2,4,6-Tribromoanisole: a Potential Cause of Mustiness in Packaged Food

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Strains of the ubiquitous xerophilic fungus *Paecilomyces variotii* can quantitatively convert the fungicide 2,4,6-tribromophenol to its musty-smelling metabolite, 2,4,6-tribromoanisole. Conversion was complete at the end of 6 weeks. A trained sensory panel found the odor threshold concentrations of 2,4,6-tribromoanisole to be 2×10^{-5} μg/L in water and 2×10^{-3} μg/kg in sultanas. Studies also showed that 2,4,6-tribromoanisole could produce a musty taint in sultanas packaged in polyethylene pouches after only 1 week of storage in the presence of fiberboard that contained 300 ng of this compound; the taint was produced at both 22 and 30 °C. An analytical method based on multiple-ion detection GC−MS was used for the measurement of 2,4,6-tribromoanisole in packaging materials and food.

Keywords: *2,4,6-Tribromoanisole; 2,4,6-tribromophenol; fungal metabolite; musty odor; odor threshold; GC−MS analysis*

INTRODUCTION

2,4,6-Tribromoanisole (TBA) has been identified as a trace environmental contaminant in marine fauna and sediments (Miyazaki et al., 1981; Watanabe et al., 1983), in topsoil and earthworms (Knuutinen et al., 1990), and in the atmosphere (Wittlinger and Ballschmiter, 1990). In these materials, it is believed that 2,4,6-tribromophenol (TBP) is its precursor and that bacteria are responsible for the *O*-methylation of the parent phenol (Allard et al., 1987). In nature, TBP has been identified in marine algae (Whitfield et al., 1992a), sponges and bryozoans (Whitfield et al., 1992b), benthic organisms (Sheikh and Djerassi, 1975; Weber and Ernst, 1978), and higher marine animals such as crustaceans (Whitfield et al., 1988) and fish (Boyle et al., 1992; Whitfield et al., 1995). TBP is also readily formed from the chlorination of water containing phenol and bromide (Sweetman and Simmons, 1979). Industrially, derivatives of synthetic tribromophenols, and in particular those of TBP, have been widely recommended as fireproofing agents in resin laminates (Takahashi and Sato, 1992) and in organic polymers destined for use in electronic equipment (Nishibori and Kondo, 1993). They are also used as wood preservatives (Towa Mokuzai Co. Ltd, 1982) and as general fungicides for use with leather, textiles, paint, plastics, paper, and pulp (Pulido and Ayzaguer, 1991).

During a recent survey of the occurrence of bromophenols in Australia, it was found that some samples of corrugated fiberboard, manufactured from imported medium and linerboard, were contaminated with high concentrations of TBP. As some of this material had been used for the packaging of nonhermetically sealed food, we were concerned that this compound could be the source of a new chemical taint in packaged foods. It was possible that TBP would be converted to TBA by analogy with the conversion of 2,4,6-trichlorophenol to its musty-smelling metabolite, 2,4,6-trichloroanisole (TCA), by fungi present in fiberboard (Whitfield et al., 1985, 1991). Anecdotal information received by this laboratory indicated that TBA possessed a musty odor that could be detected at low concentrations. As a consequence, the current study was undertaken to establish (a) whether TBP can be converted to TBA by fungi present in fiberboard, (b) the odor threshold concentration of TBA in water and a food, and (c) whether TBA present in packaging materials can contaminate nonhermetically sealed foods in a high enough concentration to produce a chemical taint.

The ubiquitous fungus, *Paecilomyces variotii*, a known methylator of TCP, was chosen for the metabolic studies. This species is one of some 17 that have been isolated from both fiberboard cartons and timber floors of freight containers and that are capable of methylating TCP to TCA (Tindale et al., 1989). Many species of fungi other than *P. variotii* could likewise have the capacity to metabolize TBP to TBA.

