

## REVIEW

# Fungal Volatiles as Indicators of Food and Feeds Spoilage

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Schnürer, J., Olsson, J., and Börjesson, T. 1999. Fungal volatiles as indicators of food and feeds spoilage. *Fungal Genetics and Biology* 27, 209–217. Fungal growth leads to spoilage of food and animal feeds and to formation of mycotoxins and potentially allergenic spores. Fungi produce volatile compounds, during both primary and secondary metabolism, which can be used for detection and identification. Fungal volatiles from mainly *Aspergillus*, *Fusarium*, and *Penicillium* have been characterized with gas chromatography, mass spectrometry, and sensory analysis. Common volatiles are 2-methyl-1-propanol, 3-methyl-1-butanol, 1-octen-3-ol, 3-octanone, 3-methylfuran, ethyl acetate, and the malodorous 2-methyl-isoborneol and geosmin. Volatile sesquiterpenes can be used for taxonomic classification and species identification in *Penicillium*, as well as to indicate mycotoxin formation in *Fusarium* and *Aspergillus*. Developments in sensor technology have led to the construction of “electronic noses” (volatile compound mappers). Exposure of different nonspecific sensors to volatile compounds produces characteristic electrical signals. These are collected by a computer and processed by multivariate statistical methods or in an artificial neural network (ANN). Such systems can grade cereal grain with regard to presence of molds as efficiently as sensory

panels evaluating grain odor. Volatile compound mapping can also be used to predict levels of ergosterol and fungal colony-forming units in grain. Further developments should make it possible to detect individual fungal species as well as the degree of mycotoxin contamination of food and animal feeds. © 1999 Academic Press

**Index Descriptors:** fungal biomass; ergosterol; colony-forming units; adsorbing polymers; fusel alcohols; dimethyl disulfide; 1-methoxy-3-methylbenzene; sorbate; *trans*-1,3-pentadiene; 2,4,6-trichloroanisole.

Fungal growth in food and animal feed leads to substantial damage, manifested as general spoilage, nutritional losses, formation of mycotoxins, and potentially allergenic spores (Filtenborg *et al.*, 1996). Methods able to detect and identify fungi with different levels of specificity are needed to evaluate the mycological quality of food and feeds. Techniques are also needed which can provide quantitative data on fungal mass and activity. Assays based on DNA sequences are the most specific and are currently being developed also to provide quantitative data, e.g., competitive PCR for detection of *Fusarium* species in plant material (Nicholson *et al.*, 1998). The main emphasis of this review is on the use of fungal volatiles to detect the presence of fungi in food and feeds. However, as new assays always have to be compared with “basal” methods, the quantification of colony-forming units (CFU), hyphal lengths (volume), or ergosterol will also be briefly covered.

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## COLONY-FORMING UNITS

The use of selective substrates for various ecological groups can increase the precision of CFU determinations. In food mycology substantial progress has been made in the development of selective and indicative growth media (Samson *et al.*, 1995). Xerotolerant fungi can be detected on dichloran glycerol 18% agar (DG18) with a water activity of 0.95 or on malt extract yeast extract 50% glucose agar (MY 50G) with a water activity of 0.89. "Acido tolerant" fungi, e.g., *Penicillium roqueforti* and preservative resistant yeasts, are among the few fungi able to grow on malt extract agar with 0.5–1.0% acetic acid. Color changes of the colony reverse (backside of agar plate), based on reactions between substrate components and secondary metabolites, indicate the presence of certain mycotoxigenic fungi, e.g., *Aspergillus flavus* on *Aspergillus flavus* and *parasiticus* agar (AFPA) and *Penicillium verrucosum* on dichloran rose bengal yeast extract sucrose agar (DRYES). However, it has to be kept in mind that the results of fungal CFU determinations are highly influenced by the degree of fungal sporulation (Schnürer, 1993).

