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STUDY OF CURING COMPONENTS OF SMOKE

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Although smoke-curing as a technological and food preserving method has a century - old history there are but few data regarding its chemical and physico-chemical nature.

The most part of earlier research was aimed at studying technological problems of smoking (1,2,3), determining composition of smoke (4,5,6,7) and others (8,9,10). Quite a number of data about chemical and mechanical nature of smoking are of a general or hypothetical character. The lack of deep knowlegde hampers improvement of smoking technique and technology including such alluring methods as electric smoking and smokeless curing by smoking preparations.

Therefore a systematic and profound study of smoking problems began in the All-Union Scientific Research Institute for Meat Industry (Moscow) in 1958. This paper deals with the first results of this work on research of curing components, i.e. those components of smoke, which penetrate into the product and give it particular odour, flavour and other properties.

Methods of research

Collection of curing components. Methods used in the study of odour and flavour of the other food-stuffs did not fit our aim because of two reasons at least: 1) The presence of large quantities of volatile substances in the products itself (meat, fish) hinders the identification and isolation of curing components; 2) The isolation of smoke constituents directly from smoked products in quantities sufficient for a subsequent separation, on a destillation tube, for instance, requires processing of hundreds and thousands of kg of smoke-cured products, making thus the study practically impossible.

Therefore curing components were collected by means of sausage models. Among various models (casings filled with cotton, minced paper, solutions of gelatine etc) casings filled with distilled water gave the best results. Curing components of smoke penetrated into such models rather intensively and accumulating phenols were completely similar to those of an ordinary sausage smoked under the same conditions.

Curing components of smoked models were extracted and divided into main organic groups along the scheme based on methods of the organic analysis and chemical investigations of woods (14,15).

The phenol components were divided on a rectification tube (with the capacity of 10 theoretical plates) in an atmosphere of nitrogen at the pressure of 3-4 mm of mercurial column.

Several methods of chromatography of smoke phenols on paper were used (16,17,18,19,20 etc.). The method (20) gave sufficiently good results. For a better division the chromatograms were passed through the solvent 3 times. Aliphatic aldehydes and ketones were chromatographed on the paper, processed by dimethylphormamid, in the form of 2,4-dinitrophenylhydrosones (21).

The infra-red spectrometry was used for identification of one of the cyclic ketones.

The antioxidant activity of different fractions of smoking components was determined by the accelerated kinetic method (22).

The diffusive method of Reddish and Roulet (23) was used ~~for a~~ comparison of bactericide strength of the extracted curing fractions.

A simple, spectacular and quick method was developed for the determination of the rate of penetration of phenols into the product during smoke-curing and storage. The method consists in receiving a reprint of phenols from the cut of a smoked product (24).

Results and discussion

A complicated mixture of the organic compounds which penetrated into the models was divided along the scheme (fig.1) into similar fractions: phenols, neutral substances (including hydrocarbons and aldehydes, ketones, alcohols), acids, organic bases and carbohydrates.

The isolated groups of substances were studied in regard to their antioxidant, bactericide and organopeltic substances. The results of the determination of the antioxidant activity for different smoking fractions are shown on fig.2. The curves present the accumulation of peroxides depending on the oxidation time when similar quantities of a fraction (0,02%) are added to the fat. The comparison of the curves shows, that while carbohydrat s, bases and neutral substances practically possess no antioxidant properties and acids possess these properties in a small degree, phenols significantly inhibit the fat turning rancid.

Microbiological studies were conducted with the microflora which is most tipical for sausages: sporeforming - B.subtilis, B.megaterium and B.mesentericus, conditional pathogenic - colibacillus and B.proteus, as well as on the staphylococcus culture.

