Influence of Storage, Curing and Heating on the Electrophoretic Behaviour of Sarcoplasmic Proteins.

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SUMMARY: The influence of storage, salting, curing and heating on the electrophoretic behaviour was investigated regarding to electrophoretic meat species identification of raw and processed material. Materials under investigation were raw beef and pork stored at different temperatures, raw and heated beef and pork salted and cured with nitrite. Protein separation was carried out by isoelectric focusing and zone electrophoresis total sarcoplasmic proteins, myoglobin and esterases were visualized by different staining methods.

The protein patterns are only influenced by storing if the sensoric quality of the meat was impaired. The addition of salt with or without nitrite results in a decreasing or a fading of some sarcoplasmic proteins. This effect could also be recognized in samples heated to 65° and 70°C, heating to 75°, 80° and 100°C reduce or neutralize the described influence of salt. Myoglobin and esterase bands of raw samples are not influenced by curing, but heating of cured meat effects a reduction of the intensity of the myoglobin bands in comparison to samples without addition of nitrite.

INTRODUCTION: The origin of the animal, the kind of feed, the different muscles of the animal, the age of the meat and the storing conditions are possible factors which can influence the electrophoretoic behaviour of the sarcoplasmic proteins (HOFMANN, 1986). So, meat species identification based on electrofocused sarcoplasmic proteins can only be carried out by comparing the unknown sample with references applied on the gel side by side (BAUER, 1990). Using only the myoglobin bands for detection (BAUER and HOFMANN, 1987 a-c) meat from closely related animals cannot be distinguished.

The influence of heating on the electrophoretic behaviour and the solubility of sarcoplasmic proteins and myoglobin was desribed and discussed previously (BAUER and HOFMANN, 1990 a,b). In addition to heating meat and meat products are salted or cured for human nutrition. Comparing sarcoplasma protein patterns of heated meat and sausages some differences could be obtained (BAUER and HOFMANN, 1989). Also in this case the myoglobin method cannot be used in general ((BAUER and HOFMANN 1987 c, 1989)

The objective of this work was to investigate the influence of storage, salting, curing and heating on the electrophoretic behaviour of sarcoplasmic proteins in order to test the suitability of electrophoretic methods for reliable species identification in meat and meat products.

MATERIALS and METHODS:

Materials: Raw beef and pork stored for 18 resp. 14 days at 5°C and 20°C. Minced beef and pork with addition of 0.5, 1, 2, and 4% NaCl or NPS (NaCl containing 0.5 to 0.6% sodium nitrite). Heating to 65°, 70°, 75°, 80° and 100°C for 15 minutes

Extraction: see BAUER and HOFMANN (1987a, 1989); salted samples were dialysed.

Electrophoresis: Zone elctrophoresis in 7.5% polyacrylamide gel and isoelectric focusing (pH gradient 3-10) was used according to BAUER and KELNER (1989)

Staining: Sarcoplasmic proteins by Coomassie Brilliant Blue R-250 (NEUHOFF, 1985), Serva Violet 17 (PATESTOS et al., 1988) and silver staining (HEUKESHOVEN and DERNICK, 1985); myoglobin staining (BAUER and HOFMANN, 1987); esterase staining (BGA, 1988; BAUER and KELNER, 1989)

RESULTS and DISCUSSION:

Storing meat for 18 (beef) or 14 (pork) days at 5°C doesn't influence the electrophoretic behaviour of sarcoplasmic proteins independent from the electrophoresis technique. Differences in the protein pattern could not be obtained neither using IEF (Fig.1, left gel) nor zone electropheresis (Fig.2). Also the myoglobin and esterase bands (Fig.1, right gel) are not influenced by storage at 5°C. Changes such as decreasing of the intensity of the myoglobin bands, disappearing of some sarcoplasmic poteins (Fig.1, left gel; Fig. 2) and of the acidic esterases (Fig. 1, right gel) could be obtained by IEF and zone electrophoresis at the earliest after 6 days of storage at 20°C . These changes occur simultaneoulsy with the senoric spoilage of the meat.

Salting and curing of raw meat effects a decreasing of the intensity or disappearing of the electrophoretically separated sarcoplasmic proteins in the neutral region with increasing concentrations of salt. Fig.3 shows a comparison between unsalted and salted meat regarding to the differences of sarcoplasmic protein pattern. In contrast to this the myoglobin and esterase bands visualized specifically are not influenced by the addtion of salt and curing agent.