EXPERIMENTAL PROCEDURES

Materials. 2,4,6-Tribromoanisole and 2,4,6-tribromophenol were purchased from Aldrich Chemical Co. Inc., Milwaukee, WI, and 2,6-dibromophenol-d_3 from C/D/N Isotopes Inc., Quebec, Canada. The purity of each compound (>99%) was confirmed by analysis using gas chromatography−mass spectrometry (GC−MS). 3,5-Dimethyl-2,4,6-trichloroanisole was synthesized in this laboratory. Export grade sultanas were supplied by Angas Park Co., Angaston, South Australia, and virgin kraft fiberboard was supplied by Mildura Co-operative, Mildura, Victoria. Analyses of these materials showed that the sultanas were free from TBA and TCA and that the fiberboard was free from TBA and TBP. The dried fruit was stated to contain about 0.3% hydrogenated vegetable triglycerides added during processing as a drying oil. Distilled water was purified through a Milli-Q Purification System (Millipore Corporation, Bedford, MA). All inorganic chemicals and organic solvents were reagent grade. Extruded low-density polyethylene film (50 μm) was obtained from a local supplier. Fungi were obtained from the CSIRO Food Research Collection; the strains of *P. variotii* had previously been isolated from fiberboard containing mg/kg concentrations of a mixture of chlorophenols.

Isolation of 2,4,6-Tribromoanisole and 2,4,6-Tribromophenol. For the analysis of TBA, samples of sultanas (25

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g) and fiberboard (12, 18, or 25 g) in distilled water (1.5 L) were extracted by combined steam distillation solvent extraction (SDE) with 30 mL of pentane/ether (9:1) as solvent using a method previously described for the analysis of chloroanisoles in food and packaging materials (Whitfield et al., 1986). The same extraction procedure was also used for the analysis of TCA in sultanas. Similarly, for the analysis of TBP, samples of fiberboard (25 g) in distilled water (1.5 L) were extracted by SDE using the method of Whitfield et al. (1988). After extraction, a measured quantity of internal standard was added to the 30 mL extracts (100 ng in 100 μL of iso-octane). 3,5-Dimethyl-2,4,6-trichloroanisole was used as internal standard for the TBA analyses, and 2,6-dibromophenol-d_3 was used for the TBP analyses. The extracts were then concentrated by the careful removal of the pentane/ether by fractional distillation, and the concentrates in iso-octane (about 100 μL) were stored at −15 °C until required for analysis by GC−MS.

Analysis by GC−MS. A Hewlett-Packard HP5890 II gas chromatograph interfaced to a Hewlett-Packard HP5971A mass selective detector, operated in the multiple ion detection (MID) mode, was used for the analysis of TBA and TBP in all extracts. The GC was fitted with a 25 m long × 0.25 mm i.d. fused silica column coated with HP5 (0.33 μm film thickness) and a retention gap 5 m long × 0.25 mm i.d. uncoated but deactivated. Aliquots (1 μL) of the sample extracts or calibration solutions were injected automatically using a Hewlett-Packard HP 7673 autosampler. For the TBA analyses the injections were splitless but for the TBP analyses the injections were split 1:20. The GC oven was temperature programmed for both analyses as follows: the temperature was initially held at 60 °C for 1 min, programmed from 60 to 225 °C at 15 °C/min, and then 225 to 280 °C at 40 °C/min, and finally held at 280 °C for 37 min. The helium flow was 0.48 mL/min, the introducer temperature was 280 °C, and the GC−MS transfer line was 300 °C. The MS was operated in electron ionization mode with an energy of 70 eV and an ion source temperature of 180 °C. Quantitative analysis by MID was performed under software control by a Hewlett-Packard Vectra 386/25 computer running a Hewlett-Packard MS ChemStation data system. For the TBA analyses the ions monitored were *m*/*z* 238, 240, and 242 (internal standard) and *m*/*z* 329, 331, 344, 346, and 348 (TBA) and for the TBP analyses *m*/*z* 253, 255, and 257 (internal standard) and *m*/*z* 328, 330, 332, and 334 (TBP). The GC−MS was calibrated by the analysis of three different concentrations of TBA and TBP (0.5, 5, and 25 μg/mL in iso-octane) with a constant concentration of the internal standards (1 μg/mL). Response factors for TBA and TBP, with respect to the internal standards, were calculated by the data system software, and these were used to determine the concentrations of the target compounds in the extracts. The calibrations were performed on the day of the analysis, and each analysis was carried out in duplicate. If a sample contained analytes outside of the calibration range of the mass spectrometer, more internal standard was added and a diluted subsample was analyzed. Reported data has been corrected for a 7% loss of TBA and a 26% loss of TBP during extraction and concentration (see below). The detection limit for TBA in water was 0.0001 μg/L and in sultanas 0.01 μg/kg based on a factor of three times the background noise. Similar values were obtained for TBP in these materials.

