Journal of Food Engineering 94 (2009) 283–289

Contents lists available at ScienceDirect

Journal of Food Engineering

journal homepage: www.elsevier.com/locate/jfoodeng

X-ray computed tomography to study processed meat microstructure

P. Frisullo [a,c], J. Laverse [a,c], R. Marino [b], M.A. Del Nobile [a,c,*]

[a] Department of Food Science, University of Foggia, Via Napoli 25, 71100 Foggia, Italy
[b] Department of PrIME, Università degli Studi di Foggia, Via Napoli 25, 71100 Foggia, Italy
[c] Istituto per la Ricerca e le Applicazioni Biotecnologiche per la Sicurezza e la Valorizzazione dei Prodotti Tipici e di Qualità, Università degli Studi di Foggia, BIOAGROMED – Via Napoli 52, 71100 Foggia, Italy

ARTICLE INFO

Article history:
Received 25 December 2008
Received in revised form 23 March 2009
Accepted 24 March 2009
Available online 1 April 2009

Keywords:
Microtomography
Quantitative analysis
Processed meat
Microstructure

ABSTRACT

In this work, the X-ray microtomography (μCT) technique was used for the analysis of fat microstructure in five different types of Italian salami. Five different types of Italian salami, chosen to exhibit variability in terms of visible structure of fat, were used for this experiment: Milano, Ungherese, Modena, Norcinetto and Napoli. Appropriate quantitative three-dimensional parameters describing the fat structure were calculated, for example, the structure thickness (ST), object structure volume ratio (OSVR) and the percentage object volume (POV). To measure the accuracy of the three-dimensional parameter, POV, the fat content was also determined by the extraction method [AOAC, 1995. Official Methods of Analysis, 16th ed. AOAC International, Washington, DC]. The results for this study show that μCT is a suitable technique for the microstructural analysis of fat as it does not only provide an accurate percentage volume of the fat present but can also determine its spatial distribution.

1. Introduction

Given the enormous success of X-ray microtomography in medical applications, material science, chemical engineering, geology and biology; it is not surprising that in recent years much attention has been focused on extending this imaging technique to food science, as a useful technique to aid in the study of food microstructure. This technique has successfully been used for the characterisation of the three-dimensional structure of a metallic foam during compressive deformation (McDonald et al., 2006), three-dimensional visualization of human stem cell muscle homing (Torrente et al., 2006) and characterisation of entrapped liquid formed during partial remelting of a cold-rolled Al–8 wt.% Cu alloy (Terzia et al., 2009). The microstructure of food products influences to a large extent the physical, textural and sensory properties of these products. Developing a proper understanding of the microstructure, particularly the spatial distribution and interaction of food components, is a key tool in developing products with desired mechanical and organoleptic properties. Information about the three-dimensional microstructure of food products and ingredients can be obtained using various imaging techniques. Commonly used techniques to-date are bright-field, polarising and fluorescence light microscopy (LM), confocal scanning laser microscopy (CSLM) and electron microscopy (EM). Other techniques such as atomic force microscopy (AFM) and magnetic resonance imaging (MRI) are used for specific food applications. Although, these wide varieties of imaging techniques exist, they are mostly invasive, as they require sample preparation hence, formation of artifacts or are restricted to certain types of food products.

X-ray microtomography (μCT), on the other hand, is a non-invasive technique that has several advantages over other methods, including the ability to image low moisture materials. It uses the differences in X-ray attenuation arising, principally, from differences in density within the specimen. A series of two-dimensional X-ray images are obtained as a sample is rotated. The series of slices, covering the entire sample, can be rendered into a three-dimensional image that can either be presented as a whole or as virtual slices of the sample at different depths and in different directions. Manipulation and analyses of μCT data using special software also allows reconstruction of cross-sections at depth increments as low as 15 μm, and along any desired orientation of the plane of cut. A series of non-invasive μCT slices of the same sample in any direction can provide much more information than just one scanning electron microscopy (SEM) or optical imaging picture for example. X-ray microtomography has been successfully used to observe the stability of gas bubbles in dough during the bread making process (Whitworth and Alava, 1999), the microstructure of foams (Lim and Barigou, 2004), three-dimensional quantitative analysis of breadcrumb (Falcone et al., 2005), the study of bread porous structure (Falcone et al., 2004) and ice crystals within frozen foods (Mousavi et al., 2005). Recently this technique has also been used to study the bubble size distribution in wheat flour dough (Bellido et al., 2006), the effect of far-infrared radiation assisted drying on microstructure of banana slices

* Corresponding author. Address: Department of Food Science, University of Foggia, Via Napoli 25, 71100 Foggia, Italy. Tel./fax: +39 881 589 242.
E-mail addresses: delnobil@unina.it, ma.delnobile@unifg.it (M.A. Del Nobile).

0260-8774/$ - see front matter © 2009 Elsevier Ltd. All rights reserved.
doi:10.1016/j.jfoodeng.2009.03.020

(Léonard et al., 2008), three-dimensional pore space quantification of apple tissue (Mendoza et al., 2007) and the role of sugar and fat in sugar-snap cookies (Pareyt et al., 2009).

All foods can be analysed in terms of their chemical composition. This could give limited information about the structure, physical state or sensorial properties. The natural building blocks of foods can be considered as water, air, carbohydrates, proteins and fats. The way in which these are structured during processing ultimately determines the functionality of the food. Fat content in meat products is a very important compound influencing the palatability characteristics such as taste, juiciness and texture. In addition, the visual appearance of the fat could affect the consumers overall acceptability of product and therefore the choice when selecting meat product before buying (Helgesen et al., 1998). For example, nowadays, lots of meat products with different fat contents and different physical and chemical features (protein network, moisture content, ingredients, additives and so on) are being manufactured.