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Bactericide effect of the curing fractions of smoke

Table 1

Test-cultures	Bactericide zones (mm)				
	Fractions				
	phenols	acid	neutral substances	basic	carbohydrate
B. subtilis	15	24	5	2	0
B. megoterium	23	25	3	4	0
B. mesentericus	22	27	5	0	0
B. coli comm	18	15	6	2	0
B. proteus	14	8	0	0	0
Staphilococcus aureus	20	20	8	2	0

The results obtained by the Reddish and Roulet method (table 1), show the differencies in the bactericide strength of the fractions under study: a) Far from possessing a bactericide effect, carbohydrate substances stimulate the growth of bacteria; b) Organic bases and mixtures of neutral substances possess rather weak bactericide properties; c) Summarized phenols and acids show the strongest bactericide effect with regard to all bacteria, including sporogenic flora.

Under experimental conditions the mixture of phenol substances proved to be the most effective with regard to B. coli comm and B. proteus, which are killed with more difficulties or not killed at all by the other curing fractions.

The results of the organoleptic test of the curing fractions of beech smoke are shown on table 2.

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Table 2

Organoleptic characteristics of smoke-curing fractions

Fraction	Colour	Flavor	Odour
Mixture of all neutral compounds	Light brown	Acrid, fatty	Peculiar, not pleasant
Summarized acids	Dark-yellow	Sour	Acrid, sour with a shade of acetic acid
Organic bases	Slightly yellowish	Almost without any flavor	Peculiar, not pleasant
Corbohydrates	Reddish-brown	Without any flavor	Very weak with a shade of caramel
Aromatic aldehydes	Colourless	-	Pleasant with a shade of almonds
Hydrocarbonic part of neutral substances	Dark-yellow	-	Peculiar, not pleasant.
Summarized phenols	Light brown	Acrid	Complicated phenol odour, rather pleasant.

Different quantities of these curing fractions were introduced into minces, which were subsequently used for preparing samples of sausages. It was found that all fractions except phenols had a negative influence on the flavour and odour of the prepared samples. Sausages in which summarized phenols were added, had sufficiently satisfactory flavor and odour, although the latter differed a little from the usual odour of smoke-cured sausages.

Thus preliminary study showed that the group of phenol compounds is one of the most promising for a subsequent investigation.

As a result of experiments with a number of models, summary phenols were isolated in quantities sufficient for distillation into separate fractions (for an example of distillation see table 3). These narrow fractions were characterized according to their boiling temperature, content of metoxile groups, refraction index and molecular weight.

Table 3

Characteristic of phenol fractions of beef smoke.

Fractions	Temperature	Pressure	Weight of fractions(%)	Specific weight	Content of metoxil	Refraction index	Molecular weight
1	58-64	4	6,01	1,1121	3,96	- x/	133
2	64-70	4	5,71	1,1117	13,2	- x/	123
3	70-76	4	5,62	1,0999	13,35	- x/	123
4	76-79	4	7,52	1,0985	9,96	1,5328	109
5	79-89	4	7,43	1,0967	9,18	1,5332	164
6	89-109	4	7,68	1,1224	12,6	1,5398	152
7	109-111	4	8,28	1,1833	30,4	1,5532	178
8	111-114	4	10,74	1,1872	33,2	1,5549	171
9	114-119	3	5,48	1,1827	31,4	1,5517	-
10	119-126	4	8,73	1,1652	27,9	1,5413	162
	126	Rest	1,56				
			6,56				
		Washed out	11,22				
		Total	92,64%				
		Losses	7,36%				

x/ mm of mercury column;
atmospheric pressure - 739 mm.

Table 3 shows that narrow phenol fractions differ from each other according to some indices and therefore are to differ according to chemical composition and their properties. Microbiological investigations showed that the fractions of the type 8, 9 and 10 had the strongest bactericide effect.

Antioxidant properties of the fractions strengthen as the molecular weight and boiling temperature grow (fig.3). While the antioxidant activity of the first fractions (table 4) is rather weak, that of the fractions, boiling at 119-126°C and 4 mm of the mercurium column (type 10), is no less than that of butyloxitoluene (table 4).

Organoleptic studies showed rather interesting results. It was found that while the fractions of the type 1,2,3 (table 3) had a "phenol-guaiacol" odour and the last fractions (6,7,8,9,10) had a specific phenol odour too, the middle fractions (4,5) had a different special odour, approaching to some degree to the smoky flavour.