Compared to raw samples heating salted or cured meat to 65 or 70°C intensifies the impact of salt to the decreasing intensity of the protein patterns (Compare Fig.3 and Fig.4, lower gel). The sarcoplasmic protein patterns of samples heated over 65°C can only detected by the silver staining (Fig.4, lower gel). Also the intensity of the myoglobin bands visualized using their peroxidase activity is reduced in contrast to raw samples (Fig.4, upper gel). Esterases cannot be detected after heating of meat. The comparison of heated samples with and without the addition of nitrite shows that curing effects an increasing of the specific stained myoglobin bands. This effect is caused by dissociation of dinitrosoheme from the globin during heating of nitrosomyoglobin (LEE, 1976). The described influence of salt and curing agent on the electrophoretic behavior of the sarcoplasmic proteins is reduced or extinguished

Figure 1: IEF of sarcoplasmic proteins stained by Coomassie Blue (left gel) and specific stained esterases (right gel) of pork stored under different conditions; cathode on the top.

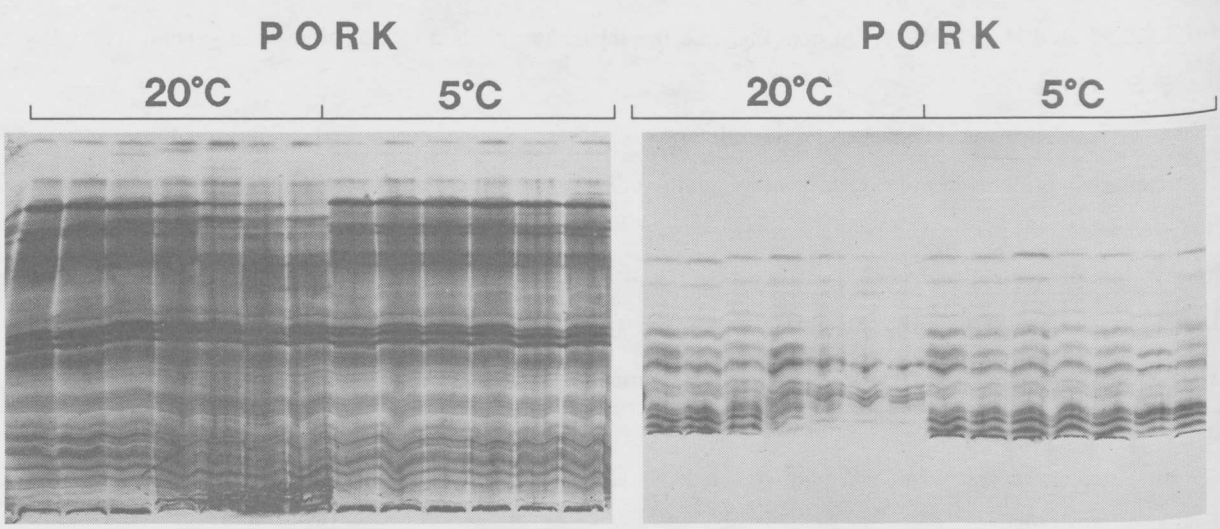


Figure 2: Zone electrophresis of sarcoplasmic proteins stained by Coomassie blue of pork stored under different conditions.; cathode on the top.

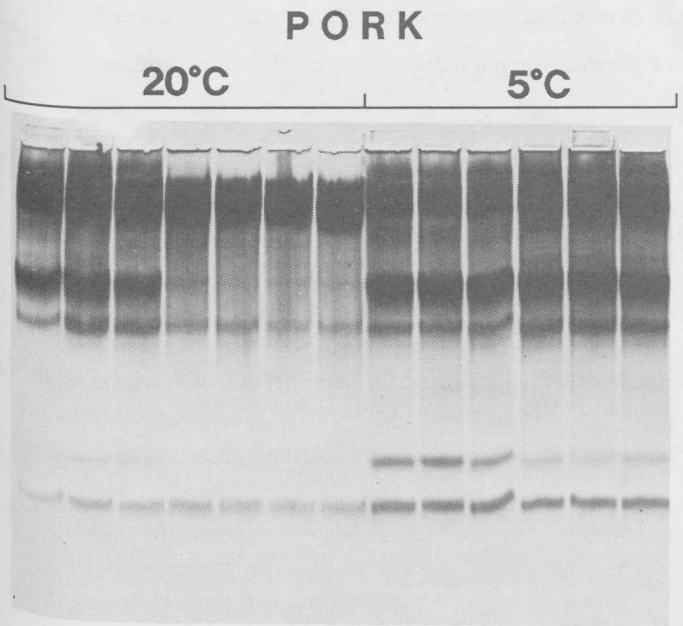


Figure 3: IEF of sarcoplasmic proteins from salted and unsalted pork; cathode on the top.

