Contribution of Phenols to Boar Odour

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by R.L.S. Patterson

Meat Research Institute,

Agricultural Research Council,

Downing Street, Cambridge.

Introduction

The muscle and particularly the fat of entire adult male pigs (boars) has long been known in many cases to develop an unpleasant, nauseating odour when heated. This characteristic, which prevents its widespread use as a source of bacon and pork products, appears to be confined largely to the boar, because meat from the castrated animals (hogs) and the virgin females (gilts) does not generally show this taint or off-odour during cooking. However, a survey of the incidence of boar odour or sex odour carried out by Pearson [1] showed that it was detectable in only 64% of the boars tested, of which 28% were rated as strong and 36% as slight, and also in 5% of the barrows (hogs) and gilts. This suggests that the component(s) responsible for the odour may not only occur in the boar in variable amount, but may also occur to a small extent in hogs and gilts. If these latter two classes do produce the odorous compounds or their precursors, presumably they do so in quantities which normally result, except in the case of the five per cent, in the production during cooking of an odour below the average threshold of detection.

Previous work [2,3] has shown that in the case of entire boars the odour is strongest in the fatty tissue surrounding the penis and prepuce. Heated subcutaneous fat, especially from the flank, of 200lb live weight boars can be distinguished subjectively by odour from that of hogs or gilts with comparative ease, but preliminary gas chromatographic analyses of volatile products from the heated fats failed to show any significant differences which could be responsible for the characteristic boar taint. Similar findings have been reported by Pearson et al. [4, 5].

Since fat from the flank appeared subjectively to be richer in the odorous component(s), the reproductive tract and prepuce were considered to be possible sources of the odorous compounds or their precursors responsible for the

production of taint. The preputial diverticulum, situated near the orifice of the penis, is a sac which, in some boars, perhaps one in ten, becomes filled with fluid. The volume of the fluid can vary from a few drops to about 100 millilitres.

Experimental

Samples of the opaque, dark brown fluid, with a pH between 8.5 and 9.5 and having frequently a strong ammoniacal odour, were obtained from boars (Large Whites) by "milking" the pouch behind the penis. After filtration to remove pieces of straw, grit and other solid material, the fluid was extracted batchwise at the original pH with diethyl ether (carbonyl- and peroxide-free), until no further odour was extracted into the ether. This was judged by dipping a glass rod into each batch and smelling it after the ether had been allowed to evaporate. The pH of the fluid was then lowered to 4.5 by addition of 1 N hydrochloric acid and extraction with fresh ether continued until no further odour was removed. At this stage, the remaining aqueous fluid was almost completely odourless. The two ether extracts were concentrated separately by slow fractional distillation at 38°C to volumes of approximately 100 ml. before repeated extraction of each with 5% sodium bicarbonate, followed by similar extraction with 1 N sodium hydroxide. Acidification of these four alkaline extracts with 1 N hydrochloric acid, followed by individual extraction with diethyl ether, produced two pairs of ethereal solutions, each pair containing, respectively, acidic and phenolic components of the preputial fluid. The solutions had entirely different odours, that of the phenolic fractions being especially characteristic of pigsties. These fractions were therefore combined and analysed by gas-liquid chromatography after concentration by slow fractional distillation to approximately 1 ml.

The identification of the compounds present in the phenolic fraction was carried out using two columns containing different stationary phases 1) a 4ft x 4mm glass column packed with 15% phenyldiethanolamine succinate (PDEAS) on Chromosorb W (80-100 mesh) operated isothermally at temperatures of 130°C and 150°C and 2) a 5ft x 4 mm glass column packed with 5% xylenyl phosphate (XP)

on Celite (100-120 mesh) at 120°C. The G.L.C. instruments used were both manufactured by Pye Co. Ltd., one equipped with a strontium -90 ionisation detector and the other with dual flame detectors. Argon carrier gas was used throughout.

One major peak and five smaller peaks were recorded. The major component corresponded in retention behaviour with the meta and para isomers of cresol and was present in sufficient quantity to permit trapping from the chromatograph. Mass spectral analysis showed an intense parent ion peak (M) at mass 108 (cresol C_7H_8O = 108), accompanied by a slightly higher peak at (M-1), resulting from the characteristic loss of a hydrogen atom from a methyl group. Loss of the elements of water from the molecule resulted in a strong peak at mass 90 (M-18), confirming the presence of a hydroxyl group. The ratio of the M/(M-1) peaks differ for the three isomeric cresols depending upon the relative positions of the methyl and hydroxyl substituents [6]. The ratios of M/(M-1) are 1.327, 1.250 and 0.926 for the ortho, meta and para isomers respectively; the ratio for the experimental material was 0.966, indicating that the compound was almost exclusively p-cresol. This was confirmed by comparison of the infrared spectrum of the isolated material with infrared spectra of authentic samples of the three isomers. The spectra of the isolated material and p-cresol corresponded exactly. Considering the three isomeric cresols, the para isomer would be anticipated since it could be theoretically explained as a metabolite of the amino acid tyrosine.

