The Annals of the University *Dunarea de Jos* of Galati
Fascicle VI – Food Technology, New Series Year III (XXXII)
Paper presented at the International Symposium *Euro - aliment 2009*, 9th – 10th of October 2009, Galati – ROMANIA

INFLUENCE OF THE EXTRACTION METHOD ON THE RHEOLOGICAL PROPERTIES OF MYOFIBRILLAR PROTEINS FROM DIFFERENT SOURCES

PATRASCU LIVIA, CERCEL FLORIN, ALEXE PETRU

Department of Biochemistry,
"Dunarea de Jos" University, 111 Domneasca Street, 800201 Galati, Romania
livia.mantoc@ugal.ro

Received 10 July 2009
Revised 13 October 2009

Extraction of myofibrillar protein was performed using three different water solutions in order to obtain different values of pH), from chicken breast and organs (liver and heart). This study investigates the impact of the extraction methods on viscoelastic properties of myofibrillar proteins. Three types of viscoelastic measurement were made. The first was an oscillatory analysis with strain sweep step and a frequency sweep step. Secondly, creep testing was performed with an initial load of 30 Pa. Finally, the products were subjected to heat – induced gelation. The gelation characteristics of myofibrillar proteins are indicative of meat product texture. Defining the performance of myofibrillar proteins during gelation is beneficial in maintaining quality and developing processed meat products and other processes. Rheological properties were measured while heating at 2.0°C/min from 5 to 80 °C. Results have shown better rheological characteristics of myofibrillar protein extracted in a solution of a low pH.

Keywords: myofibrillar proteins, viscoelastic properties, creep.

1. Introduction

Extraction of myofibrillar protein presents interest from the technological point of view for water-holding capacity enhancing and for their binder properties. Hydration surface properties, binding and rheological behaviour are terms used to describe protein functionality in meat-processing. Offer and Trinick (1983) suggest that the myofibrils are responsible for water retention in meat. Protein gels demonstrate viscoelastic behavior in that they exhibit both fluid-like (viscous) and predominantly solid-like (elastic) behaviour (Xiong, 1994). Thermally induced protein–protein aggregation is considered to be the key element to transform the viscous protein extract (sol) to a three-dimensional matrix during meat-processing (Samejima et al, 1969; Samejima, Ishioroshi, & Yasui, 1981, 1982; Xiong, 1992). By extracting the functional myofibrillar protein from the muscles, the protein may be used as an ingredient for the formulation of some whole-muscle processed meat products(Li, 2006).

Lesiow and Xiong (2003) evaluated the effect of pH and muscle fiber type on the gelation properties of chicken muscle homogenates. Using both oscillatory and compression testing, the authors determined that the strongest gel from the chicken breast muscle homogenates had a pH of 6.30. These findings illustrate that muscle types respond differently to pH.

The objectives of this study were to determine the impact of the extraction method (especially the extraction solution pH) on the rheological properties of myofibrillar protein and the extent to which muscle type influences the viscoelasticity of myofibrillar proteins. Three types of meat, chicken breast, beef liver and chicken heart were investigated to determine effects of myofibrillar protein source and of extraction solution pH (1.8; 7.0 and 10.5) on protein viscoelastic characteristics.

2. Methods and materials

2.1. Myofibrillar protein isolation and preparation

Chicken breast, liver and heart (300g each), were used after 1 day postmortem storage at 4 ºC. Highly-purified myofibrillar proteins were prepared using three different conditions. The methodology used included, in all cases, solubilizing/washing the minced meat four times followed by protein

precipitation; for the first sample distilled water was used with pH adjusted to 10.5 using Na_2CO_3; for the second sample distilled water was used with pH adjusted to 1.8 using HCl; for the third sample distilled water was used with pH adjusted to 7.0 using NaOH. Samples were centrifuged after each solubilization/washing for 10 min at 3000 rot/min. In order to characterize the wet myofibrillar proteins from the rheological point of view all protein concentrates were first brought to a dry matter of 14.5±0.5%. Thus we obtained nine myofibrillar protein samples: Breast 7.0; 10.5; 1.8; Liver 7.0; 10.5; 1.8; and Heart 7.0; 10.5; 1.8. Protein samples were stored at 4 ºC prior to analysis.

2.2. Rheological measurements

The viscoelastic characterisation of samples were investigated by a controlled stress rheometer (Advance Rheometer AR2000, TA Instruments) equipped with an cone-and-plate probe 40 mm 2º measuring system and 1500µm gap. Strain sweep step were first performed with a variation of % strain from 0.025 to 28.636 % a frequency rate of 1 Hz for all samples. The strain stress method was found to be a good approach for yield stress analysis of medium viscosity materials.

