APPLICATION OF X-RAY PHASE-CONTRAST TOMOGRAPHY IN QUANTATIVE STUDIES OF HEAT INDUCED STRUCTURAL CHANGES IN MEAT

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Abstract – X-ray computed tomography is increasingly used in the studies of food structure. This paper describes the perspectives of use of phase contrast computed tomography in studies of heat induced structural changes in meat.
From the data it was possible to obtain reconstructed images of the sample structure for visualization and qualitative studies of the sample structure. Further data segmentation allowed structural changes to be quantified.

Key Words – Connective tissue, Myofibrils, Cooking, Quantitative analysis

I. INTRODUCTION

During the last decades X-ray computed tomography has generated interest as a valuable tool in non-destructive three-dimensional imaging of microstructure of various food products [1, 2, 3] Recently a novel grating based X-ray phase-contrast tomographic method with increased contrast has been demonstrated [4, 5].
Cooking of meat results in immediate and extensive structural changes of the meat proteins. These conformational changes are caused by the denaturation of the different meat proteins and include transversal and longitudinal shrinkage of the muscle fibres, aggregation and gel formation of the sarcoplasmic proteins and the shrinkage and solubilisation of the connective tissue [6].
The objective of this study was to study the heat induced changes in the structure of beef *Semimembranosus* by the use of X-ray phase contrast tomography. Data segmentation allowed quantitative parameters as percent shrinkage of myofibrils, connective tissue, and cooking loss to be extracted from the data.

II. MATERIALS AND METHODS

Sample preparation

Raw beef *Semimembranosus* were cut to fit into an 1.5 mL Eppendorf tube. When placed in the tube, the lid was closed under the surface of degassed PBS to avoid air bubble formation during the scan. After measurements of the raw sample the exact same sample were heat treated by placing the sample in a glass of water and heated in a microwave oven until the water reached the boiling point. The Eppendorf tube was left in the water bath for 15 minutes. The sample was then reheated to the boiling point and left for 15 minutes. After heat treatment the sample was cooled in a cold water bath for 15 minutes.

X-ray tomography

Phase-contrast CT scans of the meat emulsions were obtained by use of a grating interferometric set-up at the TOMCAT beam line, Swish Light Source (SLS) at the Paul Scherrer Institute (PSI). The set-up is described in detail in [7]. Measurements were made at 25 keV and the third Talbot fractional distance. The effective pixel size was 7.4 μm.

Data segmentation

With statistical analysis the data was segmented in two steps. First the data was modelled as a mixture of Gaussians with an expectation-maximization (EM) algorithm [8]. Secondly, the data was segmented using an efficient approximate graph cut algorithm [9]. These two

steps ensure that both the spectral and spatial context of the data is considered.

III. RESULTS AND DISCUSSION

The results are based on a preliminary data analysis on 3 out of 513 slices from the data set.

Images of transverse cut of the raw and cooked sample are presented in Fig. 1. The images are prepared from single slices of the reconstructed phase contrast data sets. The dark ring surrounding the sample is the Eppendorf tube used as sample container. In the raw meat (Fig 1, A) the muscle fibre part of the muscle is represented by the grey areas, whereas the connective tissue, mainly the perimysium, is seen as white areas. The small black spots are intramuscular fat. Towards the edge of the samples darker grey areas are seen representing a water phase. Due to changes in electron density of the muscle components as a consequence of heating a colour shift is seen in the image of the cooked sample (Fig1, B) compared to the raw sample. In the cooked sample muscle fibres are seen as the white areas, whereas the connective tissue is seen as light grey. The most remarkable changes in the structure is the shrinkage of both the total sample volume and the individual fibre bundles. In the cooked sample the fibre bundles appear clearly separated by the surrounding endomysium. The shrinkage of the myofibrils have forced water to be expelled which can be observed as an increase in the area of the water phase surrounding the meat. The area of the perimysium is increased indicating partly solubilisation.