In an attempt to identify the remaining five peaks recorded on the chromatogram, a number of reference compounds, including the phenols listed in Table I, were chromatographed on the PDEAS column under identical conditions to those used for the experimental material.

TABLE I

Reference Phenols

Phenol	2:3-Xylenol
o-Cresol	2:4-Xylenol
m-Cresol	2:5-Xylenol
p-Cresol	2:6-Xylenol
o-Methoxyphenol	3:4-Xylenol
m-Methoxyphenol	3:5-Xylenol
p-Methoxyphenol	2:3:5-Trimethylphenol
2:6-Dimethoxyphenol	p-Ethylphenol
Catechol	o-Hydroxydiphenyl
Resorcinol	

The compounds which showed exact or close agreement in retention behaviour with peaks in the experimental material were, in order of increasing retention, guaiacol (o-methoxyphenol), phenol, o-cresol, [m-cresol, p-cresol], 2:4-xylenol, 2:5-xylenol, 2:3-xylenol, p-ethylphenol, 3:5-xylenol and m-methoxyphenol. Since the identity of p-cresol had been proved beyond doubt by mass spectral and infrared analyses, those compounds listed above which corresponded to this peak were disregarded i.e. m-cresol, 2:4-xylenol and 2:5-xylenol. The relative retention values, corrected to three significant figures, of the remaining compounds related to p-cresol as an internal standard are shown in Table II.

TABLE II

Relative Retention Values w.r.t. p-cresol

15% PDEAS on Chromosorb W at 130°C.

Reference Compounds		Experimental Material	
Compound	Vr_1/Vr_2	Peak No.	Vr_1/Vr_2
Guaiacol	0.580	1	0.581
Phenol	0.695	2	0.693
o-Cresol	0.737		
		3	0.867
p-Cresol	1.000	4	1.000
2:3-Xylenol	1.42		
p-Ethylphenol	1.45	5	1.45
3:5-Xylenol	1.50		
m-Methoxyphenol	4.59	6	4.52
m-Methoxyphenol	3.94(a)	6	3.95(a)

(a) at 150°C.

It can be seen from the table that guaiacol, phenol, p-ethylphenol and m-methoxyphenol agreed most closely with four of the five unidentified peaks. Furthermore, the very characteristic "smoked bacon" smell of guaiacol could be recognised in the effluent of the chromatograph at the time of emergence of the first peak.

The same reference compounds and the experimental material were chromatographed on the highly polar 5% xylenyl phosphate column at 120°C. The relative retention values expressed in relation to p-cresol are listed in Table III.

TABLE III

Relative Retention Values w.r.t. p-cresol

5% xylenyl phosphate on Celite at 120°C

Reference Compounds		Experimental Material	
Compound	Vr_1/Vr_2	Peak No.	Vr_1/Vr_2
Guaiacol	0.362	1	0.362
Phenol	0.641	2	0.647
o-Cresol	0.776	3	0.786
p-Cresol	1.000	4	1.000
2:3-Xylenol	1.61		
p-Ethylphenol	1.75	5	1.75
3:5-Xylenol	1.85		
m-Methoxyphenol	4.31	6	4.36

These figures support the results obtained with the PDEAS column since the reference phenols which agreed most closely with the chromatographic peaks in the experimental material were guaiacol, phenol, o-cresol(trace), p-ethylphenol and m-methoxyphenol. It was again possible to confirm the presence of guaiacol by extinguishing the flame of the detector and smelling the effluent. Guaiacol was also unique amongst the phenols examined on this column as it displayed partial adsorption by the column, resulting in asymmetrical, tailing peaks which served as useful diagnostic features.

A trace of o-cresol was detected on the xylenyl phosphate column but was not detected on the PDEAS column where it should have appeared on the tail of the peak for phenol. p-Ethylphenol showed exact correspondence with the fifth peak of the experimental material, whilst 2:3-xylenol and 3:5-xylenol occured closely on either side (Table II). m-Methoxyphenol displayed fair agreement with the last unidentified peak.

