Physicochemical characteristics of red pigment in Parma ham

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Background

Parma ham is a traditional procuit ham of Italy, whose production is limited to the Parma region. This ham is made only with pork (thigh) and salt taken from the Mediterranean sea. Its characteristic red color is quite stably maintained with no need for nitrite and/or nitrate addition. This pigment is easily extracted with acetone, and has been identified as a new myoglobin derivative, based on the results of optical absorption and ESR analysis[1,2]. The structure and properties of this pigment remain to be determined. The red color of Parma ham may possibly be due to the action of certain bacteria (staphylococci).

Objectives

The pigment of Parma ham differs from that, nitrosomyoglobin (NOMb), of ordinary cured meat products. The authors are presently conducting research to determine the structure of this new myoglobin (Mb) derivative. To reconfirm that this pigment differs from NOMb in structure and carry out the isolation and identification of this pigment, the physicochemical properties of the red pigment in Parma ham were examined in this study and compared with those of other Mb derivatives whose red color is due to oxymyoglobin (O_2Mb) and NOMb.

Methods

A water extract of Parma ham was prepared as follows: 1,000ml sterilized water were added to 50g ham and homogenized. At 5±2℃, the homogenate was centrifuged at 10,000×g for 10min and filtered with a series of Whatman GF/A and Toyo No.6 to obtain a clear extract. The absorption spectra of the final filtrate were recorded at 350~650nm.

Exp.1: The effects of pH on the absorption spectra were studied subsequent to the addition of NaOH and HCl to the water extract from the ham sample. Exp.2: The sterile water extract of Parma ham was obtained using a membrane filter (pore size: 0.02 μm) and kept in a sterilized test tube for 7 days at low (5±2℃)or room temperature (20±2℃) under conditions of light exposure or darkness. Optical absorption was measured for assessment of pigment discoloration. O_2Mb was extracted from fresh pork with 2 vol. distilled water and the supernatant after homogenization and centrifugation served as the O_2Mb solution. A small amount of sodium dithionite was added to a portion of the O_2Mb solution which was then kept at low temperature for 2 days. Nitric oxide (NO) gas was prepared by reaction of sodium ascorbate with sodium nitrite and bubbled through the meat extract. pH of the extract was adjusted with NaOH to 6.5 for use as the NOMb solution. Exp.3: To the water extract and each of the O_2Mb and NOMb solutions, ferricyanide was added. Sodium dithionite was added only to the water extract after it had been flushed with NO gas. At 30min, the absorption spectrum was recorded between 350 and 650nm so as to determine whether NOMb had been formed. Exp.4: Water extract of Parma ham and the two solutions were each heated at 40~90℃ for 30min followed by measurement of the absorption spectrum of each. The heated extract was filtered under sterile conditions. The filtrate was kept in a sterilized test tube at 20℃ for 5 days under a regime of light and darkness and examination was made to assess pigment stability.

Results and discussion

Exp.1: Fig. 1 shows the spectra of the water extract at 30 min following pH adjustment. A single absorption peak was noted at 423nm in the Soret band and 2 peaks at visible wavelengths of 549 and 587nm. The red pigment showed no change at pH 6~10. At pH 5.5, the red pigment started to precipitate and increasingly more

so as pH became more acidic. No typical absorption peaks for metmyoglobin (MetMb) could be seen at 505 and 630nm at any pH. MetMb formation from Parma ham pigment is thus shown not to occur at either acidic or alkaline pH. Exp.2: Fig. 2 shows the effects of light and temperature on red pigment stability of the water extract after being stored for 7 days. The red pigment was stable in the dark. Discoloration due to light exposure was evident after the first day of storage at either low or room temperature, particularly so at the latter. Optical peak absorption decreased for the O_2Mb and NOMb solutions even at 1 day of storage and continued to do so throughout storage. O_2Mb could be extracted from fresh pork and NOMb was prepared by reduction of Mb using sodium dithionite and NO gas bubbling. Exp.3: Fig.3 shows the effects of ferricyanide on the Mb derivatives. Ferricyanide is a strong oxidizing agent and MetMb can be easily obtained from meat or Mb model solution[3] using this compound. O_2Mb and NOMb were oxidized by ferrycianide to MetMb, but the spectrum showed no change in the Parma ham. The pigment of this ham was not affected by the sodium dithionite and NO gas flushing. Exp.4: For the water extract, absorption of the spectral peaks decreased, though the spectrum of this pigment was maintained essentially during heating, in contrast to those of O_2Mb and NOMb; precipitation of the pigment was noted at higher temperature. By acetone extraction[4], the red pigment was shown to be maintained stably as a protein-precipitate.

