presented in Figure 2. Extrapolation of the data indicated that unfortified 1R1 cigarette tobacco contained about 130 ppm of MH, which is in fair agreement with the 103 ppm reported earlier (Haeberer *et al.*, 1974). About 55 μg of MH per unfortified 1R1 cigarette, or about 0.5%, was transferred to mainstream smoke. Apparently, about 99% of the MH in cigarette tobacco is pyrolytically degraded into other products, assuming that sidestream smoke does not contain a significantly different MH concentration.

Finally, it was of interest to determine whether or not any MH distills ahead of the burning cone of a cigarette. If this were the case, then considerable quantities of MH would be found in the butts. Cigarette butts from unfortified and fortified cigarettes (0.8 or 1.6 mg added MH/g of tobacco) were analyzed for MH content. Butts with 1.6 mg of MH added contained 0.4 mg of MH. A 23-mm butt represents about 27% of an 85-mm cigarette, and should contain about 27% of the applied MH. The analysis showed that the butt contained 25% of the MH. This indicated that MH does not distill ahead of the burning zone to be condensed and concentrated in the butt. Since distillation can now be ruled out, the disperse phase or aerosol particles must be responsible for transport of MH into smoke.

The behavior of MH under pyrolytic conditions could be anticipated from its stability during melting point determinations. It was observed that MH does not melt but degrades at 296°. This substantiates our findings of pyrolytic decomposition of MH in the burning cigarette.

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Chloroanisoles as Off-Flavor Components in Eggs and Broilers

Musty eggs and broilers were analyzed for the presence of chloroanisoles and chlorophenols. The taint was caused by tri- and tetrachloroanisole. The contamination of the poultry products was brought about either by the use of woodshavings or by the use of contaminated feed.

In 1972 and 1973 a distinct musty taint was often observed in eggs and broilers from poultry farms in different parts of The Netherlands.

In earlier investigations (Engel *et al.*, 1966; Curtis *et al.*, 1972) it was shown that a musty taint in eggs and broilers was caused by 2,3,4,6-tetrachloroanisole and pentachloroanisole. These compounds were also found to be present in litter and woodshavings from the relevant poultry houses where they were picked up by the chickens. It was assumed, therefore, that the odor was thus transferred to the eggs and the carcasses.

It has been proved by Curtis *et al.* (1972) that the presence of chloroanisoles is caused by the ability of the microorganisms in the litter to form 2,3,4,6-tetrachloroanisole and pentachloroanisole from the corresponding chlorophenols. Furthermore, Cserjesi and Johnson (1972) found that *Trichoderma* sp. were able to convert pentachlorophenol partially into pentachloroanisole.

Technical grade pentachlorophenol, which is used as a wood preservative and contains up to 13% impurities, mainly isomeric tetrachlorophenols (Melnikov, 1971), may act as a precursor for the chloroanisoles. In addition, Ide *et al.* (1972) found that pentachlorophenol itself was decomposed by microorganisms into mono-, di-, tri-, and tetrachlorophenols.

Since chlorophenols may act as precursors for the corresponding chloroanisoles, both the presence of chloroanisoles and chlorophenols was determined in the products in question. The results of the investigation are described here.

Woodshavings and tainted products from poultry houses were analyzed. Since tainted products were also produced in poultry houses where no woodshavings were used, feed samples and in a few instances some of the raw materials used in the feed were examined as well.

MATERIALS AND METHODS

The methods applied to concentrate the chloroanisoles and chlorophenols were specific for each type of product. Eggs having a musty taint were selected from suspected lots. As the taint was located in the egg yolks, only this fraction was subjected to analysis. The yolk samples were saponified in an alcoholic KOH solution and subsequently extracted with ether. The extract was washed with water until all alkali was removed.

Tissue samples of tainted broilers, mainly consisting of depository fat and skin, as well as feed samples, were steam distilled and extracted with pentane–ether in an apparatus described by Likens and Nickerson (1964). The woodshavings were extracted in a Soxhlet apparatus. All extracts, which were dried over sodium sulfate and concentrated by distilling off the solvents, were used for mass spectrometric analysis.

Samples of animal grease, used for the preparation of feed, were dried over sodium sulfate and subjected for 2.5 hr to the high vacuum sublimation procedure of Lea and Swoboda (1962) at 80°. The volatile material, which was condensed on a cold finger, was washed off with ether and used for analysis.