In the analyses for TBA the presence of this compound was confirmed by the appearance of a single peak in the total ion chromatogram at linear retention index 1637, by the presence of the five characteristic ions listed above, and by the appearance of the correct isotopic ratios for these ions.

Extraction Efficiencies. The extraction efficiencies of the SDE technique for the recovery of TBA from sultanas and fiberboard and for TBP from fiberboard were determined as follows. Samples of sultanas (25 g) or fiberboard (25 g), previously shown to be free of TBA and TBP, were each inoculated with an iso-octane solution containing 100 ng of the compounds. The treated materials were then extracted, and the extracts were analyzed by GC−MS as described above. The extractions were performed in duplicate. The average recovery of TBA from both materials was 93%. The average recovery of TBP from fiberboard was 74%.

Metabolism of 2,4,6-Tribromophenol by *P. variotii*. Spore suspensions of *P. variotii* isolates were prepared by adding an aqueous solution of Tween 80 (1 g/L) to sporulating slope cultures to achieve spore suspensions of about 10^6 spores/200 μL of inoculant. The fiberboard growth medium was prepared from a sample of fiberboard that was cut into 12 squares (13 cm × 13 cm, about 12 g). Ten score marks were made with a scalpel along the corrugations of each square, and the fiberboard was bent so that the cuts were expanded. A stock solution (200 μL) containing 20 μg of TBP was injected along the cuts of the fiberboard square. The square was rolled into a cylinder and placed into a wide neck 1 L glass jar containing a small beaker of saturated potassium dichromate solution (25 mL) to provide a relative humidity of 98% (Whitfield et al., 1991). A total of 11 squares treated with TBP and one with no treatment were prepared in this fashion. The jars were closed by a metal screw cap with aluminum foil as liner and were sterilized by autoclaving at 121 °C for 15 min. When the relative humidity in the jars had reached equilibrium, the lids were unscrewed and aliquots (200 μL) of the spore suspension of *P. variotii* were aseptically injected into ten of the 11 fiberboard cylinders treated with TBP and the one cylinder with no treatment. The remaining cylinder treated with TBP but not inoculated with the spore suspension and the inoculated cylinder with no TBP were the controls. The jars, containing 21% oxygen, were sealed and then incubated at 30 °C. One treated and two control jars were held for 10 weeks; other jars were sampled weekly commencing at the end of the first seven days. When a cylinder was removed from a jar it was either immediately extracted, as previously described, or was wrapped in aluminum foil and stored at −20 °C until required for extraction. The concentrated extracts were analyzed for TBA as previously described.

Preparation of Samples for Sensory Assessment. TBA was dissolved in absolute ethanol at a concentration of 100 μg/L and was stored at −10 °C as a stock solution. From this solution, by successive dilution with purified water, nine concentrations were prepared covering the range 1×10^{-7} to 1×10^{-3} μg/L. These aqueous solutions were assessed for off-odor by the sensory panel and the results so obtained were used to determine the odor threshold value of TBA in water. The aim was to vary the odor intensity from "no odor" to "moderately odorous".