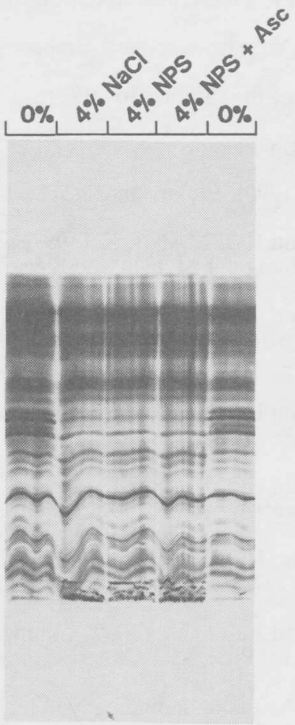
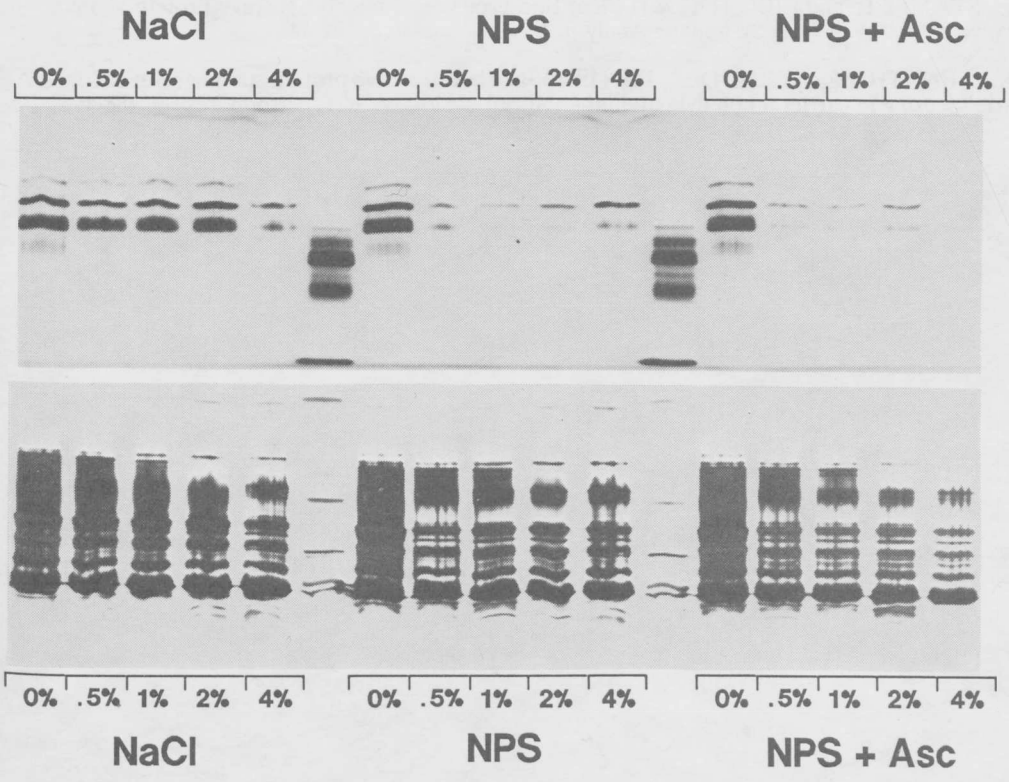


Figure 4: IEF of heated and salted pork using a "triple eletrofocusing cell"; cathode in the middle; specific staining of the myoglobi (upper part) and silver staining (lower part).



by heating the meat to 75, 80 or 100°C. As recommanded by BAUER and HOFMANN (1989) for reliable identification of heated meat a second heating of the samples to 80°C also compensates differences of the protein pattern caused by salting and curing.

CONCLUSIONS: Isoelectric focusing and zone electrophoresis in polycrylamide gels are suitable methods for meat species identification of raw meat independent on the age of the meat as long as the senoric quality is not insured. Changes in the protein patterns appear at first from spoiled samples. Heating and salting or curing of meat influences the pattern of sarcoplasmic proteins. A second heating of the samples to 80°C results in protein pattern which are not influenced by conditions of production.

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