Fractions of the concentrates were introduced with a cold probe into the ionization chamber of a double focus-

Table I. Presence of Chloroanisoles and Chlorophenols in Musty Eggs and Broilers, in Feed, and in Woodshavings Used in Poultry Houses[a]

Sample	No.	DCA[b]	DCPH[c]	TriCA[d]	TriCPH[e]	TeCA[f]	TeCPH[g]	PCA[h]	PCPH[i]
Eggs	1	−	−	+	Tr	Tr	Tr	−	+
	2	0	0	+	+	+	−	−	−
Broilers	1	0	0	+	Tr	++	Tr	−	−
	2	0	0	+	−	+	−	−	−
Chicken feed	1	0	0	Tr	++	++	+	−	++
	2	0	0	+	++	Tr	+	−	++
	3	−	0	+	++	−	−	−	−
	4	−	−	++	Tr	−	−	−	+
Animal grease	1	0	0	0	0	+	0	0	0
	2	0	0	0	0	++	0	0	0
Woodshavings	1	0	0	+−	+	++	−	++	++
	2	++	0	+	++	++	+	−	−
	3	−	−	+−	Tr	++	−	−	Tr
	4	0	0	0	0	++	++	−	++
Pentachlorophenol (Puriss.)		−	−	−	Tr	−	++	−	0
Sodium pentachlorophenate		0	0	−	+	−	++	−	0

[a] +, present; ++, present in large quantity; tr, traces present; −, not present; 0, not determined. [b] Dichloroanisole. [c] Dichlorophenol. [d] Trichloroanisole. [e] Trichlorophenol. [f] Tetrachloroanisole. [g] Tetrachlorophenol. [h] Pentachloroanisole. [i] Pentachlorophenol.

ing mass spectrometer (Varian-MAT, Model 731). The analysis for the presence of chloroanisoles and chlorophenols was carried out by single ion detection at a resolution of 15,000 using a peak-match system, calibrated to the exact masses of the components of interest. The detection limit of this procedure was 0.1 ppb. Some of the results are summarized in Table I.

RESULTS AND DISCUSSION

The tainted eggs and broilers were found to contain tri- and tetrachloroanisole, as well as tri- and tetrachlorophenol, while the eggs also contained pentachlorophenol. In earlier investigations by Engel *et al.* (1966) and Curtis *et al.* (1972) tetrachloroanisole was reported to cause a musty taint in eggs and broilers. Trichloroanisoles—having extremely low threshold values (Curtis *et al.*, 1972)—have not been detected in poultry products so far. Recently the presence of 2,4,6-trichloroanisole in a number of essentil oils was reported by Stoffelsma and de Roos (1973), causing a musty naphthalenic odor.

In the investigation described here no chloroanisoles and chlorophenols were found in eggs and broilers of good organoleptic quality.

The musty taint was caused either by contaminated woodshavings or feed. The woodshavings appeared to contain di-, tri-, tetra-, and pentachloroanisole, as well as tri-, tetra-, and pentachlorophenol (Table I). When no shavings were used in the poultry house, it was always the feed that proved to be contaminated, in fact mostly with trichloroanisole.

When starting the examination of raw materials incorporated in the feed in 1972, only the analysis of tetrachloroanisole was carried out. This substance was found to be present in several lots of animal grease. At a later stage there was no evidence of any contamination of the grease although the feed still proved to be contaminated. Unfortunately, the reason for the contamination could not be ascertained.

Both technical grade sodium pentachlorophenate and pentachlorophenol (Puriss.) contain tri- and tetrachlorophenol, whereas chloroanisoles were absent (Table I). Therefore, chlorophenols have to be converted to act as off-flavor components.

From Table I it can be seen that the contamination does not show any particular pattern. Contamination with tri- and tetrachloroanisoles appears to occur more frequently then with pentachloroanisole.

CONCLUSIONS

The data show that in musty eggs and broilers tetrachloroanisole and trichloroanisole are present. The latter compound is a potent off-flavor component that has not been previously reported to occur in poultry products. The taint was not only caused by the use of woodshavings, as has been found in earlier investigations, but also by contaminated feed. This may have been caused either by the use of contaminated animal grease or another contaminated raw material.

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