

H/3 : PUOLANNE E. : DER EINFLUSS VON PH-WERT, TEMPERATUR UN
SALZGEHALT AUF DIE NITRATREDUKTASEAKTIVITÄT
DES MIKROKOKKENSTAMMES M₁₁₁

QUESTION DE M. HOFMANN :

Würden Sie bitte noch einmal darauf hinweisen, auf welche Veränderungen sich die beiden Kurvenzüge beziehen, die in der von Ihnen gezeigten graphischen Darstellung Nr. 1 aufgezeichnet sind ?

REPONSE :

Die obere Linie ist von meinen eigenen Untersuchungen (Tabelle 1 und Abbildung 1) und die untere ist von Niinivaara.

QUESTION DE M. MOERMAN :

Wäre es nicht besser, die Nitratkonzentration auf die logarithmische Skala zu zeichnen ? Vielleicht bekommt man dann eine gerade Linie.

REPONSE :

Wenn man die Konzentration in einer logarithmischen Skala und die Geschwindigkeit in einer arithmetischen Skala zeichnet, bekommt man eine gerade Linie. In der Enzymchemie ist es gewöhnlicher, die Konzentration in einer arithmetischen Skala und die Geschwindigkeit in einer reziproken Skala zu zeichnen. Dann bekommt man eine gerade Linie, wovon man auch die Michaelis-Menten Koefficient K_m auslösen kann. Aber für einen Nicht Biochemiker ist eine normale arithmetische Skala leichter zu verstehen.

QUESTION DE M. VENDEUVRE :

Rohwurstbräte wurden normalerweise vor dem Abfüllen in den Darm einer 12 bis 24-stündiger Reifung bei 2°C bis 9°C unterzogen. Ist dieses Verfahren Ihres Erachtens noch begründet, weil Sie eben gezeigt haben, dass die Aktivität der Mikrokokkenstämme unter 10°C stark eingeschränkt ist ?

REPONSE :

Ich weiss nicht, warum man in Deutschland die Rohwurstbräte Würste so kurz bei 2 - 9°C unterzeigen. In Finnland werden die Würste ungefähr eine Woche bei 8°C gereift, und die Mikrokokken haben Zeit genug, Nitrat zu reduzieren. 12 bis 24 St. ist nach meiner Meinung zu wenig für genügende Nitratkonzentration, aber natürlich wird Nitrat nach Erhöhung der Temperatur mehr reduziert.