No simple phenol was found which agreed with the third peak of the experimental material when analysed on the PDEAS column (Table II) nor was there a corresponding peak detected on the XP column (Table III). This peak was small and presumably coincided with one of the larger peaks on the latter column, or its concentration in the carrier gas at the time of elution may have been too low for detection under the operating conditions.

The results obtained from these two gas chromatographic columns show that the compounds detectable in the phenolic fraction of the boar preputial fluid are guaiacol, phenol, o-cresol (trace), p-cresol, p-ethylphenol and m-methoxyphenol, and one other unidentified compound. It is recognised that this method of characterising compounds is not infallible but confirmation of the minor components by mass spectroscopy was not considered feasible with the quantity of material available without access to a coupled gas chromatograph - mass spectrometer unit, which would be capable of analysing microgramme quantities of material without intermediate trapping from the gas chromatograph.

Quantitative Data

Three samples of preputial fluid were obtained, two from the same boar and

the third from a litter-mate. The boars were Large Whites and the samples of fluid were obtained at live weights of 166lbs, 193lbs and 195lbs respectively.

The quantities of the five phenols present in the ether extracts of the three samples, concentrated to known volumes, were determined by comparing their respective peak heights with those of standard solutions of each of the identified compounds under identical conditions of injection and analysis on the PDEAS column. The standard solutions were prepared as weight or volume per cent in diethyl ether depending upon the physical state of the compound at room temperature.

The analyses of the guaiacol and phenol were carried out together at 130°C using 40µl injections of the experimental solutions and mixed guaiacol - phenol standards. The p-cresol was also analysed at 130°C but in this case 1µl injections were adequate. The p-ethylphenol was analysed at 145°C and the m-methoxyphenol at 159°C, 40µl injections being necessary in both cases. The introduction of relatively large volumes of solvent did not have a detrimental effect on the subsequent analysis, provided the injection system was sufficiently hot to ensure instant volatilisation of the injection.

The values for each constituent phenol are given in Table IV, expressed in two ways 1) as microgrammes per millilitre of preputial fluid and 2) as a percentage of the total phenol content.

TABLE IV

Quantitative Data

Compound	Boar 1				Boar 2	
	Weight 166 lbs		Weight 193 lbs		Weight 195 lbs	
	µg/ml	% of Total	µg/ml	% of Total	µg/ml	% of Total

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	µg/ml	% of Total	µg/ml	% of Total	µg/ml	% of Total
Guaiacol	0.26	0.26	0.33	0.31	0.12	0.27
Phenol	0.83	0.84	0.66	0.62	0.20	0.45
p-Cresol	90.23	91.70	98.39	92.90	42.72	95.67
p-Ethylphenol	6.44	6.55	5.76	5.44	0.84	1.88
m-Methoxy-phenol	0.63	0.64	0.77	0.73	0.77	1.72
TOTAL PHENOL	98.39		105.91		44.65	

Agreement is reasonably good between values for the two samples from the same boar and also between the percentage figures for all three samples except for the p-ethylphenol and m-methoxyphenol values. The reason for the variation in the quantities of the latter two compounds and in the overall quantities of phenolic material from the two sources is not understood at present and will require analyses of further samples to establish the norm.

Discussion

It is clear that p-cresol is the principal component and also the major contributor to the odour of the preputial fluid. Guaiacol, which has a very low threshold of detection, will contribute a "smoky" note to the odour, although it is present in the least concentration. Phenol and m-methoxyphenol appear to have little direct effect on the odour at this level of concentration but the p-ethylphenol contributes a harsher "rubbery-phenolic" note to the rather sweeter odour of the p-cresol.

Judged subjectively by smell, neither p-cresol nor the other phenols, either individually or in combination, appear to be directly responsible for the taint of heated boar fat. However, a possible contribution by these compounds to the undesirable odour of tainted fat cannot be entirely ruled out until complete identification of the odorous volatile products of heated boar fat has been achieved.

On the other hand, a blend of these phenols is partly responsible for the "external" odour of a live, entire boar, whenever any of this fluid is expelled from the preputial sac, perhaps during copulation or repose. This is especially so when the contaminated areas, for example, the ground or belly of the animal, have an alkaline pH. During evaporation of the aqueous fluid, loss of ammonia occurs, with a resultant fall in pH to weekly acid conditions, causing the odour to alter due to contributions by acidic compounds.