Batches of dried fruit containing eight concentrations of TBA covering the ranges 1×10^{-4} to 0.48 μg/kg were prepared as follows. TBA was dissolved in absolute ethanol at a concentration of 1 mg/mL and was stored at −10 °C as a stock solution. Sultanas (1 kg) were placed in a 5 L cylindrical glass jar. A predetermined amount of TBA stock solution was diluted with purified water and the solution was added to a piece of circular filter paper (15 cm o.d.). The impregnated paper was placed in a cylindrical cage of stainless steel gauze (40 mesh, 18.5 cm long × 3.8 cm o.d.) that was suspended in the jar containing the sultanas. The jar was sealed with a screw cap. Contamination of the sultanas with TBA was achieved by placing the jar on a ball-mill (Pacific Electric Co. Pty Ltd. USA) and tumbling the dried fruit for 24 h. The contaminated sultanas were stored in a sealed jar at 20 °C until required for sensory assessment. The concentrations of TBA in the contaminated sultanas were determined by GC−MIM−MS as previously described.

Sensory Assessment. Assessors were members of the CSIRO Division of Food Science and Technology, Sydney Laboratory, who demonstrated an ability to detect and identify the odor of TBA in preliminary studies. A panel of 20 assessors served in each of the 12 assessments (water and sultanas). For the assessment of TBA in aqueous solution the sample volumes were 20 mL and were served at ambient temperature (20 °C) in small glass tumblers. For the sultanas, 20 g samples were served in small tumblers wrapped in aluminum foil and covered with glass dishes. The dried fruit was warmed at 60 °C for 60 s just before serving to concentrate the odor in the headspace above the sultanas.

The order of presentation of the samples was determined by a William's Square Design. A triangle of samples was presented to the panelist; two of these were control samples

and one a contaminated sample. Position of the contaminated sample was varied randomly throughout the assessments. The panelists were asked to smell each sample in turn, select the odd sample, and if the sample did not have a musty smell to describe any perceived odor. Two assessments were made each day by the panelists, a morning session and an afternoon session. Each concentration of TBA was presented to the panelist twice.

Analysis of Sensory Data. The 20 panelists scored the samples of water plus TBA as musty (Y) or not musty (N). Two trials were performed. The concentrations of TBA in Trial 1 were 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6}, and 10^{-7} μg/L, and Trial 2 had concentrations of 7.5×10^{-4}, 5×10^{-4}, 2.5×10^{-4}, and 7.5×10^{-5} μg/L. In both trials each panelist tested duplicates and the means of the percent of "Y" answers for the nine concentrations were determined. Threshold was defined to be the concentration at which there was a two-thirds positive response. This concentration was determined by linear calibration (Snedecor and Cochran, 1989). However, the raw data contained two outliers for the concentration 10^{-4} μg/L. The responses to this concentration were both less than those given to the most dilute concentration (10^{-7} μg/L); therefore, they were rejected as being implausible. With the elimination of these values, a better fit was obtained for the regression model [proportional positive response $y = a + b$ log(concentration)], due to less variance across the concentrations.

Similarly, the above technique was applied to the data obtained from the sultana trials. Again, two trials were performed; the concentrations of TBA in Trial 1 were 10^{-1}, 10^{-2}, 10^{-3}, and 10^{-4} μg/kg, while Trial 2 had concentrations of 0.48, 0.42, 0.29, and 0.16 μg/kg. The raw data for the sultanas also contained two outliers at a concentration of 10^{-1} μg/kg. Elimination of these values again provided a better fit for the regression model.