Reduktion

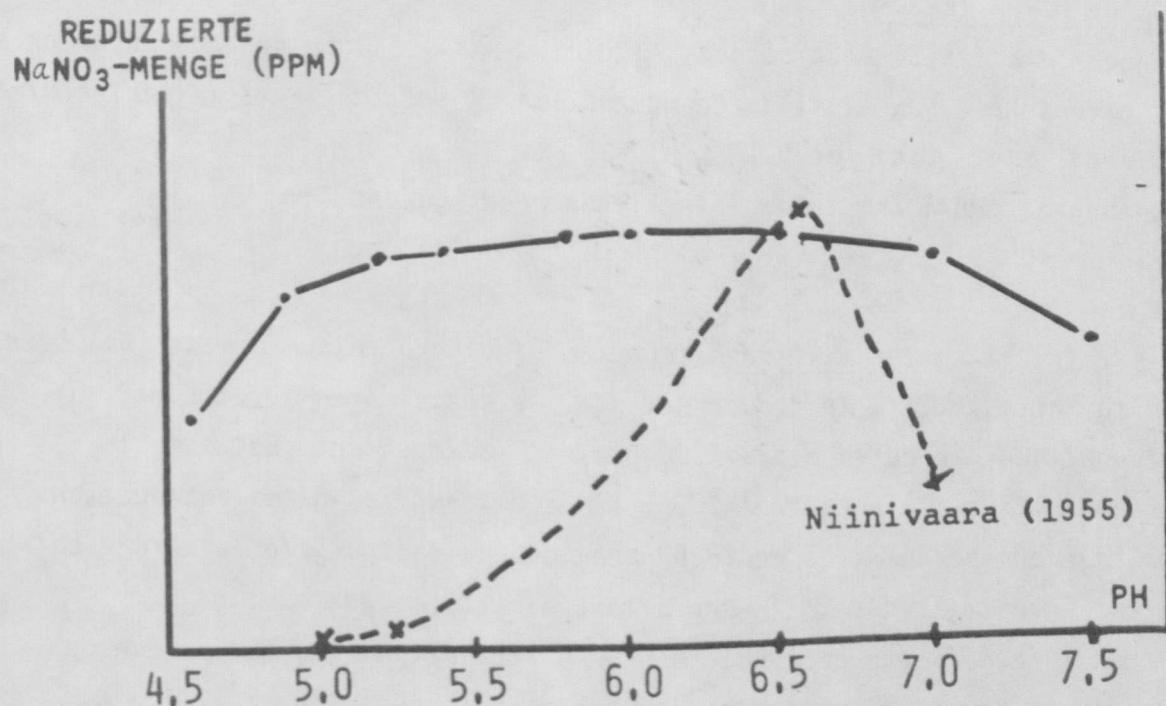


ABB. I. DER EINFLUSS DES PH-WERTES AUF DIE NITRATREDUKTASE-AKTIVITÄT DES MIKROKOKKENSTAMMES M_{III} VERGLICHEN MIT NIINIVAARA (1955) (X—X—X).

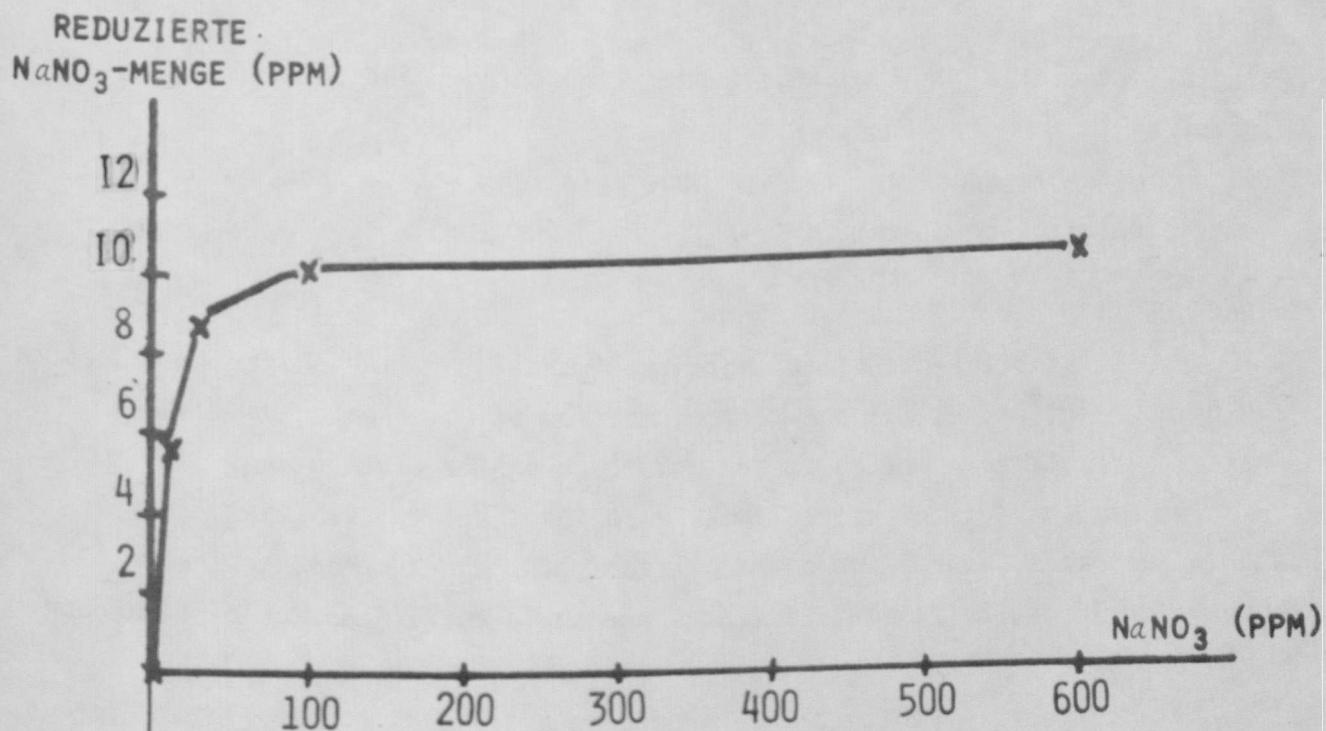


ABB. 2. DER EINFLUSS DER SUBSTRATKONZENTRATION AUF DIE GESCHWINDIGKEIT DER NITRATREDUKTION.

H/4 : GARDNER : THE OCCURENCE OF VIBRIO IN THE MICROFLORA OF WILTSHIRE CURED BACON SIDES.

QUESTION de M. PATTERSON :

You suggest that these vibrios are associated with certain defects in bacon. To what extent have you studied the mechanisms of spoilage - is it proteolytic, lypolitic or saccharolyptic ? Can you suggest control methods (other than good hygiene) ?