## HYPHAL AND SPORE VOLUME

Direct microscopy for hyphal length (and spore number) determinations has been commonly used in soil microbiology as well as in studies of litter decomposition. This provides a determination of fungal volume which can be converted to fungal (bio)mass.

In combination with staining techniques, e.g., immunofluorescence, reliable determinations of fungal biomass can be obtained even for difficult samples. The technique is seldom used in food mycology since only limited numbers of samples can be handled (<20 per day). Newell (1992) has written an excellent review on advantages and problems of using this and other methods for estimating fungal biomass in plant material.

## ERGOSTEROL

Fungal-specific marker substances, such as ergosterol and chitin, offer a third possibility for quantification of fungi. Determination of ergosterol, originally proposed by

Seitz *et al.* (1977) for determining the degree of fungal infestation of grains, has become increasingly used with a variety of samples. Ergosterol is a dominant membrane sterol in all eumycota fungi except chytrids, rusts, and some yeasts, but is absent or very rare in oomycetes and hyphochytridiomycetes (Newell, 1992). It is not found in bacteria, plants, and animals, but has been found in certain algae and protozoa (Newell, 1992). The presence of double bonds at positions 5 and 7 leads to a highly specific ultraviolet absorption spectrum with a maximum at 282 nm, making possible a quantification after separation with high-pressure liquid chromatography (Seitz *et al.*, 1977, 1979; Newell *et al.*, 1988). Fungal ergosterol contents normally range between 0.2 and 0.6% of dry weight, but the content can vary more widely depending on mycelial age and growth conditions (Newell, 1992; Schnürer, 1993). The ergosterol assay has now become a commonly used method for the quantification of fungal mass in food and feeds, especially in grains (Schwadorf and Müller, 1989). However, it has to be remembered that the assay is nonspecific and does not provide any information on species composition.

## FUNGAL VOLATILES

The most commonly used method to detect fungal growth in food and feeds is sensory analysis, i.e., the human nose. Sensory analysis is used to detect mold and other objectionable odors of grain in all international and in most national trade (Smith *et al.*, 1994; Börjesson *et al.*, 1996). In the Swedish cereals trade, grain odors are described as either normal, musty, moldy, acid, sour, burnt, or foreign, and the intensities are given as weak, pronounced, or strong. The odor analysis is most commonly performed by a single employee at a laboratory or a grain reception station, and only seldomly by a trained sensory panel. This procedure for evaluating mycological quality, although fast and sensitive, has a number of drawbacks. Most important among these is that the method is subjective (Dickinson *et al.*, 1998). The inhalation of fungal spores from moldy grain is hazardous (Rylander, 1986) and recent investigations also suggest that fungal volatiles can cause damage to human respiratory organs (Larsen *et al.*, 1998; Walinder *et al.*, 1998). There is thus a need to develop alternative detection methods for fungal volatiles that are efficient, not too expensive, and preferably as fast as human sensory analysis.

## Collection of Volatiles

Volatiles can be analyzed using different collection methods. The simplest is to take samples with a gas-tight syringe directly from the headspace gas above a sample and then to inject the sample into a gas chromatograph. Alternatively, volatiles can be collected on various porous polymers (e.g., Tenax GC or Chromosorb 102) and released either through extraction with organic solvents or through thermal desorption (Börjesson, 1993; Larsen, 1994). The volatiles can be adsorbed using diffusive sampling from the headspace (Larsen and Frisvad, 1994) or active purging and trapping of headspace gases (Börjesson *et al.*, 1990, 1992). Cereal grains can also act as adsorbing polymers, allowing for determinations of both past and present fungal growth (Börjesson *et al.*, 1994). Nilsson *et al.*, (1996) described an elegant headspace solid-phase microextraction (HS-SPME) method, using collection of fungal volatiles on polyacrylate and polydimethyl-siloxane-coated microfibers, for direct injection into a thermal desorption unit on the gas chromatograph. Results obtained from collecting volatile organic compounds from the genus *Penicillium* on HS-SPME were similar to those obtained by Tenax adsorption (Nilsson *et al.*, 1996).