Organoleptic assessment of sausages with different quantities of the fractions N 1-10 showed, that all the fractions except N 4 and N 5 gave the products an outside flavour.

Though the sausages with the phenol fractions N 4 and N 5 were not completely identical to those made according to the technology generally used, their taste qualities were found satisfactory. At the same time the flavour of nonsmoked sausages without these fractions was unpleasant.

These result seem to show, that in the beech smoke there are phenol compounds participating in the composition of the specific smoky flavour.

We tried to determine the qualitative composition of the phenol fractions by means of paper chromatography.

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Table 4

Antioxidant activity of phenol fractions of beech smoke

Fraction	Time of oxidation / Peroxide index						
I	75 / 0,027	120 / 0,1[illegible]7	1400 / 0,297				
IV	60 / 0,022	120 / 0,o41	180 / 0,507				
VI	60 / 0,o18	120 / 0,031	150 / 0,040	255 / 0,113	255 / 0,164		
VII	75 / 0,002	145 / 0,025	205 / 0,04	265 / 0,06	325 / 0,062	385 / 0,089	445 / 0,154
BHT x/	60 / 0,016	185 / 0,031	290 / 0,047	340 / 0,063	405 / 0,073	475 / 0,093	515 / 0,125
VIII	60 / 0,012	180 / 0,022	275 / 0,038	335 / 0,048	445 / 0,082	515 / 0,120	545 / 0,164
IX	60 / 0,015	195 / 0,027	315 / 0,047	375 / 0,055	465 / 0,104	495 / 0,126	525 / 0,182
X	60 / 0,0093	180 / 0,020	33o / 0,031	415 / 0,046	51o / 0,066	580 / 0,o85	[illegible] / 0,1[illegible]

x/ Butyloxitoluene (0,02% added to the fat) is a common antioxidant, used for comparison.

The chromatograms of all the 10 fractions showed 17 different stains, including those seen only in ultra-violet rays. The following substances were identified: phenol (carbolic acid), metacresol, guaiacol, methylguaiacol (fractions 1-5), pyrocatechol, methyl ethers of pyrogallol and its homologues (presumably, methyl and dimethyl ethers of methyl-, ethyl- and propylpyrogallol) (fractions 6-10). We did not succeed in separating methyl ethers of pyrogallol and its homologues, since they had a single stain.

The lack of witnesses prevented us from identifying some other compounds too.

Such phenols as ethyl - and propylguaiacol, orthocresol and α and β -naphthols were not found.

The identification of the rest of phenol compounds still remains the task for further investigations.

A white crystalline substance with a specific odour was isolated from the phenol fractions N 4 and N 5. The reaction with 2,6-dichloro-quinone-chlorimid did not show a blue coloration, typical for phenol compounds. The reaction with the chloride of iron gives a violet coloration, which disappears with the addition of saturated solution of sodium bicarbonate. This reaction is typical for methylcyclopentenolone. The same results were obtained with the pure reagent. The melting temperature of the isolated substance was $105,5^{\circ}$, that is the melting temperature of the pure methylcyclopentenolone.

The infra-red spectrometry was used for an additional study of this substance. It was found to be completely identical to methylcyclopentenolone (Fig.4).

The presence of this ~~[illegible]~~ ketone in phenol fractions is due to the fact that it exists in two tautomeric forms: keto-enol and diketone, which easily pass into each other.

The properties of methylcyclopentenolone in the enol form are similar to those of phenols(with alkalies it makes salts of the phenolate type; it dissolves easily in alcohol, ether etc). During the distillation of curing components of smoke it is isolated together with phenols.

Methylcyclopentenolone has a specific odour with a shade of dried mushrooms and, partly, roasted nuts. We believe, that methylcyclopentenolone is one of the components of smoke, which causes the formation of the tangy flavour.

Other ketones and aldehydes of smoke penetrating through the casings of sausage models, are: formaldehyde, acetic and butyric aldehydes, furfural, traces of vanillin, acetone and methylethylketone.

Using the method (24) we found, that phenols of smoke penetrate into the middle of dry sausages both in the process of smoke-curing and during the drying. They reach the middle of the sausage approximately 25-30 days after curing.