REPONSE :

Our main interest in the in vivo metabolism of vibrio in cured meats has been related to their ability to reduce nitrate. As there is an upper limit for permitted nitrite in cured meats (200 ppm) it is important that a meat cannot attain levels higher than 200 ppm as a result of nitrate reduction. The spoilage characteristics could be regarded as proteolytic in nature but I have not investigated this in any detail as yet. As for control , prevention I feel is the best but in brines filtration or dilution can be used to reduce levels of contamination and as they are very sensitive to heat mild pasteurization treatments can be carried out on the cured meat.

QUESTION DE M. RIORDAN :

Three short questions :

1. In table 3 of your paper we note very high vibrio counts for weeks 4 - 5 - 6. Were these high counts due to washing matured sides with brine ?
2. In factories 5 and 7 was there poor temperature control at the time of sampling ?
3. How often in your experience of bacon factory brines should the immersion brine be discarded ?

REPONSE :

1. Yes, I established that in factories 5 and 7 the sudden increases in Vibrio in the brines was due to the addition of brine used for washing sides with slime formation to the main bulk of the cover brine.
2. The reasons for the necessity to wash the sides in the first place at both factories 5 and 7 was due to excessively high temperatures and humidity in the cellar which allowed the rapid multiplication of Vibrio on the sides.
3. With respect to Vibrio brines should be discarded when the count exceeds 100,00 /ml but it is essential to trace and eliminate the cause of such high vibrio counts.

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QUESTION DE M. BARNES :

In recent times *Vibrio parahaemolyticus* has been identified as an organism causing food poisoning. e.g. *Proteus* have been incriminated in one reported case as cause of this type of food poisoning. Has M. GARDNER with his special interest *Vibrio* species any comment to make on this ?

REPORSE :

The bacon Vibrios may in a taxonomic sense have some characters in common with *Vibrio parahaemolyticus* but they are different in many respects. Work on the pathogenicity of *Vibrio costiculus* the brine vibrios, has shown that there is no potential public health danger from these organisms.

QUESTION DE M. PIVNICK :

Have attempts been made to add vibrios isolated from spoiled product to fresh product sterilized by irradiation in an attempt to determine if the organism actually causes the spoilage.

REPORSE :

These experiments have been done not with bacon sterilized by irradiation but with bacon obtained by aseptic removal from large pieces. I recovered the culture inoculated in a pure state and was quite satisfied that it can cause spoilage.

QUESTION DE M. NICKELS :

Have any tests (trial) been conducted with pure cultures of *Vibrio* strains on bacon ? What where the results ? (slime, odour, discolouration)

Thank you.

REPORSE :

I have carried out some pure culture work with *Vibrio* on bacon and they have the ability to produce slime, off-odour and discolourations. The-off-odours are of a putrefactive nature and the discolourations are fiery or bright red in nature.

QUESTION DE M. DEVEREUX :

Regarding the different morphological appearance of the *Vibrio* under different conditions do you think that it is the same strain of *Vibrio* in all cases or are there various strains involved ?

REPORSE :

Preliminary examination indicates that in the bacon *Vibrio* group there are a number of species sub-species or types. I have as yet not associated different types with different ecological environments.

Have you isolated and identified different Vibrio strains from brines.
If you have, have you observed all vibrios harmful?

REPONSE :

The technique of enumerating the bacon Vibrios from brines is specific for this group only. The brine strains used widely in Europe will not grow on the selective medium, so although they are very useful in the latter circumstance the bacon types I feel have to be regarded as important spoilage species in the final cured meat.

QUESTION DE M. HOFMANN :

Sie erwähnen einerseits, dass Vibrio zur Entwicklung Salz benötigen, andererseits aber sich in Gegenwart von Salz nicht entwickeln. Ist das nicht ein Widerspruch, oder habe ich Sie falsch verstanden?

REPONSE :

As I have stated the bacon Vibrios require salt for growth (minimum 0.5 - 1.0%). The upper limite of tolerance is around 10-12 % NaCl in the medium. So, in relation to mixture curing brines (25 - 27 % NaCl) there is no doubt that growth is inhibited at these high levels.

H/5 : M. PIVNICK H. ET AL : A RATIONALE FOR THE SAFETY OF CANNED SHELF-STABLE CURED MEAT : PROTECTION = DESTRUCTION + INHIBITION.

QUESTION DE M. RALPH :

Can the author clearly define "Destruction" and "Inhibition"? Is the effect of NaCl, NO₂ etc... during heating the former or the latter?

REPONSE :

At levels of salt and nitrite used in canned cured shelf stable meat, these additives do not increase the killing activity of heat, and I do not believe that they increase the damage to the spores that survive the heating process. If one increases the concentrations of salt and nitrite considerably above that usual industrially, then one does find that these additives increase the killing activity of heat.