Alternatively, volatiles can be extracted from various materials using steam distillation, organic solvents, or supercritical fluid carbon dioxide (Nielsen *et al.*, 1991; Börjesson *et al.*, 1994). Extraction techniques that rely on heat and/or organic solvents can cause degradation of sample volatiles, while the supercritical fluid extraction technique is considered a nondestructive method (Charpentier, 1986; Larsen and Frisvad, 1995a).

## Chemical Characterization

Separation of volatiles is primarily carried out using gas chromatography (GC) or gas-liquid chromatography. The gas chromatograph is very often combined with a mass spectrometer (GC-MS) for separation and identification of compounds. Through mass spectroscopy the molecular mass and typical fragmentation pattern of an unknown fungal volatile can be obtained and compared with reference libraries (Börjesson, 1993; Larsen, 1994). Infrared spectroscopy using Fourier transform methods can also be combined with a gas chromatograph (GC-FTIR), and due to its ability to differentiate between isomers, it can complement GC-MS (Larsen, 1994). To detect odorous fungal metabolites the gas chromatographic separation of volatiles can be combined with sensory analysis of indi-

vidual peaks, using a split gas-stream GC-technique (Börjesson *et al.*, 1993).

## Volatile Fungal Metabolites

Volatile fungal metabolites can be formed during both primary and secondary metabolism from a wide variety of starting compounds, e.g., acetate, amino acids, fatty acids, and keto acids (Fig. 1; Kinderlerer, 1989; Börjesson, 1993; Larsen, 1994; Jelen and Wasowicz, 1998). Fungal volatiles can indicate spoilage but are also important as flavor compounds of many fermented foods (Kinderlerer, 1989; Janssens *et al.*, 1992). Some of the most commonly reported volatiles from filamentous fungi are listed together with examples of producing species in Table 1. Generally, there have been fewer studies concerning volatiles from spoilage yeast than for filamentous fungi (Whitfield, 1998). However, *Saccharomyces cerevisiae*, one of the 10 most commonly isolated food contaminating yeasts (Pitt and Hocking, 1997), form "fusel" alcohols during branched chain amino acid catabolism (Webb and Ingraham, 1963). Fusel alcohols are the alcohols with higher boiling points than ethanol that remain after distillation of a yeast fermentation. Among these are 3-methyl-1-butanol formed from leucine and 2-methyl-1-propanol from valine catabolism (Berry, 1988). The osmotolerant *Hansenula anomala* (*Pichia anomala*), frequently associated with spoilage of cakes and pastry, is further known to produce large quantities of malodorous ethyl acetate (Lanciotti *et al.*, 1998, Whitfield, 1998).

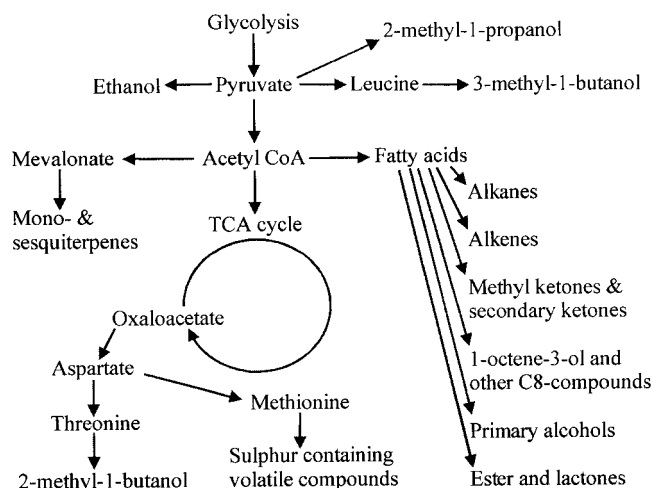










FIG. 1. Overview of metabolic pathways for biosynthesis of the main fungal volatile metabolites (Börjesson, 1993; Larsen, 1994).