Conclusion

It would appear that the odorous compounds which have so far been identified are not directly implicated in the problem of sex odour as related to consumer

acceptance of boar meat. They may however play a part in another important aspect of the sex odour problem, namely, the stimulating influence of the odour of a boar on a female pig in oestrus. Preliminary trials have indicated that p-cresol *per se* is not a pheromone. Its smell alone does not appear to be of great interest to a sow in heat but it is possible that in the presence of other odours emanating from a boar, the combined or synergistic effect may be quite different.

Summary

The meat and particularly the fat of boars frequently develops an unpleasant smell when heated. This taint or sex odour, which prevents the widespread use of boar meat as bacon and pork products, is confined largely to the boars since castration is an effective method of reducing the odour.

Since the odorous compound(s) responsible for taint may be the products of some secondary sex-dependent metabolism, secretion originating in the prepuce have been studied.

The odorous phenolic fraction of boar preputial fluid has been analysed by gas-liquid chromatography and five of the six phenols detected have been identified by retention data and were found to be guaiacol, phenol, p-cresol, p-ethylphenol and m-methoxyphenol. Quantitative data for the five phenols are presented; p-cresol is the major component comprising over 90% of the phenol content.

None of these phenols appears to be directly responsible, either individually or in combination, for the taint of heated boar fat when judged subjectively by smell. However, they do contribute to the odour of the live animals whenever any preputial fluid is released and, on account of this, they may play a part in the physiological stimulation of the female pig in oestrus when she is in the presence of a boar.

SOMMAIRE

La viande, et plus particulièrement la graisse du pourceau, a fréquemment une mauvaise odeur lorsqu'elle est chauffée. Ce relent, qui est une odeur sexuelle, empêche son utilisation pour la fabrication du bacon et des produits à base de porc. Elle se limite en grande partie aux pourceaux car la castration est devenue un moyen efficace de la réduire.

Etant donné que le(s) composé(s) étant à l'origine de ce relent peuvent être attribués à un métabolisme secondaire lié au sexe de l'animal, on a procédé à l'examen des secrétions provenant du prépuce.

La fraction phénolique de la secrétion préputiale dégageant une odeur chez le pourceau a été analysée au moyen de la chromatographie gaz-liquide. Cinq des six phénols détectés ont été identifiés par retention; ce sont le gaïacol, le phénol, le p-crésol, le p-éthylphénol et le m-méthoxyphénol. Des données quantitatives concernant les cinq phénols sont fournies. Le p-crésol est le composant principal puisqu'il comprend plus de 90% du contenu en phénol. Aucun de ces phénols ne semble directement responsable, individuellement ou associé aux autres, du relent que dégage la graisse du pourceau, si on en juge subjectivement par l'odeur. Cependant, ils concourrent à l'odeur que présente l'animal vivant chaque fois qu'une secrétion préputiale a lieu et, de ce fait, il est possible qu'ils jouent un rôle dans le processus de stimulation physiologique de la truie en rut quand elle se trouve en présence d'un pourceau.

Zusammenfassung

Das Fleisch, in ganz besonderem Maße das Fett des Ebers entwickelt beim Erhitzen einen übelriechenden Geruch. Kastrierung ist ein wirksames Mittel zur Geruchsverminderung - in diesem Fall ist dieser Makel, der Sexgeruch, der sich ausschließlich auf Eber beschränkt, ein Grund dafür, daß weitverbreiteter Gebrauch dieser Fleischart als Schinken oder Schweinefleischprodukt ausgeschlossen ist.

Die Geruchszusammensetzung mag ihre Ursache in einigen sekundären, sex-gebundenen Stoffwechsel haben; nämlich Ausscheidungen aus der Vorhaut, die bereits

untersucht wurden.

Die riechenden Phenolanteile der präputialen Flüssigkeit, wurden durch eine Gas-Flüssigkeit farbgraphisch analysiert und 5 von 6 Phenolanteilen, die festgestellt wurden, waren durch Verhaltungsergebnisse als Guajakol, Phenol, p-Kresol, p-Ethylphenol und m-Methoxyphenol identifiziert worden. Mengenmäßige Zahlenergebnisse der fünf Phenole sind gegenwärtig: p-Kresol ist mit 90% der Hauptkomponent des Phenolgehalts.

Keiner dieser Phenole, ob in reiner oder gebundener Form, scheint in erster Linie ausschlaggebend für die Geruchsursache erhitzten Fettes des Ebers zu sein. Dennoch spielen sie für den Geruchssinn der lebenden Tiere, wennimmer eine Vorhautabsonderung stattfand eine große Rolle, nämlich die physiologische Stimulation des Menstruationszyklus der weiblichen Artgenossen.

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