QUESTION DE M. HOFMANN :

Are you of the opinion that nitrites are absolutely necessary for inhibiting C. botulinum significantly ?

Have you tried to find an equation to include all seven inhibitory factors about which you spoke ?

REONSE :

1. It depends on the product. For canned shelf stable meats, one might consider higher salt concentrations, more heat but less finished quality, etc... However, in pasteurized canned meats, because they are frequently not held under refrigeration, the removal or decrease of nitrite would probably level eventually to some outbreaks of botulism.

2. The equation presented would be useful for all inhibitory factors. One can determine the effect of each individual inhibitor by changing the experimental conditions, but one then is left at guessing whether the individual inhibitors act only on their own, or whether there is a synergistic effect.

QUESTION DE M. FROUIN :

Pouvez-vous nous apporter quelques précisions sur l'action des peroxydes sur C. Botulinum ?

REONSE :

The oxides and peroxides of fatty acids do not prevent germination of spores that survive heating, but they do prevent outgrowth from the spores. It has been standard practice for many years, when enumerating spores of C. Botulinum, to add starch TO THE BACTERIOLOGICAL MEDIUM USED FOR THE ENUMERATION. The starch adsorbs the fatty acid peroxides and negates their inhibitory activity. If starch is present in the meat product it would probably negate the inhibitory effect of fatty acid peroxides, if they should be present. Possibly also, meat proteins may negate the inhibitory of fatty acid peroxides ; it is known that undenatured albumin adsorb fatty acids.

QUESTION DE M. NERAAL :

Which effect have polyphosphates on the growth of C. Botulinum, and how pH-dependent is this effect ?

REPONSE :

I can not comment on the inhibitory effect of polyphosphates, per se, on C. Botulinum. However, since the polyphosphates increase the pH, they decrease the inhibitory effect of the nitrite. Nitrite decreases in the product during storage and, therefore, this combined pH nitrite effect probably decreases with time.

QUESTION DE M. HUGHES :

Will it be possible to apply this approach to pasteurized canned cured meats ?

REPONSE :

Probably not, if one is interested in the part played by destruction and damage of surviving spores caused by the heating process. However, one could apply the principles with respect the inhibition to quantify the protection given to a pasteurized cured meat. This protection would result from salt and nitrite level, pH , temperature of storage, etc...

QUESTION DE M. GEORGALA :

1. Is Dr. PIVNICKS calculation influenced greatly by using different strains of clostridium botulinum ?
2. What is a safe level for the "protection factor" ?
3. Is the approach described by Dr. PIVNICK coming into use in Canadian meat industry ?

REPONSE :

1. We used 10 strains. I believe that at least several strains of type A and heat resistant type B should be mixed. Whether the use of 10 trains in a mixture is worth the work, I cannot answer, but certainly one should use more than a single strain of type A and type B. For this type of experiments, the strains should be mixed rather than used singly in the meat.
2. I dont think that anyone wants to publicly state what a safe level of Protection Factor is, because the history of the product with respect to botulism is so good that industrial experience along with some government control by Departments of Agriculture, advice by various industrial organizations etc... has sufficed to the present. However, as technological changes occur, and there is no history to apply, one should do the experiments necessary to prove that the new technology does not give a less safe product than the technology that is replace.
3. The Canadian meat industries have a history of safe products, but I do not know whether or not the concept is coming into use throughout the industry.

QUESTION DE M. ROSKIS :

1. M. PIVNICK peut-il ,à titre d'orientation générale, nous préciser les modalités d'application de l'équation qu'il suggère dans les conditions pratiques, par exemple d'un jambon pasteurisé ?

2. Observations d'ordre général :

Sur le plan de la sécurité (salubrité) des saucissons de viande, il y a un autre problème qui a pris dernièrement des dimensions d'une actualité internationale : celui des nitrosamines et l'on pourrait même regretter que ce problème n'ait pas été porté à l'ordre du jour de la présente réunion. Ne serait-il pas intéressant d'organiser une table ronde sur ce sujet, pour avoir une idée sur l'état actuel des travaux en la matière et l'importance qu'il y a lieu de lui accorder ?

REPONSE :

1. The experiments of CHRISTIANSEN et al. on pasteurized hams published in Appl Microbiol. in 1973 give some very practical demonstrations of the importance of nitrite in pasteurized canned hams and, at the same time, the very small effect of nitrate in such products. However, it must be emphasized that the pasteurization is not likely to kill many spores nor to damage the spores to any great extent that would make them more susceptible to the inhibitors.

2. The matter of nitrites and nitrosamines, in the context of safety of cured meat, will be discussed in ZEIST, the Netherlands, during the week of Sept. 10 at 13 a conference called specifically for this purpose.