Samples were subjected to oscillatory measurement at a frequency range of 0.01 – 10.00 Hz (0.6283 – 62.83 rad/s for ang. frequency) by a controlled osc. stress of 20 Pa chosen from the strain sweep test. Subsequently, their storage modulus (G`) and loss modulus (G``) including the delta (δº) were determined.

Creep testing

Creep compliance, under a constant stress, was measured at fixed intervals with an initial load of 30 Pa at a holding time of 10 min. Creep tests examine the way the products respond to steady low stresses. Since compliance (J,[1/Pa]) is the reciprocal of an initial elastic modulus (G_0) (Steffe, 1996) a lower compliance means higher G_0 and exhibits a greater rigidity of the gels. (Fig. 3)

Temperature ramp step

Measurements were conducted within the linear range at a strain of 0.0025 and a constant frecquency of 1 Hz. Samples were heated from 5 to 80 ºC at a rate of 2 ºC/min through the use of a Peltier temperature controller. Edges of samples were covered with light silicone oil to prevent drying out. Evolution of storage modulus G`with temperature was monitored.

3. Results and discussion

3.1. Dynamic viscoelastic oscillatory measurement

Viscoelastic measurement of storage (G`), loss (G``) modulus and delta are among parameters that characterise myofibrillar protein indicating gel strength. G` and G`` moduli are a measurement of the energy stored and dissipated in the material under test. For all samples, both G` and G`` developed in a similar way. However, the G` values were considerably greater in magnitude than the G`` values, showing that predominantly elastic gels were formed. Thus, only the G` values will be discussed (Vardhanabhuti et al. 2001). The bigger G` is the stronger structure material with a solid - like (elastic) behaviour. Also, if a material is an ideal solid, its phase shift angle (δ) value will approach 0º, whereas if it is an ideal liquid, δ will approach 90º. A purely elastic material will have a δ of 45º.

All myofibrillar protein had a phase of shift angle (δº) closer to a solid behaviour regardless the pH value. Fig. 2 showed the variation of G` values of myofibrillar proteins as a function of frequency. Small δº values (Fig.1) with large G` values indicate that, although the elastic component predominates in the system, it still behaves as a weak viscoelastic gel (Apichartsrangkoon et al, 1998; Messens et al. 2000). Measurements showed a bigger G` for chicken breast, this meaning a lot of pressure requested for material to initiate flow. This indicates a better internal structure with a solid-like behaviour. For samples extracted with a solution of a neutral pH, storage modulus values were very small, tending to zero. This indicates a very weak structure.

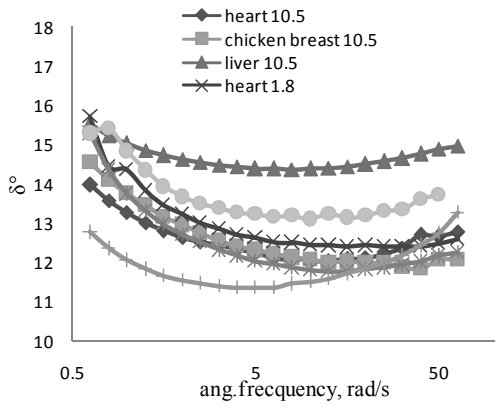

Figure 1. δ variation as a function of frequency for myofibrillar protein extracted with distilled water at different values of pH

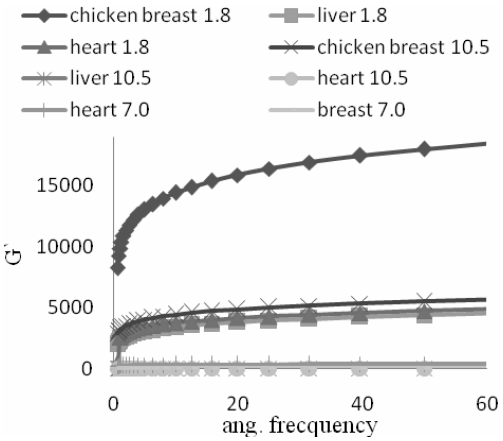

Figure 2. Typical storage modulus as a function of frequency for myofibrillar protein extracted with distilled water at different values of pH

3.2. Creep test

A creep test is a measure of how a material behaves on the application of a sudden stress which is maintained at a constant value for a specified time (Chattong, Apichartsrangkoon, 2008). Fig. 3 shows a smaller compliance for chicken breast sample extracted at a low pH meaning that poultry myofibrillar proteins have a stronger structure and a better binder capacity in comparison with liver or heart myofibrillar proteins as shown in the temperature ramp step also. The influence of the extracting pH is clearly seen in the liver case where the 10.5 sample has a much bigger compliance than the 1.8 sample. Samples extracted for neutral pH had a decuman compliance exceeding the others, on this account they didn't appear in the figure.