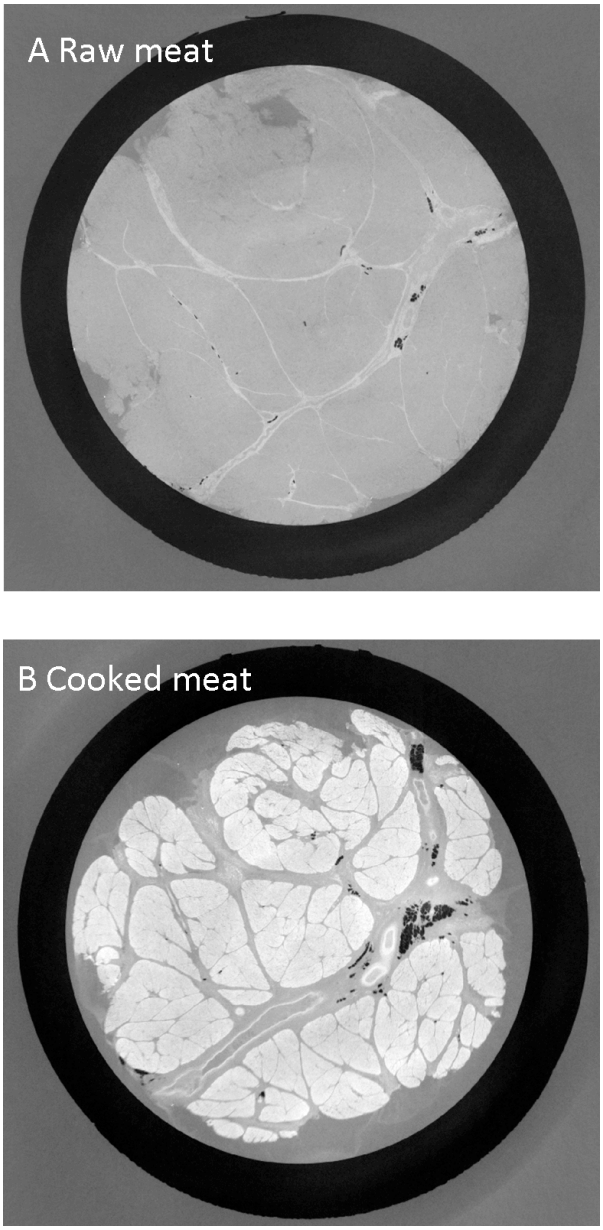

Fig 1. Transverse sample cut reconstructed from the phase contrast tomogram.
A: Raw beef. B: Cooked beef.

The observed difference in microstructure can be further explored by comparison of the histograms provided for each sample (Fig 2).

The histogram from the raw sample (Fig 2, A) shows that water, muscle fibres and connective tissue can be clearly separated from the data set.

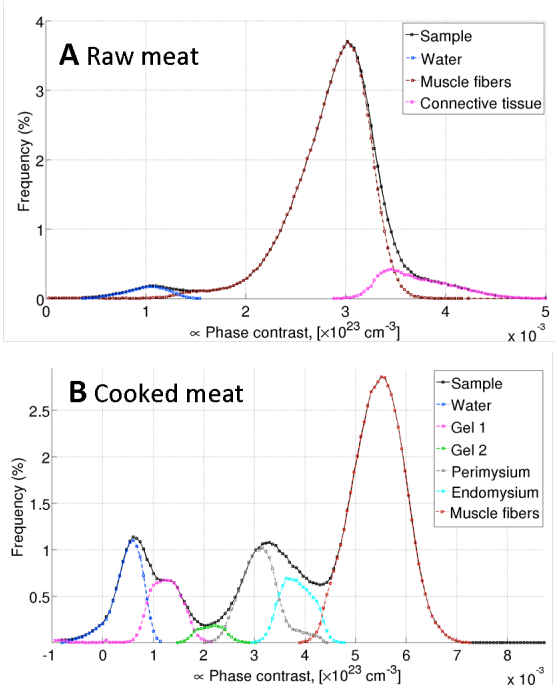

Fig 2. Histograms of the distribution of the muscle components before and after cooking.

The volume is mainly taken up by the myofibrils. The connective tissue can only be separated into one component.

From the histogram of the cooked sample (Fig 2, B) it is clear that cooking changes the composition of the components as the electron density i.e. the muscle fibres increases. Further differences in the behavior of the two types of connective tissue allows the perimysium and endomysium two be quantified separately. The endomysium shrinks resulting in increased density, while the density of the perimysium is lowered indicating partly solubilisation. Further two peaks assigned to gel-like populations appear in the histograms, which may be ascribed to the extraction and gelling of sarcoplasmic proteins [6].

The changes in the volumes of the main components are summarized in Table 1. In the Table the volumes of the observed gel-like structure are included in the water phase.

Table 1. Distribution of the different muscle components in the raw and the cooked samle. The distribution is calculated in percentage of the total sample volume.

Component	Raw (vol%)	Cooked (vol%)
Water	2.72	22.05
Muscle fibres	87.81	55.32
Connective tissue	9.47	22.62

The heat induced changes in muscle structure all compares to previously observations on the subject as studied by microscopy, spectrometric methods and differential scanning calorimetry [6]. Compared to these methods phase contrast tomography offers unique possibilities as both qualitative and quantitative studied can be made on the same sample without prior sample preparation as isolation of single muscle components or histology. In addition information is obtained from the full sample volume, which may serve to minimize variation between measurements.

The presented qualitative data is based on preliminary data segmentation. Completion of the data analysis is expected to make it possible to extract more detailed information on the volumetric distributions.

IV. CONCLUSION

Phase contrast tomography offers the possibility to study structural changes of meat caused by cooking. The non-destructive characteristics of the method made it possible to study the exact same sample before and after heat treatment. The high contrast in the data set made it possible to both visualise and quantify structural variation within the meat structure

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the experimental work by Torsten Lauridsen, Rasmus Laurberg Hansen and Karin E. Ibsen on obtaining the sample data set. H.E., M.S.N. and R.M. acknowledge financial support through the NEXIM research project founded by the Danish Council for Strategic Research (contract no. 11-

116226) within the Program Comission on Health, Food and Welfare.

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