Diffusion of 2,4,6-Tribromoanisole through Polyethylene Wrapping Film. Sultanas (25 g) were weighed into a pouch made of 50 μm polyethylene film. The weight of film used in the pouch was 1 g. The pouch was heat-sealed and placed between two pieces of 7 × 18 cm virgin kraft fiberboard (9 g each) that had previously been contaminated with TBA (150 ng/board); the sultanas were therefore in contact with a total of 300 ng of TBA. To ensure close contact of each part of the diffusion cell, the pouch and fiberboards were placed between two pieces of aluminum plate held with metal clips. The cell was then wrapped in aluminum foil and was stored in a glass jar. A total of eight cells were prepared of which four were stored at 22 °C and the remainder at 30 °C. At the end of seven days, two cells were removed from each of the constant temperature rooms; the remaining cells were removed at the end of 14 days. The fruit was removed, and the fruit and pouches were separately extracted using the method previously described. The extracts after concentration were analyzed for TBA by GC−MIM−MS.

RESULTS AND DISCUSSION

Metabolism of 2,4,6-Tribromophenol by *P. variotii*. Data obtained from the analysis of the 10 samples of fiberboard contaminated with TBP and inoculated with strains of *P. variotii* showed that the maximum production of TBA (equivalent to 100% conversion of TBP) occurred after 6 weeks of incubation at 30 °C (Figure 1). However, in the following weeks the concentration of TBA at first decreased dramatically (weeks 7 and 8) and then more slowly, to almost zero, over the last two weeks. The metabolite was not detected in either of the control samples. The reduction in TBA concentration after week 7 has been attributed to its evaporation and partial adsorption on the surfaces of the jars and lids. This was supported by the observation that each jar opened after week 3, including that opened after week 10, had an intense musty odor. Furthermore, under similar experimental conditions, TCA was also lost from the fiberboard medium (Whitfield et al., 1991). However, the possibility that TBA may be degraded by the fungus to other products cannot be excluded.

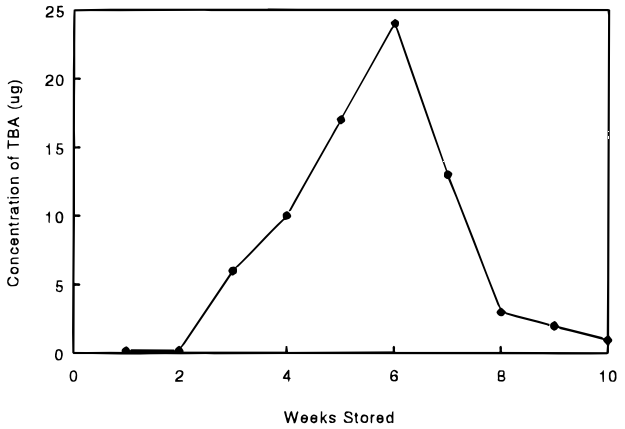

Figure 1. Production of 2,4,6-tribromoanisole from 20 μg of 2,4,6-tribromophenol on fiberboard medium by *P. variotii*.

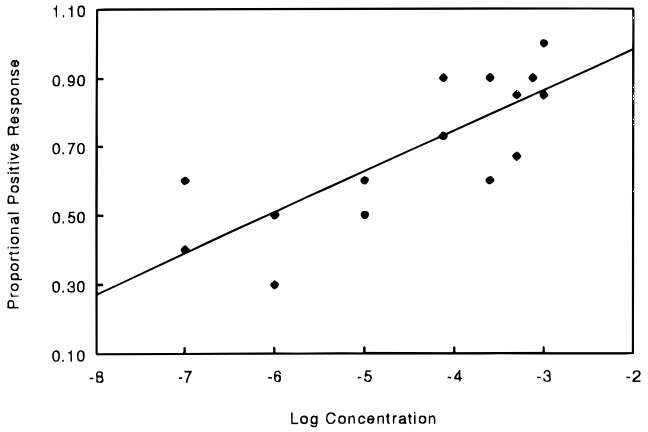

Figure 2. Plot of proportional positive odor response against the log of the concentration of 2,4,6-tribromoanisole in water.