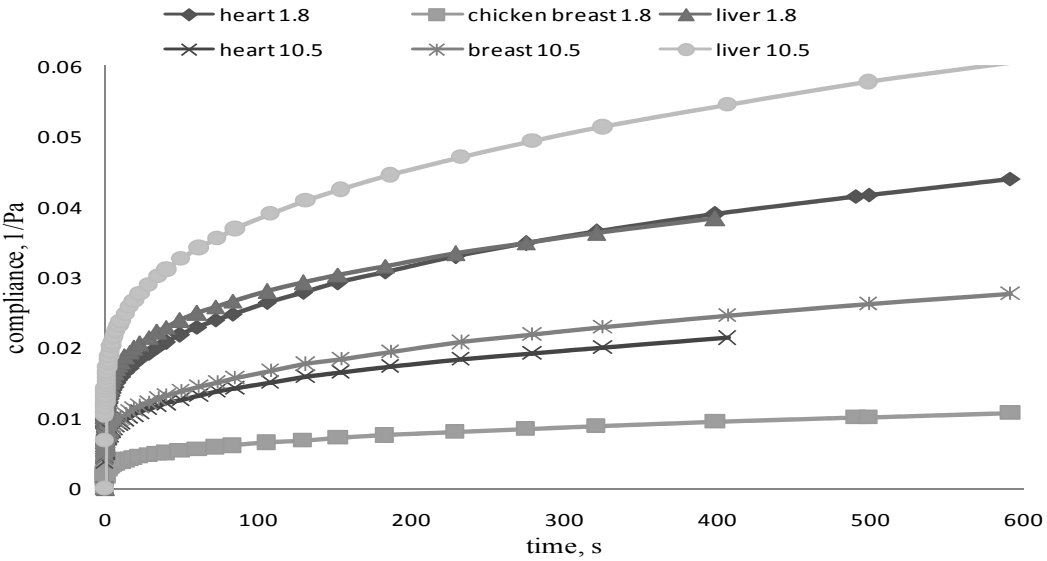

Figure 3. Complience-time relation for myofibrillar protein extracted from different sources at a pH = 1.8 and 10.5

3.3. Temperature induced gelation

All samples began to form a gel shortly after 50 ºC, as indicated by an increasing G` modulus after a long falling until gelation started indicating transition from a liquid-like state to a solid-like one. This temperature is usually taken as gelation temperature and corresponds to the temperature for which G` increases and becomes greater than the background noise, which is one of the common methods of

detecting the gelling point in the absence of a crossover between G` and G`` (Matsumura, Mori, 1996; Ould Eleya, Gunasekaran, 2002; Lamsal et al 2007). The mechanism in which myosin forms a gel is suspected to involve the aggregation of the heavy meromyosin (HMM), which is the head of the protein, and the formation of a network amongst the LMM, or tail portions of the protein (Samejima, Ishioroshi, Yasui, 1981). This continued until the temperature reached approximately 70 ºC when the gel strength increased again.

