VOLATILE NITROGEN - COMPOUNDS AND DETERIORATION OF MEAT

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Introduction and general consideration. In meat research and particulary in meat trade there is some confusion as to the value of different methods for the judgment of keeping qualities of meat on one hand and as a criterion of sanitary quality of meat on the other.

Today we are all convinced of specific etiologies like enterotoxins or infective bacteria beeing the main causes for the symptomatology found in gastrointestinal irritation and that f.i. split proteins from "normal" putrefaction of meats seldom give rise to gastrointestinal upsets.

This is one side of the problem. Another, just as important economically, is the fact that meat in the state of putrefaction or microbiological or enzymochemical deterioration as a whole cannot be sold or used in the manufacture of most meat products.

This means that we need not only such methods which give us full information about meat from a public-health point of view, but also methods which as objectively as possible can inform us about the state of deterioration and so give us means for the testing of keeping qualities of meat and for the judgment of the best disposal of the meat in question. Sometimes it is of value to have a method enabling the food technologist to judge f.i. what influence different slaughtering methods will have on the keeping quality of the meat.

Detecting meat decompositions which might lead to food defects, by means of chemical methods, therefore is an interesting problem. But we have no comparison-standard. Bacteriological examinations are of little value, since the reason for the deterioration may be principally chemical in one case, mainly microbiological in another. Correlations are very seldom found between results of chemical tests and bacteriological examinations. This fact is, of course, a good reason for the view of many bacteriologically experienced scientists, that existing chemical methods are of little value compared to bacteriological examinations, so far as a judgment of meat from a public-health point of view is concerned but it is not relevant if we want to judge of deterioration as such.

Only organoleptic tests are today our tools for comparison. Everybody who has followed the results of a sufficient number of organoleptic tests by even very trained persons for years must regret this fact deeply.

Methods. Very many chemical methods for determination of, say, incipent putrefaction of meat have been published (1, 2) and it is very likely that several methods described are valuable for the special task here treated, namely objective detection of deterioration of meat without regard to public-health points of view. In our testing of well-known chemical methods, original and modified by us, during several years we have found the determination of volatile ammonia and ammonia compounds after treating of the sample with a very weak alkali worth more attention than it is usually paid by meat technologists.

According to Edelmann and coworkers (3) ammonia production does not afford one a criterion of sanitary quality of meat. Maybe this statement has discouraged food chemists from studying ammonia production as a measure of meat deterioration. There are later contributions on ammonia production in meat to be found in literature. As a rule organoleptic and bacteriological tests are used as comparison standard (4).

We have found a slightly modified distillation method of the type described in Allen's handbook (1) to be the most suited mothod for our purpose:

Twenty grm. of the well-hashed sample are weighed into a 1 l. distilling flask. 450 ml. of water and 10 grm. of freshly calcined, light magnesium oxide are added, and the whole mixed. The mixture is quickly brought to the boiling-point and the distillate collected in an Erlenmeyer-flask containing N/10 hydrochloric acid and cochineal indicator. Methyl red indicator can also be used. The distillation is carried out in 50 - 60 minutes and exactly 200 mls. are distilled over. The Erlenmeyer-flask is marked at the exact volume of 200 mls + volume of acid. Sticking of the material to the bottom of the flask and charring is prevented by careful distillation especially at the start.

When 200 mls have been distilled over the Erlenmeyer-flask is heated to boiling and boiled for 2 minutes with some small pieces of unglazed china. Then the flask is cooled by cold water and back titration carried out using N/10 sodium hydroxide.

A sharp titration point and exactly comparable results are obtained. It is well known that some meat bases, as well as some amino acids and acid amines will give some ammoniacal nitrogen by distillation (5) even when the weakest alkalies as magnesium oxide are used. This fact seems to be of no importance for the use of the method for the purpose here treated as will be seen from the results below.

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Results. In table 1 some measurements of volatile nitrogen compounds in meat from the same muscles of beef are collected. Samples were taken under sterile precautions as well as without sterile measures of precaution but in ordinary hygienic way in order to avoid an unusual microflora. Sample II S, taken under sterile precautions, remained steril during the whole time it was under investigation. Sample II I.S. which was not taken under sterile precautions but from the same muscle as sample II S showed (5/3) a total count on nutrient agar of 14 000 000 bacteria/gm together with a low count of anaerobes on liver extract. Samples I A and I B were taken from the same muscles of other beef carcasses, as samples II immediately after slaughter. They were collected in the abattoir of Stockholm. Though no true sterile precautions were taken sample A was collected in a most aseptic way where-as sample B was collected with only such precautions taken which would avoid other infection than with ordinary air-born flora of the abattoir. At the end of the investigation (5/3) sample I A showed a bacterial content of 10 000 000 and sample I B 22 000 000 pr gm.

Table 1.

Volatile nitrogen compounds in sterile and unsterile samples of the same beef-muscles.