Conclusions

Examination of the characteristics of the red pigment in Parma ham indicated it to apparently be a new derivative, differing from other water soluble Mb derivatives. The pigment was stable against change in pH, particulary so in the dark. The pigment precipitated with foreign proteins denatured by heating, but could be detected in its acetone extract.

Pertinent literature

1) Morita, H., Niu, J., Sakata, R., Okayama, T. and Nagata, Y., Proc. 41st ICoMST, 382-383, 1995.
2) Morita, H., Niu, J., Sakata, R., Okayama, T. and Nagata, Y., J. Food Sci., 61, 1021-1023, 1996.
3) Sakata, R., Honikel, K.O., Morita, H. and Nagata, Y., Fleischwirtsch., 76, 1046-1048, 1996.
4) Sakata, R. and Honikel, K.O., Fleischwirtsch., 76, 951-957, 1996.

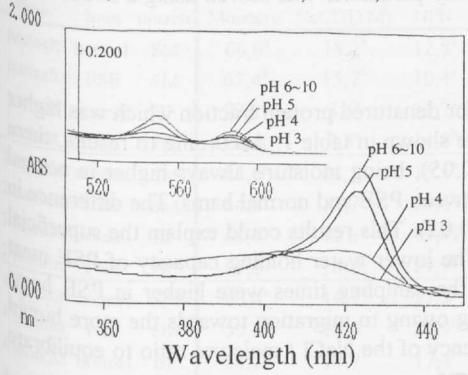

Fig.1. Effects of pH on the absorption spectrum of the water extract from Parma ham.

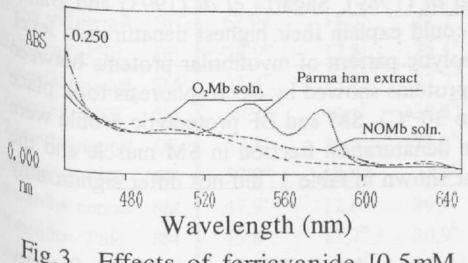

Fig.3. Effects of ferricyanide [0.5mM $K_3Fe(CN)_6$] on the absorption spectra of Mb derivatives.

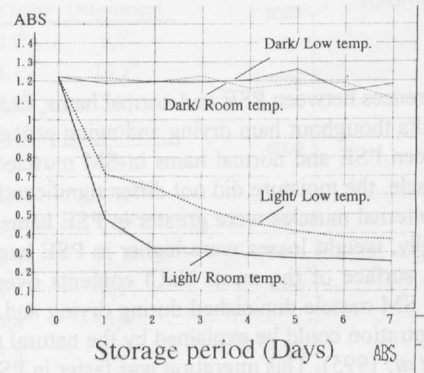

Fig.2. Effects of light exposure and temperature on red pigment stability in water extract from Parma ham.

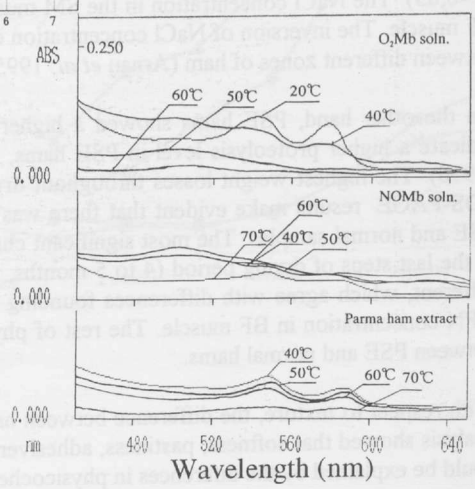

Fig.4. Effects of heating on the absorption spectra of the red Mb derivatives.