This fact indicates that the penetration of smoke-curing components into sausages in the process of curing is of diffusive nature. The rate of the penetration of the phenols through casings depends on the concentration of smoke, composition of minces and the material of casings. The rate of the penetration through natural casings is higher than through artificial.

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Conclusion

Practically all groups of organic substances of smoke penetrate through casings in the process of smoke-curing: phenols, acids, bases, neutral compounds, carbohydrates, hydrocarbons.

Phenols, especially their high-boiling fractions, possess the strongest antioxidant and bactericide properties.

The odour of such groups of substances as acids, hydrocarbons, bases and carbohydrates, is far from tangy flavour. A crystalline substance, identified as methylcyclopentenolone, was isolated from the phenol fractions boiling at, approximately 76-89^{o} and 4 mm of the mercurium column.

It is believed that methylcyclopentenolone and other neutral substances (furfural, alcohol), - probably some phenols too, - participate in the formation of the specific heady tangy flavour.

Perhaps the pungency and shade of smoke of this flavour are given by the compounds of formaldehyde and methyl alcohol type.

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SCHEME OF A GROUP ORGANIC ANALYSIS OF SMOKE-CURING COMPONENTS

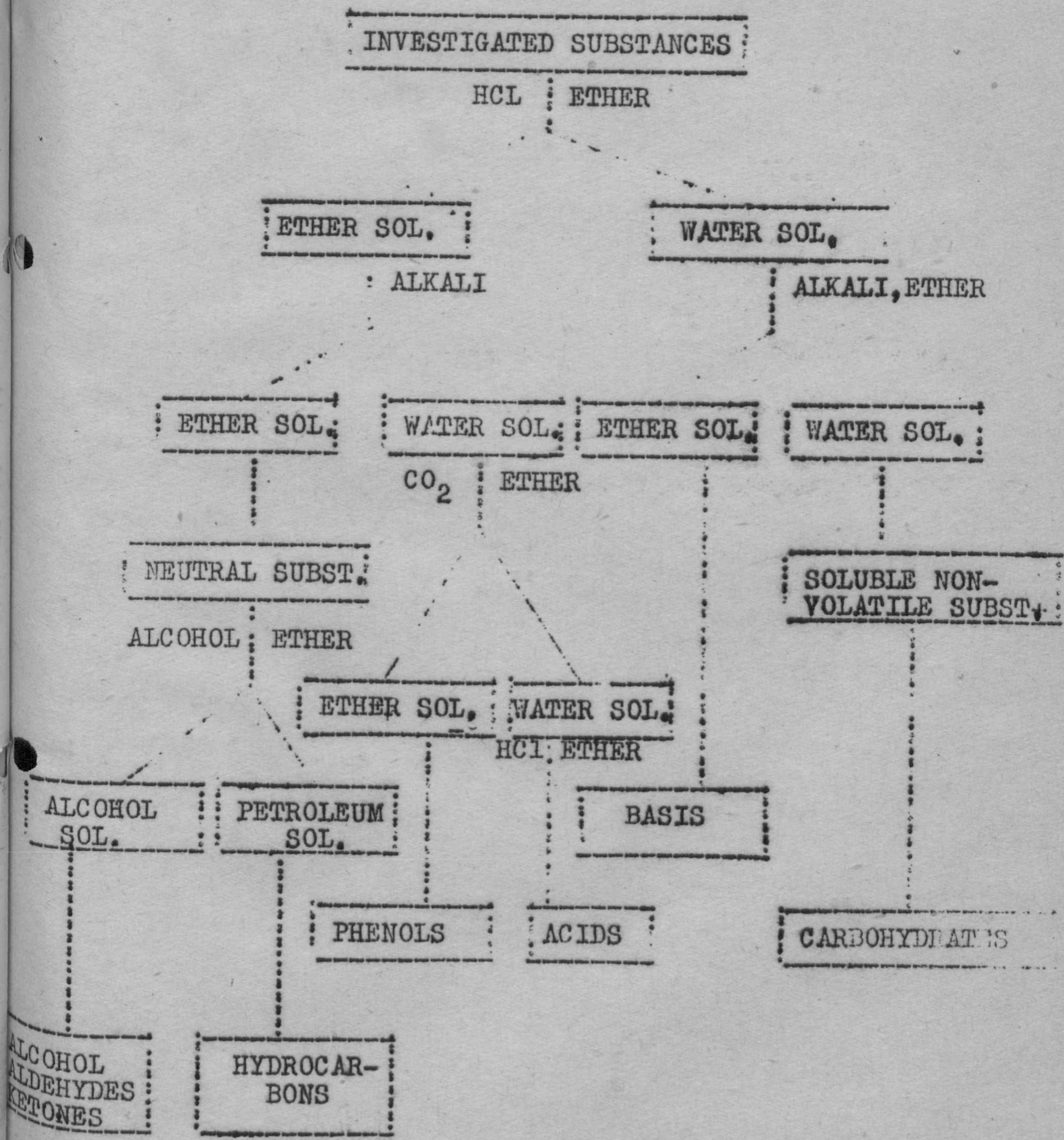

Fig. 1

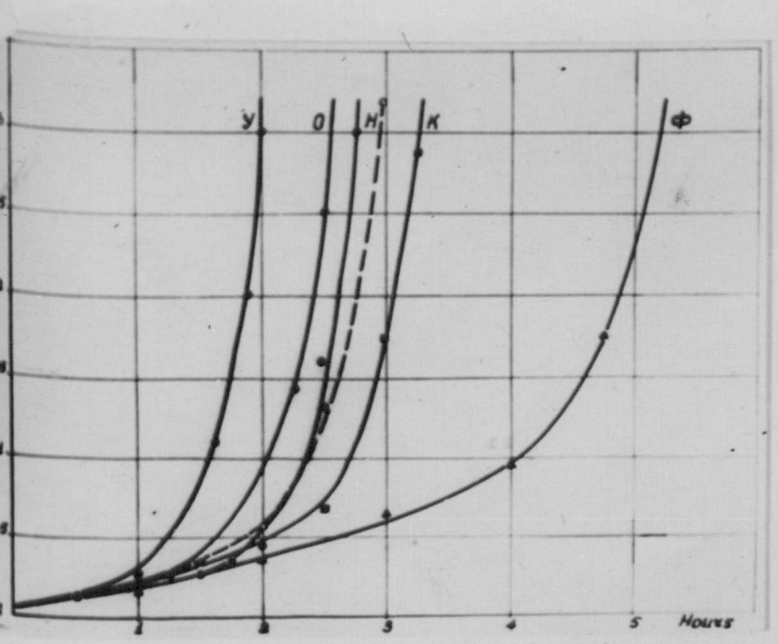

Fig 2 Curves of the oxidation of pork fat: H-neutral compounds; K-acids; Ф-phenols; У-carbohydrates; O-bases. Dotted line-control (primary pork fat only), t=110° C.

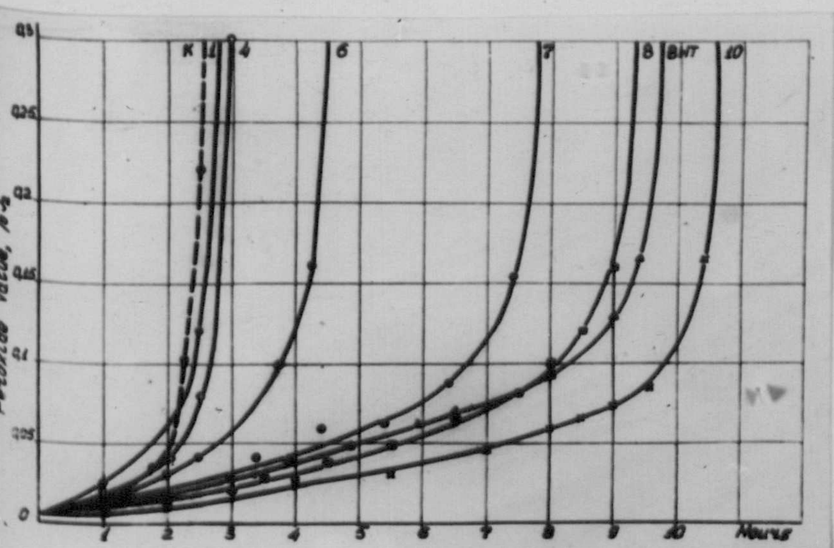

Fig. 3. Comparative antioxidant strength of phenol fractions: 1, 4, 6, 7, 8, 10 - fractions; BHT - butyloxitoluene; K-control (fat only).