**TABLE 1**  
Examples of Common Volatile Metabolites from Food- and Feed-borne Spoilage Fungi

Class	Compound	Structure	Producing fungus	Reference
Alcohols	2-Methyl-1-propanol		<i>Geotrichum candidum</i> <i>Penicillium roqueforti</i>	(1) (2)
	3-Methyl-butanol		<i>Fusarium graminearum</i> <i>Penicillium aurantiogriseum</i>	(3) (4)
	1-Octene-3-ol		<i>Penicillium glabrum</i> <i>Penicillium verrucosum</i>	(2) (5)
Ketones	3-Octanone		<i>Fusarium sporotrichoides</i> <i>Penicillium commune</i>	(5) (4)
	Esters	Ethyl acetate		<i>Penicillium digitatum</i> <i>Pichia anomala</i>
Furans	3-Methyl furan		<i>Aspergillus flavus</i> <i>Penicillium brevicompactum</i>	(2) (2)
Monoterpenes	2-Methyl-isoborneol		<i>Aspergillus niger</i>	(7)
			<i>Penicillium solitum</i>	(8)
Sesquiterpenes	Geosmin		<i>Penicillium discolor</i> <i>Penicillium expansum</i>	(4) (9)

*Note.* A more complete tabulation of fungi and volatile compounds is given by Jelen and Wasowicz (1998).

References: (1) Jacobsen and Hinrichsen (1997); (2) Börjesson *et al.* (1992); (3) Jelen *et al.* (1997a); (4) Larsen and Frisvad (1995b); (5) Pasanen *et al.* 1996; (6) Lanciotti *et al.* (1998); (7) Börjesson *et al.* (1993); (8) Hocking *et al.* (1998); (9) Matheis and Roberts (1992).

Börjesson *et al.* (1993) identified volatiles from 10 *Aspergillus* and *Penicillium* species, 4 with a unpleasant musty odor, and 6 reference fungi without any characteristic colony odor when cultivated on oatmeal agar. Volatile metabolites were collected on an adsorbent and analyzed with GC/MS and a combined GC and sensory analysis. Multivariate analysis of GC/MS and fungal odor data revealed strong associations between 6 of the 65 volatile compounds and colony odors. The sensory analysis showed that 5 of these, dimethyl disulfide, 1-octen-3-ol, 2-methyl-isoborneol, and two C<sub>11</sub>H<sub>18</sub> compounds, had strong musty odors. Geosmin, 1-methoxy-3-methylbenzene, and methylphenol were also produced in large amounts by some malodorous fungi and contributed to their unpleasant odor. 3-Methylfuran, 2-methyl-1-propanol, and 3-methyl-1-butanol were much more commonly produced than the malodorous compounds. Both odorous and other volatile metabolites could be detected after 2 days of fungal growth. The production of odorous metabolites was enhanced at the time of sporulation.

Sunesson *et al.* (1995) analyzed the production of

volatiles from *Aspergillus versicolor*, *Penicillium commune*, *Cladosporium cladosporioides*, *Paecilomyces variotii*, and *Phialophora fastigiata*, cultivated on malt extract agar and dichloran glycerol agar. The most commonly produced substances were 2-methyl-1-propanol, 2-methyl-1-butanol, 3-methyl-1-butanol, 3-methylfuran, and dimethyl disulfide. The production was highly dependent on both medium and species.

Fungi can degrade triacylglycerols to free fatty acids to provide a carbon and energy source. Degradation of lipids found in coconut and palm kernel oils and butter leads to formation of medium chain (C6-C12) fatty acids, which can inhibit fungal growth (Kinderlerer, 1993). A number of fungal bioconversions of medium chain fatty acids can occur, leading to the production of volatile methyl ketones and secondary alcohols. The important food spoilage fungi *Aspergillus*, *Penicillium*, *Cladosporium*, and *Fusarium* have all been shown to produce volatile methyl ketones and secondary alcohols from medium chain fatty acids. For example, Hocking *et al.* (1998) detected ketones ranging from heptan-2-one to the major compound undecan-2-one when *Penicillium solitum*, isolated from moldy margarine,



was grown on margarine agar. Naturally off-flavored margarine contained  $\mu\text{g g}^{-1}$  quantities of volatile methyl ketones (mainly heptan-2-one) that were absent in control margarine. Kinderlerer (1993) has suggested that the production of these volatile methyl ketones is a fungal strategy to eliminate toxic metabolites.