The total fat content of processed-meat products such as salami is an important aspect used in numerous studies. Thus, reliable methods for the quantitative analysis of fat from this type of food products are of critical importance. There are several methods to analyse fat content quantitatively (Monin, 1998), although the method that is commonly used is based on chemical analysis (AOAC, 1995), it is quite expensive and time consuming. Furthermore this technique is destructive to the sample, as a result, the same sample cannot be measured more than once and sometimes uses harmful, flammable solvents with health and environmental hazards. On the other hand, X-ray microtomography is non-destructive and provides in-depth information on the microstructure of the food product being tested, therefore a better understanding of the physical structure of the product and from an engineering perspective, knowledge about the microstructure of foods can be used to identify the important processing parameters that affect the quality of a product. Structure–property relationships can strongly affect the physiochemical, functional, technological and even nutritional properties of foods. For example, with regards to solid food foams like bread, extruded cereals, biscuits and cakes, the consumer appreciation of these products is strongly linked to the texture. For texture, sensory properties of solid food foams are related to both mechanical properties and cellular structure. In this context, determining the relationships between a given mechanical property and the cellular structure is thus of prime importance. Processes are no longer designed from a macroscopic level; knowing the properties of foods on the micro scale determines the process specification. Hence, X-ray microtomography is fast becoming a very useful tool to aid in the study of food microstructure.

This work has served to demonstrate the capability of X-ray microtomography as a useful technique to study fat distribution and the percentage of fat in food products such as salami.

2. Materials and methods

2.1. Raw materials

Five different types of Italian salami, chosen to exhibit variability in terms of visible structure of fat, were used for this experiment: Milano, Ungherese, Modena, Norcinetto and Napoli. They were purchased locally and were tested on the same day of purchase. Three samples were prepared for each type of salami, each 28 mm in diameter and a thickness of 18 mm. Each sample used for X-ray microtomography (μCT) analysis was wrapped with parafilm to avoid dispersion of moisture; the parafilm does not interfere with the X-rays. The same samples used for μCT analysis were also used for chemical analysis of fat composition.

2.2. Tomographic analysis

For μCT the samples were imaged under the same conditions, using the Skyscan 1172 high-resolution desktop X-ray microtomography system (Skyscan, Belgium). A salami sample was placed on a rational plate; the source and the detector were fixed, while the sample was rotated during measurement. Power settings of 82 kVp and 125 μA were used. A CCD camera with 2000×1048 pixels was used to record the transmission of the conical X-ray beam through all samples. The distance source–object–camera was adjusted to produce images with a pixel size of 15 μm. Two-frame averaging, a rotation step of 0.70° and an exposure time of 1475 ms were chosen to minimize the noise, covering a view of 180°. Smoothing and beam-hardening correction steps were applied to suppress noise and beam hardening artifacts, respectively. Beam hardening correction was only moderately applied (set to 25% within NRecon) due to the use of an aluminum filter during acquisition. This filter acts to suppress low energy X-rays from the source, thus minimizing-beam-hardening artifacts. A fast ring artifacts reduction (set to 7 within NRecon) was also applied. Once initial parameters were set, the acquisition step was completely automated and did not require operator assistance. Scan time, on average, required 20 min. A set of flat cross-section images, was obtained for each sample after tomographical reconstruction by the reconstruction software NRecon (Skyscan). Three-dimensional reconstructions of samples were created by effectively stacking all two-dimensional tomographs, a total of 125 slice images with a slice spacing of 0.069 mm.

For image processing and analysis, the Skyscan software, CT-analyser (CTAn) was used. Image segmentation was firstly carried out on the smoothed 8-bit grey-scale images obtained from the reconstruction step, using CTAn (Skyscan) software. Segmentation is the process of converting the grey-scale images into black and white images by assigning the value 1 to all pixels whose intensity was below a given grey tone value and 0 to all the others. For this, an automatic threshold based on the entropy of the histogram (Sahoo et al., 1988) was calculated for each image. The lower grey threshold (8) and upper grey threshold (110) values were identified; each sample was processed under the same conditions.

For data analysis, prior to three-dimensional reconstruction, a component-labelling algorithm, available within CTAn, was used to isolate the largest three-dimensional connected structures. All reconstructions where created in CTAn (Skyscan) using an adaptive rendering (locality 10 and tolerance 0.25) algorithm and saved as P3G surface model (SkyScan model format). P3G models were then imported into CT vol software (Skyscan) for visualization.

The following six geometric parameters were measured using the CTAn software (Skyscan): percent object volume, object surface/volume ratio, fragmentation index, structure thickness, structure separation and degree of anisotropy. Where, (i) percentage object volume is the proportion of the VOI (volume of interest) occupied by binarised solid objects, i.e., fat; (ii) object surface/volume ratio is the basic parameter in characterising the complexity of the structures and is also the basis of model-dependent estimates of thickness, i.e., size and distribution of fat present in each sample; (iii) fragmentation index is an index of connectivity of structure, which was developed and defined by Hahn et al. (1992), it calculates the index of relative convexity or concavity of the total surface of the sample; (iv) structure thickness calculates or estimates the true structure thickness of the sample from two-dimensional measurements; (v) structure separation is essentially the thickness of the spaces as defined by binarisation within the volume of interest, i.e., fat. It can also be calculated either from