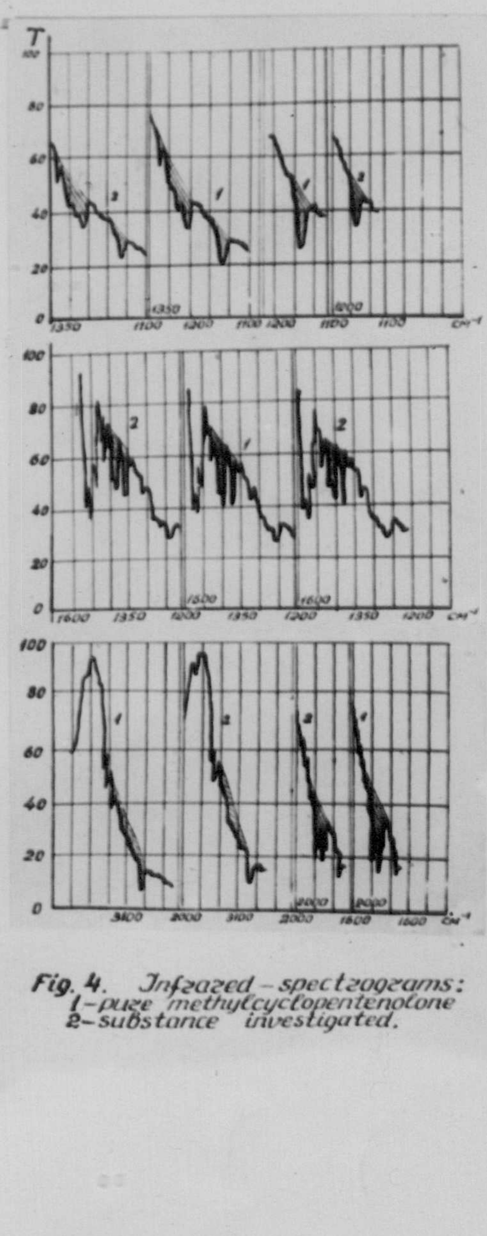

Fig. 4. Infrared-spectrograms: 1-pure methylcyclopentenolone 2-substance investigated.

SUMMARY OF THE PAPER "STUDY OF CURING COMPONENTS OF ...

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The article deals with the first results of the systematic esearch carried out by the All-Union Scientific Research Institute or Meat Industry (Moscow) with the aim of studying chemical and hysico-chemical problems of smoking, since the lack of this data mpedes further improvement of smoking technology.

The main groups of smoking components, i.e. components hich penetrate into the product and give it special odour, flavour d other properties, were isolated from beech smoke by the thods developed.

It was found that amongst these groups of smoking components acids, organic, bases, neutral substances (aldehydes, ketones, cohols), hydrocarbons, phenols, carbohydrate compounds/ phenols d acids are the most effective bactericides. Only high-boiling enol fractions were found to be strong antioxidant.

It is assumed on the grounds of organoleptic estimation at the smoke components, penetrating through casings and rticipating in the development of the tangy flavour, include enol compounds and neutral substances of the type of aromatic dehydes and ketones (furfurol, methylcyclopentenolone etc.).

The following smoking components were identified by methods organic analysis paper cromatography and infra-red spectrometry: enol (carbolic acid), metacresol, guaiacol, methyl-guaiacol, rocatechol, methyl ethers pyrogallol and its homologues, form-ldehyde, acetic and butyric aldehydes, furfural, traces of vanilla, etone, methylethylketone, methylcyclopentenolone.