In a large comparative study, 196 volatile metabolites from 47 different *Penicillium* species were characterized by GC-MS and GC-FTIR (Larsen and Frisvad, 1995b). About 70 metabolites were sesquiterpene hydrocarbons ( $\text{C}_{15}\text{H}_{24}$ ), such as  $\beta$ -caryophyllene,  $\alpha$ -bergamotene, and thujopsene. Different monoterpenes, alcohols, esters, ketones, alkenes, and a few aromatic compounds were also produced. Both geosmin and 2-methyl-isoborneol, with moldy/earthy off-flavors, were produced by a number of different species. More than half of the metabolites were detected from only one species and nearly all species produced a unique profile of volatile metabolites. This suggests that multivariate data analysis of volatile profiles should be possible to use for identification of fungi to species level. In a follow-up study, analysis of volatile metabolite production from 132 isolates of 25 different terverticillate *Penicillium* was used for species classification (Larsen and Frisvad, 1995c). Fungal volatiles were collected from Petri dishes by diffusive sampling onto the adsorbent polymer Tenax TA and analyzed by GC-MS. The relative amounts of the 131 detected volatiles collected from each of the 132 isolates were analyzed by average linking clustering (UPGMA). A clear separation was obtained for practically all species investigated. The results agreed with previous classifications of *Penicillium*, based on chemotaxonomy of biosynthetic families of nonvolatile secondary metabolites. Indeed, even isolates of *Penicillium* described as varieties, e.g., *P. roqueforti* var. *carneum* and *Penicillium roqueforti* var. *roqueforti*, clustered independently from each other. Geosmin was produced by *P. roqueforti* var. *carneum*, but not by *P. roqueforti*. Later Boysen *et al.* (1996), comparing DNA sequence data from the ITS region with data on morphological characters and nonvolatile secondary metabolites, confirmed that variety *carneum* is indeed a new species, *Penicillium carneum*, separate from *Penicillium roqueforti*.

Species-characteristic volatile terpenes from the cheese-production fungus *Penicillium roqueforti* and the cheese contaminant *P. commune* were collected by headspace solid-phase microextraction and characterized (Larsen, 1997). Selected ion monitoring of four to seven of the most characteristic ions (mainly sesquiterpenes) made it possible to identify the fungi to species level. In a mixed

culture of *P. roqueforti* and *P. commune* on yeast extract sucrose agar, inoculated at a ratio of 1000:1, specific volatiles from both fungi could be detected within 3 days, even though morphological differences were not discernible on the agar plates (Larsen, 1997). This clearly shows that volatile metabolite profiles can be used to identify fungi in mixed communities, at least *in vitro* and using selected ion monitoring mass spectroscopy.

A complete tabulation of volatile fungal metabolites from *Aspergillus*, *Fusarium*, and *Penicillium* species and their relation to spoilage of agricultural commodities can be found in the review by Jelen and Wasowicz (1998).

### *Mycotoxins and Volatiles*

It is of particular interest to investigate whether fungal volatiles can be used to detect mycotoxin formation. Zeringue *et al.* (1993) found that headspace volatiles differed between aflatoxigenic strains and nonaflatoxigenic strains of *Aspergillus flavus* grown in liquid culture. Aflatoxigenic strains of *A. flavus* produced several  $\text{C}_{15}\text{H}_{24}$  compounds (e.g.,  $\alpha$ -gurjunene, *trans*-caryophyllene, and cadinene) that were not detected in the emissions of nonaflatoxigenic strains of *A. flavus*. The production of  $\text{C}_{15}\text{H}_{24}$  volatile compounds correlated with initiation of aflatoxin biosynthesis, and the disappearance of the  $\text{C}_{15}\text{H}_{24}$  compounds coincided with the decline of aflatoxin synthesis (Zeringue *et al.*, 1993).