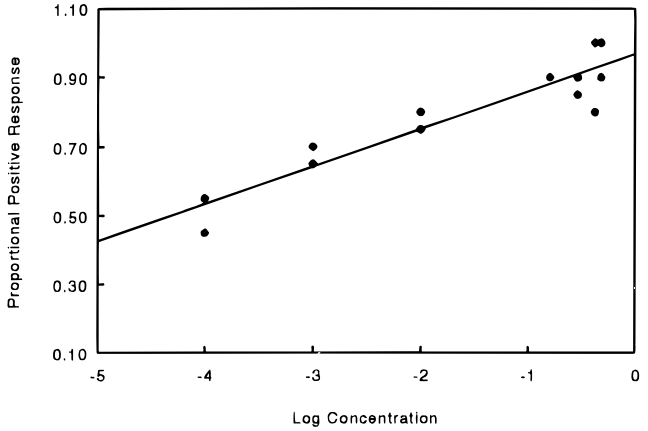

Figure 3. Plot of proportional positive odor response against the log of the concentration of 2,4,6-tribromoanisole in sultanas.

Odor Thresholds of 2,4,6-Tribromoanisole. The plot of log of the concentration of TBA in water against the proportional positive response of the sensory panel is shown in Figure 2, and that for TBA in sultanas is shown in Figure 3. In these diagrams the two outliers for the concentration 10^{-4} μg/L of TBA in water have been omitted, as have those for the concentration 10^{-1} μg/kg of TBA in sultanas. Application of the linear calibration method of Snedecor and Cochran (1989) gave a threshold concentration of TBA in water of 2×10^{-5}

Table 1. Concentration of 2,4,6-Tribromoanisole in Packaged Sultanas and Polyethylene Film Absorbed from Two Sheets of Fiberboard, Each Treated with 150 ng of Authentic Compound (Total: 300 ng)

	2,4,6-tribromoanisole[a]			
	sultanas (ng/25g)		polyethylene (ng/g)	
storage time	22 °C	30 °C	22 °C	30 °C
7 days	23 (0.9 μg/kg)	26 (1.0 μg/kg)	53	55
14 days	37 (1.4 μg/kg)	41 (1.6 μg/kg)	71	77

[a] 2,4,6-Tribromoanisole was determined by GC−MIM−MS. Values are averages of duplicate analyses of single extracts

μg/L and in sultanas of 2×10^{-3} μg/kg at the two-thirds level of positive response. The adjusted R^2 associated with these calculations in both cases gave $p < 0.001$.

The value obtained in water is comparable with the value of 8×10^{-6} μg/L obtained by Saxby (1982) and is almost identical with the value of 3×10^{-5} μg/L obtained for TCA in water by Griffiths (1974). However, the value obtained for TBA in sultanas is appreciably lower than the value of 0.12 μg/kg reported by Whitfield et al. (1987) for the flavor threshold concentration of TCA in sultanas. Based on these findings the odor threshold concentration of TBA in sultanas is about 60 times lower than that of TCA.

The fungal metabolite TCA is well-known for its ability to contaminate foods and beverages with a musty odor and flavor, making such materials unpalatable (Land et al., 1975; Buser et al., 1982; Whitfield et al., 1985). With an even lower odor threshold concentration, TBA must be regarded as a major potential threat to food quality should it become a common contaminant.

Permeation of 2,4,6-Tribromoanisole through Polyethylene Film. Table 1 records the results obtained from the storage of packaged sultanas and polyethylene film in contact with fiberboard contaminated with TBA. The results show that there is little difference in the concentrations of TBA found in sultanas at 22 and 30 °C. However, a doubling of the storage time from 7−14 days resulted in a 60% increase in the amount of TBA absorbed. Furthermore, the amount of TBA absorbed by the sultanas after 7 days of storage was equivalent to 8% of the TBA originally present in the fiberboard and after 14 days it was 12%. The presence of finishing oil on the surface of the dried fruit presumably favors the absorption of the lipophilic compound TBA. Thus if a batch of fiberboard were to contain as little as 8×10^{-2} μg/kg of TBA, sultanas could be contaminated to a concentration of 6×10^{-3} μg/kg after only 7 days of storage. This would render the sultanas unacceptable to many consumers.

