.

One-day-old broiler chicks were fed mixed aflatoxins (AF) at 2.5 mg/kg diet with or without polyvinylpyrrolidone (PVPP) at 3 g/kg diet for 21 days. AF consisted of 83.06% AFB<sub>1</sub>, 12.98% AFB<sub>2</sub>, 2.84% AFG<sub>1</sub> and 1.12% AFG<sub>2</sub>. AF treatment significantly decreased peripheral T-lymphocyte counts and caused a slight decrease in splenic plasma cell counts. The addition of PVPP to an AF containing diet significantly increased T-lymphocyte

counts. Splenic plasma cell counts were numerically intermediate between control and AF groups.

OGUZ, H. and KURTOGLU, V. 2000. **Effect of clinoptilolite on performance of broiler chickens during experimental aflatoxicosis.** *British Poultry Science* **41**: 512–517.

Broiler chicks from 1 to 21 days of age were fed diets containing mixed aflatoxins (AF) at 25 mg/kg with and without the addition of clinoptilolite (CLI) at 15 or 25 g/kg. AF consisted of 76.40% AFB<sub>1</sub>, 16.12% AFB<sub>2</sub>, 6.01% AFG<sub>1</sub> and 1.47% AFG<sub>2</sub>. The addition of CLI at 15 g/kg to an AF containing diet significantly reduced the deleterious effects of AF on food consumption and body weight gain and slightly improved the food conversion ratio. CLI at 15 g/kg was more effective than at the 25 g/kg level against the adverse effects of AF on the variables investigated in this study.

OGUZ, H., KURTOGLU, V. and COSKUN, B. 2000. **Preventive efficacy of clinoptilolite in broilers during chronic aflatoxin (50 and 100 ppb) exposure.** *Research in Veterinary Science* **69**: 197–201.

One-day-old broiler chicks were fed total aflatoxins at 50 or 100 mg/kg with or without the addition of CLI at 15 g/kg diet for 42

days. Total aflatoxins at 100 mg/kg significantly decreased body weight gains of chicks. The addition of CLI to the 100 mg/kg aflatoxin containing diet moderately reduced the adverse effects of the aflatoxins on performances of chicks. Chicks consuming 50 mg/kg aflatoxin containing diet showed no significant differences on investigated parameters compared to controls.

IBRAHIM, I.K., SHAREEF, A.M. and AL-JOUBORY, K.M.T. 2000. **Ameliorative effects of sodium bentonite on phagocytosis and Newcastle disease antibody formation in broiler chickens during aflatoxicosis.** *Research in Veterinary Science* **69**: 119–122.

The effect of dietary sodium bentonite at 0.2, 0.4 and 0.6% wt/wt of feed on *in vitro* impaired phagocytosis and suppressed immune response to Newcastle disease vaccine during aflatoxicosis in broiler chicks was investigated. Both percentage and mean of phagocytic activities were decreased significantly in chicks fed aflatoxin at 2.5 mg/kg feed. The addition of sodium bentonite was significantly effective in ameliorating the effect of aflatoxin on the percentage and mean of phagocytosis. Sodium bentonite was also effective in ameliorating the suppressive effect of aflatoxin on the Hr-titre in chicks vaccinated against Newcastle disease.

# REVIEW\*

## “Mycotoxins and Public Health: A Review” by Curtis D. Malloy and John S. Marr, J. Public Health Management Practice, 1997, (3), 61–69.

Among the plethora of papers in the medical literature, relatively few deal with the role of mycotoxins and public health. Public health practitioners gave only transient attention to mycotoxins as potentially important causes of illness in the 1960s when they were discovered, and scant attention in the 1970s despite the detection of other mycotoxins in food.

The objectives of the authors in their review were to: review the historical and contemporary importance of mycotoxins; to identify mycotoxins that may be of significant public health importance; to advocate that public health professionals refocus concern on issues surrounding mycotoxicoses; and to provide appropriate references and citations to toxicologic and mycotoxic resources.

The word “mycotoxicosis” was proposed by Forjacs and Carll (1955) to describe animal illness as a consequence of ingestion of moldy fodder. Some fungi produce a single toxin, while others may produce many, and different fungal genera may produce the same mycotoxin. Many genera, including *Acremonium*, *Alternaria*, *Aspergillus*, *Claviceps*, *Fusarium*, and *Penicillium* produce mycotoxins. While the evidence in the literature indicates that ingestion of moldy feed and fodder may be the primary route leading to animal mycotoxicoses, airborne fungal spores and infested/infected plant particulates may also induce disease leading to death in animals and man.

Mallory and Marr list 8 categories of mycotoxin action in the review: immu-

nosuppression, myelosuppression, hepatotoxicity, nephrotoxicity, neurotoxicity, dermatotoxicity, estrogenicity, mutagenicity, teratogenicity, and carcinogenicity. Moreover, mycotoxin metabolites produced by some opportunistic fungi are known to further exacerbate disease in immunocompromised hosts.

Commodities known to contain mycotoxins include cereals and grains and products thereof, seeds and nuts, fruits and vegetables and products thereof, milk and dairy products, spices and condiments, meat and meat products, and others, including prepared baby foods. Clearly, mycotoxins are common in most food supplies. Diagnostic tests (ELISA, RIA) capable of detecting 0.1 ng/mL have been developed for specific mycotoxins, and several commercial test kits are available.

Several mycotoxicoses of animals (e.g. equine leukoencephalomalacia [ELEM] in horses, porcine pulmonary edema [PPE] in swine, and stachybotryotoxicosis (in poultry, cattle, sheep, swine and sheep) are listed, and their principal pathologies and distribution are described. Human mycotoxicoses including aflatoxicosis (*Aspergillus* spp.) and ergotism (*Claviceps* spp.) are well known. At the time of publication (1997) several aflatoxins (B<sub>1</sub>, G<sub>1</sub>, and

M<sub>1</sub>) were known to be carcinogenic in animals, but only B<sub>1</sub> was carcinogenic in humans.

The review contains one chart (commodities shown to contain mycotoxins). There are 4 figures: 2 describe animal mycotoxicoses and human mycotoxicoses, their pathogens, pathologies, and distribution in the world. A third describes an historical account of the occurrence of mycotoxins, and the fourth provides evidence of carcinogenesis in humans and animals.

There are 2 [other] figures. The first illustrates the distribution of major mycotoxins (aflatoxins, deoxynivalenol, ergot, ochratoxin A, tremorgens, and zearalenone) in field crops in the United States, and the second illustrates structural formulae for aflatoxin M<sub>1</sub>, patulin, and tricothecene. 49 references are provided.

The previous review of mycotoxicology was in 1977. Malloy and Marr in their review have provided an assessment of the field of mycotoxicosis as of 1997 and, in doing so, have provided a basis for continued research leading to a better understanding of the role of mycotoxins in public health. The authors conclude their review by stating that “It may be time to work with allied fields to bring mycotoxins into the mainstream of public health.”

## AUSTRALIAN MYCOTOXIN NEWSLETTER

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\* The following review by R. Hamilton was posted on the ProMED-mail web site <<http://www.promedmail.org>> of the International Society for Infectious Diseases on 30 November 2000.