The data of Pasanen *et al.* (1996) indicate that the production of volatile terpenes relates to the formation of trichothecene mycotoxins when comparing growth of *Fusarium sporotrichoides* on cereal grains and straw. The volatile terpene trichodiene is the first metabolite in the trichothecene biosynthesis pathway (Desjardins *et al.*, 1993). A clear correlation between synthesis of trichothecenes and production of trichodiene and volatile sesquiterpenes from *Fusarium sambucinum*, *F. sporotrichoides*, *F. poae*, and *F. graminearum* was observed by Jelen *et al.* (1995, 1997a). This was evident both for inoculated wheat kernels (Jelen *et al.*, 1995, 1997a) and in incubated grain spikes with natural *Fusarium* head blight infestation (Jelen *et al.*, 1997b).

### *Fungal Volatiles from Preservatives*

The preservative sorbate is used to prevent fungal growth in a variety of foods. Several *Penicillium* species, among them *P. roqueforti*, are able to degrade sorbate to *trans*-1,3-pentadiene which has an unpleasant kerosene-

like odor (Marth *et al.*, 1966; Liewen and Marth, 1985). Fungal conversion of the wood preservative chlorophenol to 2,4,6-trichloroanisole gave a musty taint to fruits transported in freight containers made from impregnated wood (Hill *et al.*, 1995). Likewise, coffee beans can be afflicted with Rio-taint, a musty, phenolic odor caused by 2,4,6-trichloroanisole and correlated with high mold contamination of green coffee beans (Liardon *et al.*, 1989; Spadone *et al.*, 1990).

## ELECTRONIC NOSES (VOLATILE COMPOUND MAPPERS)

Recent developments of electronic sensors that respond to volatiles suggest new ways of detecting fungal growth in food and feeds (Stetter *et al.*, 1993; Dickinson *et al.*, 1998). Electronic noses, i.e., volatile compound mappers, consist of a sensor array with different types of nonspecific sensors. Exposure to volatile compounds gives rise to various kinds of electrical responses from the sensors, such as voltage shifts, changes in resistance, or changes in conductivity. The sensors can have different profiles of sensitivity and it is advantageous to include sensors with different sensing principles in the electronic noses. The discriminating ability of a sensor array is increased if the sensors show a varied response to different volatile compounds. The signal pattern from the sensors is collected by a computer and further evaluated with multivariate analysis or processed by an artificial neural network (ANN), providing a pattern recognition system. In a recent review of "artificial nose" technology Dickinson *et al.* (1998) points to the large similarities between the electronic nose and the biological olfactory system and between the ANN and the odor signal processing and recognition system occurring in the human brain.

We have used electronic nose analyses with grain samples that are heated in a chamber, from which desorbed volatiles are led over a sensor array. An ANN, trained with sensor data from samples with known odor status, could predict the odor classes of a validation set of good, moldy, weakly, and strongly musty oat samples (Jonsson *et al.*, 1997). The ANN also indicated the percentage of moldy barley or rye grains in mixtures with fresh grains. In remoistened incubated wheat, a high degree of correlation between ANN predictions and measured ergosterol, as well as with fungal and bacterial CFU, was observed (Jonsson *et al.*, 1997). When using a two-class system (good or bad smell), assigned by a sensory panel,

Börjesson *et al.* (1996) found that an electronic nose correctly classified 90% of authentic oat, wheat, and barley samples. In addition to being able to detect general fungal presence when evaluating the mycological quality of food and feeds, it is also important to be able to identify specific fungi in natural samples. The seed-borne plant pathogen common bunt (*Tilletia caries*) produces a striking fish-like odor, mainly caused by trimethylamine, which reduces the organoleptic quality of cereal end-products such as bread and breakfast cereals (Börjesson and Johnsson, 1998). We have found that a volatile compound mapper can be equally effective as a human sensory panel in separating wheat samples infested with *Tilletia caries* from noninfested wheat (Börjesson and Johnsson, 1998). Work by Keshri *et al.* (1998) further shows that it is possible to differentiate between *Wallemia sebi* and other xerophilic spoilage fungi (*Eurotium species*) using an electronic nose.