	I A				I B			
Date	Days after slaughter	Volatile N mgs %	Total N %	Volatile N in % of total N	Days after slaughter	Volatile N mgs %	Total N %	Volatile N in % of total N
4.12	2	15.1	3.58	.42	2	17.3	3.52	.49
4.18	8	19.6	3.64	.54	8	22.4	3.61	.62
4.20	10	20.3	3.52	.58	10	19.6	3.50	.56
5.3	23	30.1	3.71	.81	23	39.2	3.52	1.11

	II S.				II I.S.			
Date	Days after slaughter	Volatile N mgs %	Total N %	Volatile N in % of total N	Days after slaughter	Volatile N mgs %	Total N %	Volatile N in % of total N
4.12								
4.18					1	22.4	3.59	.62
4.20	3	18.2	3.62	.50	3	18.9	3.67	.51
5.3	16	28.0	3.82	.73	16	25.2	3.68	.68

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Organoleptically sample I B showed decay on 5.3

Independantly of the microbiological state of the meat, decay will be indicated by a certain quantity of volatile nitrogen compounds, determined as above. Determinations carried out on samples of meat from individual cattle give results as the following, Table 2 - 4.

Table 2.

Volatile nitrogen compounds in beef. Gracilis-muscle from 11 years old cow, Class II. The ground meat stored at 4° C. Protein calculated as Nx 6.25: 21.04 %.

Days after slaughter	Volatile N % of sample	Volatile N % of protein	Organoleptic test
0	0.0161	0.0765	
3	0.0191	0.0907	
7	0.0188	0.0893	
10	0.0169	0.0803	
14	0.0243	0.1154	Slight decay
17	0.0424	0.2015	

Table 3.

Volatile nitrogen compounds in beef. Gracilis-muscle from 5 years old cow classified 1-, killed 4.9.1956 at 750 a.m. in the abattoir of Stockholm.
Protein: Nx 6.25 = 21.5 %.

Date	Volatile N % of sample	Volatile N % of protein	pH
4.9	0.0165	0.0767	6.2
4.10	0.0192	0.0893	5.2
4.11	0.0187	0.0869	5.25
4.12	0.0177	0.0823	5.2
4.13	0.0162	0.0753	5.25
4.14	0.0165	0.0767	5.25
4.16	0.0152	0.0711	5.25
4.17	0.0156	0.0725	5.2
4.19	0.0167	0.0776	5.1
4.21	0.0177	0.0823	5.1
4.23	0.0186	0.0865	5.1
4.25	0.0221	0.1030	5.3

On 4.23 organoleptic tests gave indication, characterized as faintly sour.

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Table 4.

Volatile nitrogen compounds in pork. Gracilis-muscle from 75 kgs carcass of pig slaughtered in the Tranås abattoir on 1.11 and kept under refrigeration. until sample was taken on 1.20. Ground sample kept at + 4° C. Protein:N.6.25 = 18,8 % as determined on 1.20.

Date	Volatile N % of sample	Volatile N % of protein
1.20	0.0162	0.0861
1.23	0.0209	0.1111
1.27	0.0197	0.1047
1.30	0.0302	0.1606
2.3	0.0636	0.3882

Organoleptic test indicated decay on 2.3.

More than 1000 determinations show little difference between contents of volatile nitrogen in different muscles of the same carcass. Material will be published elsewhere.

During cold storage meat will deteriorate slowly even at low temperatures. Table 5 gives results of some determinations of volatile nitrogen compounds in pork stored at - 20° C for 8 months.

Table 5.

Pork stored at -20° C for 8 months. Muscles from different parts of carcass collected to general sample.

Sample collected from muscles of	pH	Volatile N % of sample
Sow	5.8	0.0296
Pork	5.4	0.0304
"	6.2	0.0280
Sow	6.1	0.0329
"	6.1	0.0312

All pH-measurements were made with glass electrode.

Theory of meat deterioration, discussions of results and conclusion. The economically most important type of meat deterioration is an enzymatic decomposition of protein and other nitrogen compounds of the muscle.

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This type of decay will take place whether microorganisms are present or not. The velocity of the deterioration processes will depend on several factors e.g. quantity and activity of originally present proteolytic enzyms of the meat, quantity and activity of proteolytic enzyms produced by microorganisms present, resistance of meat tissue to decomposition. Even a sterile sample of meat will change into an evil-smelling, viscous mass, provided enzyms have not been destroyed through heating or other treatment.

During decomposition of the kind described volatile nitrogen compounds will be formed. The quantity of these compounds need not have any correlation whatsoever to bacterial count but gives, as a rule, a good picture of what is usually and practically called deterioration. Nor is there any correlation between sanitary quality of meat on one side and deterioration on the other. The quantity of volatile nitrogen compounds is therefore of no importance for the judgment of meat from a public-health point of view.

For the most important kinds of meat e.g. beef and pork the quantity of volatile nitrogen compounds determined as described above is low and constant immediately after slaughter. If calculated as per cent of total nitrogen or protein there is only a slight change of the quantity of volatile nitrogen compounds during the first days of storage if proper chilling and refrigeration is applied. When the meat is stored for a sufficient time, and even if kept at very low temperatures the quantity of volatile nitrogen compounds will increase. When organoleptic tests are positive, the increase is marked.

Literature.

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