The polyethylene film, like the sultanas, showed little difference in the concentrations of TBA absorbed at 22 and 30 °C, although the amounts absorbed were between 90 and 130% greater than the levels found in the sultanas. Of interest, after 7 days of storage 27% of the TBA added to the fiberboard had been transferred to the sultanas or the film and after 14 days this amount had increased to 40%. Thus the rate of diffusion and the solubility of TBA in the polyethylene film were comparable with those previously observed for TCA (Whitfield, unpublished).

CONCLUSION

The detection of the fungicide TBP in fiberboard, combined with its reported use as a wood preservative, must be viewed with considerable concern. Should the use of this fungicide become as widespread as that of TCP, the occurrence of TBA-induced mustiness in food and beverages could match that of its chlorinated analog TCA. Furthermore, the use of derivatives of TBP as fire retardants in resins and polymers may create similar environmental problems through their disposal as waste materials. For example, the presence of low levels of TBP and subsequently TBA in potable water, fish, crustaceans, and shellfish could have a detrimental effect on their flavor quality and consumer acceptability. Thus the continued use of TBP, either as a fungicide or as a fire retardant, should be carefully monitored and a suitable alternative found before this compound and its metabolite TBA become costly food contaminants.

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LITERATURE CITED

Allard, A.-S.; Remberger, M.; Neilson, A. H. Bacterial *O*-methylation of halogen-substituted phenols. *Appl. Environ. Microbiol.* **1987**, *53*, 839−845.

Boyle, J. L.; Lindsay, R. C.; Stuiber, D. A. Bromophenol distribution in salmon and selected seafoods of fresh- and saltwater origin. *J. Food Sci.* **1992**, *57*, 918−922.

Buser, H.-R.; Zanier, C.; Tanner, H. Identification of 2,4,6-trichloroanisole as a potent compound causing cork taint in wine. *J. Agric. Food Chem.* **1982**, *30*, 359−362.

Griffiths, N. M. Sensory properties of chloroanisoles. *Chem. Senses Flavor* **1974**, *1*, 187−195.

Knuutinen, J.; Palm, H.; Hakala, H.; Haimi, J.; Huhta, V.; Salminen, J. Polychlorinated phenols and their metabolites in soil and earthworms of sawmill environment. *Chemosphere* **1990**, *20*, 609−623.

Land, D. G.; Gee, M. G.; Gee, J. M.; Spinks, C. A. 2,4,6-Trichloroanisole in broiler house litter: a further cause of musty taint in chickens. *J. Sci. Food Agric.* **1975**, *26*, 1585−1591.

Miyazaki, T.; Kaneko, S.; Horii, S.; Yamagishi, T. Identification of polyhalogenated anisoles and phenols in oysters collected from Tokyo Bay. *Bull. Environ. Contam. Toxicol.* **1981**, *26*, 577−584.

Nishibori, S.; Kondo, H. Halogen-containing aromatic diester flame retardants for organic polymers. Japan Patent JP 05,295,163, 1993.

Pulido, M. L.; Ayzaguer, J. M. Synergistic microbicidal combinations of 2-(thiocyanomethylthio)benzothiazole and a trihalogenated phenol. US Patent US 4,983,618, 1991.

Saxby, M. J.; Reid, W. J.; Wragg, S. *Index of Chemical Taints*. Leatherhead Food RA: Leatherhead, 1992.

Sheikh, Y. M.; Djerassi, C. 2,6-Dibromophenol and 2,4,6-tribromophenol—antiseptic secondary metabolites of *Phoronopsis viridis*. *Experientia* **1975**, *31*, 265−266.