We have used electronic nose data together with measured fungal CFU levels and ergosterol contents for *Penicillium roqueforti* inoculated wheat grain to build a cross-validated partial least squares (PLS) regression model. The PLS model could predict grain contents of both fungal CFU and ergosterol with a high degree of precision (Fig. 2; Fig. 3). We have also investigated authentic barley samples with various odor status for their volatile metabolites. The grain is heated to 50°C to desorb fungal volatiles in a specifically designed glass vessel which is purged with air. Released volatile compounds are collected on the adsorbent Chromosorb 102 and analyzed with GC/MS. GC/MS data indicated that grain samples with a perceived moldy/musty odor had larger volatile concentrations of 2-methylpropanol, 3-methylbutanol, 3-octanone, and 1-octen-3-ol than sound grain (Olsson *et al.*, unpublished). Since these are common fungal volatiles, it is reasonable to assume that the degree of fungal infestation (CFU or ergosterol) should be possible to predict with an electronic nose also with authentic, i.e., noninoculated, grain samples.

## CONCLUDING REMARKS

Careful analytical work has identified chemical and sensory characteristics for volatile metabolites from many different fungi relevant to food and feed spoilage. Fungal species can produce a multitude of volatiles, some of which are common to many fungi, and others that seem to be unique for one species, e.g., many sesquiterpenes. By using proper analytical techniques and data processing methods, monitoring of fungal volatile metabolites can be used both