UNTERSUCHUNG VON RÄUCHERBESTANDTEILEN DES RÄUCHERRAUCHS

(Zusammenfassung)

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In diesem Artikel werden die ersten Ergebnisse der in dem Allunions*Forschungsinstitut für Fleischwirtschaft (WNIIMP,Moskau) durchgeführten Untersuchungen, die die Klärung der chemischen und physico-chemischen Fragen der Räucherung zum Zweck haben, dargelegt.

Es wurden mit Hilfe der ausgearbeiteten Methodik aus dem Buchenrauch die Hauptarten der Räucherkomponente isoliert, d.h. solche Bestandteile die beim Räuchern in das Erzeugnis eindringend demselben spezifisches Aroma, Geschmack und andere Eigenschaffen verleihen.

Es wurde festgestellt dass unter den isolierten Arten von Räucherbestandteilen (Säuren , organischen Basen, neutralen Stoffen: Aldehyden, Ketonen, Alkoholen, Kohlenwasserstoffen, Phenolen, Kohlenhydratverbindungen) als stärkste Bakterizide Phenole und Säuren gelten können, Unter den Phenolfraktionen erwiesen sich nur die hochsiedenden als starke Antioxidantien.

Auf Grund der organoleptischen Beurteilung kann man annehmen, dass unter den durch die Wursthüllen eindringenden Räucherbestandteilen Phenolverbindungen und eine Art aromatischer Aldehyde und Ketone (Furfural, Methylcyklopentenolon u.a.) darstellende neutrale Stoffe an der Aromabildung teilnehmen.

Anwendung von Methoden der organischem Analyse, Papierchromatographie sowie Infrarotspektrometrie half uns folgende Räucherbestandteile zu identifizieren: Phenol (Carbolsäure), Metakresol, Guajakol, Methylguajakol, Pyrocatechusäure, Methylpyrogallussäure sowie Honologen der Pyrogallussäure, Formaldehyd, Essigsäure- und Buttersaurealdehyde, Furfural, Spuren von Vanilin, Aceton, Methyläthylketon, Methylcyklopentenolon.

SOMMAIRE DE L'ARTICLE "L'ETUDE DES COMPOSANTS DE FUMÉE".

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Dans cet article on expose les premiers résultats des recherches systematiques, réalisées par l'Institut de recherches scientifiques de l'Industrie des viandes de l'U.R.S.S. (Moscou) dans le but d'éclairer les questions chimiques, relatives au fumage; l'absence de renseignements de ce genre présente un obstacle sérieux pour l'amelioration de la technologie de la production des aliments fumés.

En se servant de la méthode élaborée on parvint à isoler de la fumeé de hêtre les groupes principaux des constituants de fumage, c'est a dire ceux, qui penètrent à l'intérieur du produit au cours du fumage et lui donnent l'odeur, spécifique, la saveur et autres propriétés.

On a constaté que parmi les groupes isolés de constituants principaux de fumage (acides, bases organiques, substances neutres: aldéhydes, cétones, alcools, hydrocarbures, phénols, hydrates de carbone) les phénols et les acides sont les antiseptides les plus effectifs.

Entre les fractions phénoliques ce ne sont que celles qui ont une haute température d'ébullition qui sont les antioxygenes les plus effectifs.

Se basant sur l'éxamen organoleptique on peut supposer, que de tous les constituants de la fumée, qui penètrent à travers l'enveloppe du saucisson seulement les composés phénoliques et les substances neutres du type des aldéhydes aromatiques et des cétones, contribuent à la formation de l'arome des produits fumés (furfurol, methylcyclopentenolone etc.).

A l'aide de l'analyse organique, de la chromatographie sur papier et de la spectrométrie infra-rouge on parvint à identifier les constituants de fumage suivants: phénol (acide phénique), métacresol, guayacol, méthylguayacol, pyrocatechine, esters méthyliques de pyrrogallol et de ses homologues, formaldéhyde, les aldéhydes acétique et butyrique, furfurol, les traces de vanilline, acetone, methyléthylcétone, methylcyclopentenolone.