Snedecor, G. W.; Cochran, W. G. *Statistical Methods*, 8th ed.; Iowa State University Press: Ames, Iowa, 1989; p170.

Sweetman, J. A.; Simmons, M. S. The production of bromophenols resulting from the chlorination of waters containing bromide ion and phenol. *Water Res.* **1980**, *14*, 287−290.

Takahashi, K.; Sato, J. Fire-resistant thermoplastic resin compositions, fireproofing agents, and manufacture of the fireproofing agents. Japan Patent JP 06,157,873, 1994.

Tindale, C. R.; Whitfield, F. B.; Levingston, S. D.; Nguyen, T. H. L. Fungi isolated from packaging materials: their role in the production of 2,4,6-trichloroanisole. *J. Agric. Food Chem.* **1989**, *49*, 437−447.

Towa Mokuzai Co., Ltd. Preservation of wood. Japan Patent JP 57,131,506, 1982.

Watanabe, I.; Kashimoto, T.; Tatsukawa, R. Polybrominated anisoles in marine fish, shellfish, and sediments in Japan. *Arch. Environ. Contam. Toxicol.* **1983**, *12*, 615−619.

Weber, K.; Ernst, W. Occurrence of brominated phenols in the marine polychaete *Lanice conchilega*. *Naturwissenschaften* **1978**, *65*, 262.

Whitfield, F. B.; Nguyen, T. H. L.; Shaw, K. J.; Last, J. H.; Tindale, C. R.; Stanley, G. Contamination of dried fruit by 2,4,6-trichloroanisole and 2,3,4,6-tetrachloroanisole adsorbed from packaging materials. *Chem. Ind. (London)* **1985**, 661−663.

Whitfield, F. B.; Shaw, K. J.; Nguyen, T. H. L. Simultaneous determination of 2,4,6-trichloroanisole, 2,3,4,6-tetrachloroanisole and pentachloroanisole in foods and packaging materials by high-resolution gas chromatography-multiple ion monitoring-mass spectrometry. *J. Sci. Food Agric.* **1986**, *37*, 85−96.

Whitfield, F. B.; McBride, R. L.; Nguyen, T. H. L. Flavour perception of chloroanisoles in water and selected foods. *J. Sci. Food Agric.* **1987**, *40*, 357−365.

Whitfield, F. B.; Last, J. H.; Shaw, K. J.; Tindale, C. R. 2,6-Dibromophenol: the cause of an iodoform-like off-flavour in some Australian crustacea. *J. Sci. Food Agric.* **1988**, *46*, 29−42.

Whitfield, F. B.; Nguyen, T. H. L.; Tindale, C. R. Effect of relative humidity and incubation time on the *O*-methylation of chlorophenols in fibreboard by *Paecilomyces variotii*. *J. Sci. Food Agric.* **1991**, *55*, 19−26.

Whitfield, F. B.; Shaw, K. J.; Svoronos, D. The volatile aroma compounds of Australian marine algae. *Proc. Int. Congr. Flav. Frag. Essen. Oils, 12th*, **1992a**, 365−372.

Whitfield, F. B.; Shaw, K. J.; Walker, D. I. The source of 2,6-dibromophenol: cause of an iodoform taint in Australian prawns. *Water Sci. Technol.* **1992b**, *25*, 131−138.

Whitfield, F. B.; Helidoniotis, F.; Svoronos, D.; Shaw, K. J.; Ford, G. L. The source of bromophenols in some species of Australian fish. *Water Sci. Technol.* **1995**, *31*, 113−120.

Wittlinger, R.; Ballschmiter, K. Studies of the global baseline pollution. XIII. C_6-C_{14} organohalogens (α- and γ-HCH, HCB, PCB, 4,4′-DDT, 4,4′-DDE, *cis*- and *trans*-chlordane, *trans*-nonachlor, anisols) in the lower troposphere of the southern Indian Ocean. *Fresenius J. Anal. Chem.* **1990**, *336*, 193−200.

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