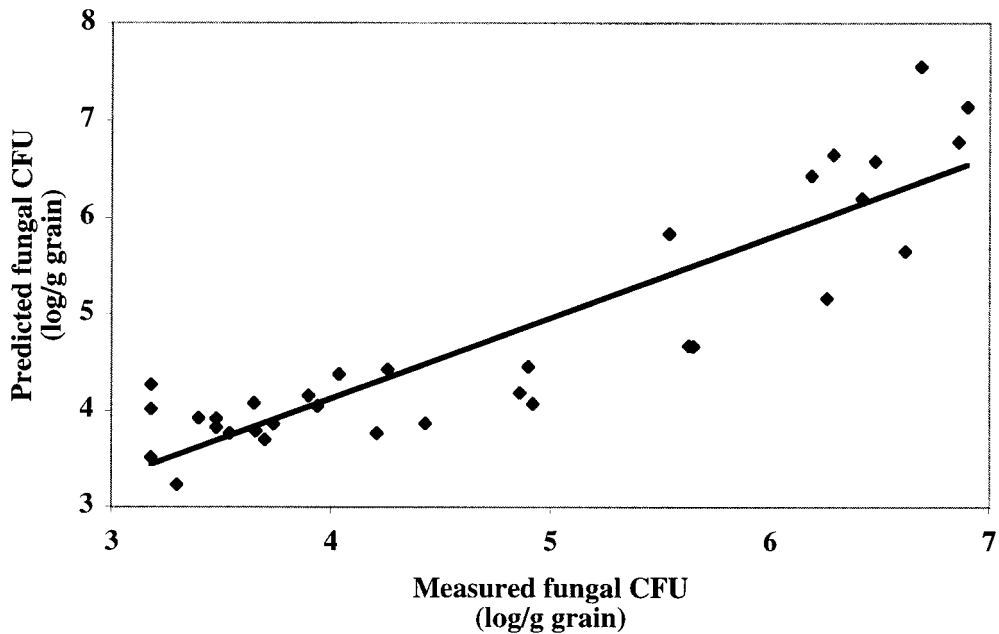


FIG. 2. Prediction of grain fungal CFU from volatiles detected with the electronic nose. Wheat kernels ( $a_w$  0.94) were inoculated with  $10^3$  spores of *Penicillium roqueforti* per gram and incubated at 2, 4, 10, 15, 20, and 25°C in six replicate flasks for 7 days, and CFU values were determined on DG18 agar. A cross-validated partial least square regression model, based on sensor responses and measured fungal CFU values, gave a high degree of correlation between measured and predicted CFU values ( $R^2 = 0.82$ ,  $n = 33$ ).

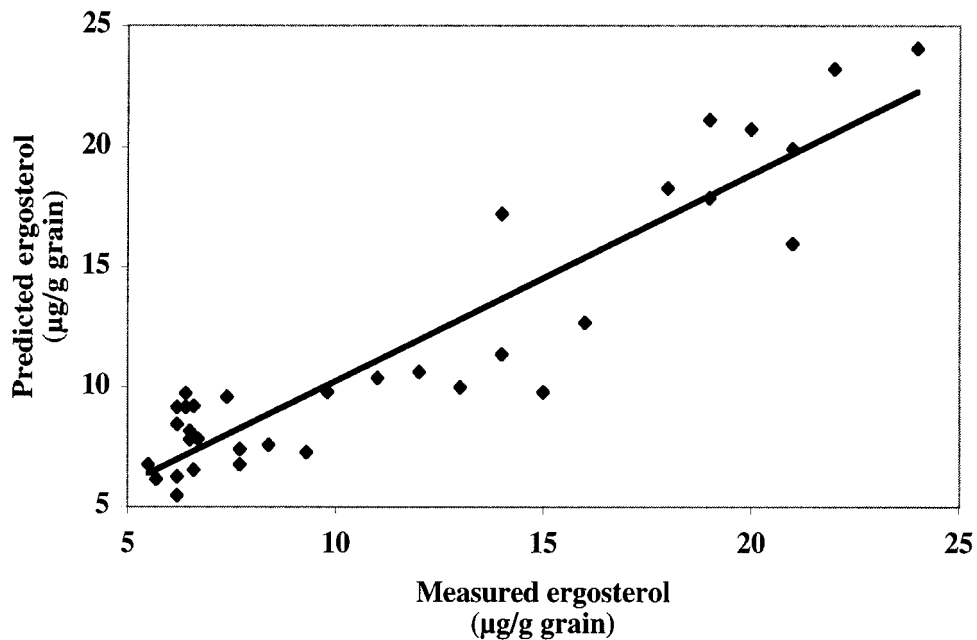


FIG. 3. Prediction of grain ergosterol content from fungal volatiles detected with the electronic nose. Wheat kernels ( $a_w$  0.94) were inoculated with  $10^3$  spores of *Penicillium roqueforti* per gram and incubated at 2, 4, 10, 15, 20, and 25°C in six replicate flasks for 7 days, and ergosterol levels were determined. A cross-validated partial least square regression model, based on sensor responses and measured ergosterol contents, gave a high degree of correlation between measured and predicted ergosterol values ( $R^2 = 0.86$ ,  $n = 34$ ).

to detect general fungal infestation and to indicate specific fungi. Placing polymer cartridges that adsorb fungal volatiles in storage environments and analyzing at regular intervals should provide an opportunity for early warning/detection of spoilage activity in food and feeds. The ability of starch and cellulose in grains to accumulate volatile metabolites, i.e., act as natural adsorbents, offers a possibility of tracking past fungal growth in cereal-based food and feed. Although applications of electronic noses until now have been slow to appear in the literature, this is rapidly changing. The present fast development of sensor technology methods for the detection of volatiles, as well as data processing techniques able to treat large data sets of volatile profiles, will certainly improve the possibility for species-specific detection of spoilage and mycotoxin-forming fungi in food and feeds.

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