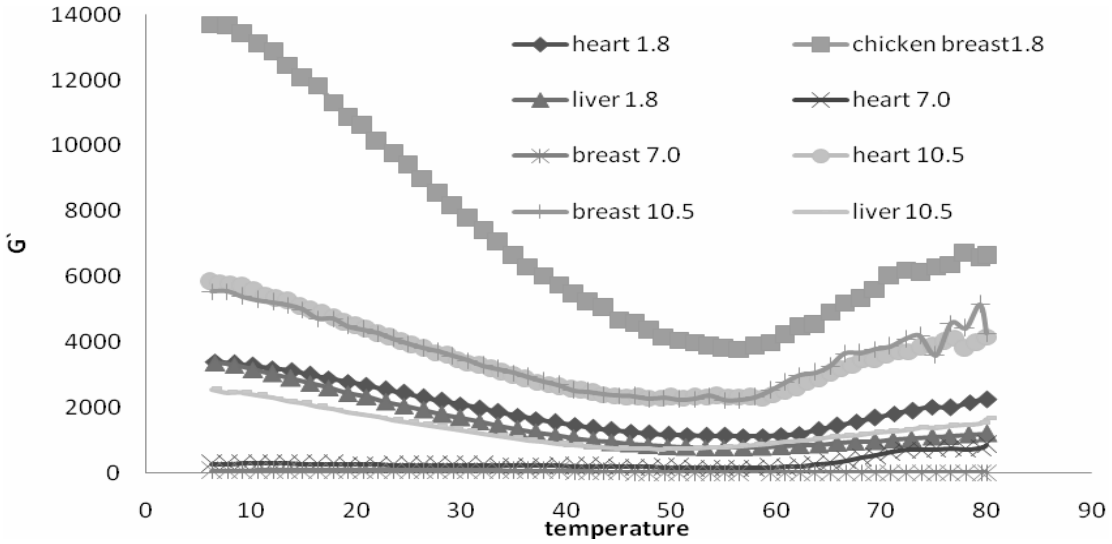

Figure 4. Average storage modulus (G`, Pa) measurements of myofibrillar proteins during heating, of heart, liver and chicken breast extracted at different values of pH

Near 80°C value, no significant changes in G` were observed, indicating that thermally-induced denaturation and aggregation of the protein concentrate were complete (Smyth et al, 1998 Doerscher et al. 2003,). As in the previous tests (strain sweep step and creep test) it can be seen that chicken breast myofibrillar proteins (solution pH=1.8) had the bigger G` values. 7.0 samples had a very low G` value, moreover, chicken breast assessed a bad structure.

4. Conclusions

The results of the current work document that myofibrillar proteins from different sources of meat respond differently from the rheological point of view. Also, it can be affirmed that myofibrillar proteins, extracted at pH=1.8, had a better structure, those from chicken breast being stronger, with a solid-like behaviour. pH also influences the response of myofibrillar protein when speaking of rheology. Results have shown that the extracting method for a neutral pH is not efficacious when we want to use myofibrillar proteins for their great rheological characteristics. It is incontestable that liver protein has a great quality from the nutritional point of view, but rheologicaly it has not a great value, as shown by the phase shift angle (δº) values and storage modulus (G`) in the temperature ramp test.

References

Apichartsrangkoon, A., Ledward, D. A., Bell, A.E., Brennan, J. G., 1998 – Physicochemical properties of high-pressure-treated wheat gluten. *Food chemistry,* **63**(2), 215-220.

Chattong, U., Apichartsrangkoon, A., 2009, Dynamic viscoelastic characterisation of ostrich-meat your following pressure, temperature and holding time regimes, *Meat science, **81**, 426 – 432.*

Doerscher, D.R., J.L. Briggsc, S.M. Lonergan ,2003 - Effects of pork collagen on thermal and viscoelastic properties of purified porcine myofibrillar protein gels, *Meat science,**66**, 181-188.*

Lamsal, B.P., Jung, S., Johnson, L.A., 2007, Rheological properties of soy protein hydrolisates obtained from limited enzymatic hydrolysis. *LWT*, (**40**) 1215-1223.

Lesiow, T., Xiong, Y. L., 2003, Chicken muscle homogenate gelation properties: effect of pH and muscle fiber type. *Meat science, **66**, 69 -74.*

Li, C.T., 2006 - Myofibrillar protein extracts from spent hen meat to improve whole muscle processed meats, *Meat science,* **72***,581-583.*

Matsumura, Y., Mori, T., 1996, Gelation. In G. M. Hall. (Ed.), *Methods of testing protein functionality.* **76-106**, London, UK: Blackie.

Messens, W. J., Van de Walle, D., Arevalo, J., Dewettinck, K., Huyghebaert, A. 2000, Rheological properties of high-pressure-treated Gouda cheese. *International Dairy Journal,* **10** *(5-6), 359 – 367.*

Offer, G., Trinick, J. 1983, On the mechanism of water holding in meat: the swelling and shrinking of myofibrils. *Meat science,* **8***, 245 – 281.*

Ould Eleya, M. M., Gunasekaran, S. 2002,- Gelling properties of egg white produced using a conventional and a low-shear reverse osmosis process. *Journal of Food Science,* **67**(2), 725-729.

Samejima, K., Hashimoto,Y., Yasui, T., & Fukazawa, T., 1969 – Heat-gelling properties of myosin, actin, actomyosin and myosin-subunits in a saline model system. *Journal of Food Science*, **34**, 242–245.

Samejima, K., Ishioroshi, M., Yasui, T., 1981, Relative roles of the head and tail proportions of the molecule in heat – induced gelation of myosin. *Journal of Food Science,* **46***, 1412 – 1418.*

Samejima, K., Ishioroshi, M., Yasui, T., 1982, Heat induced gelling properties of actomyosin: effect of tropomyosin and troponin. *Agricultural and Biological Chemistry*, **46**(2), 535–540.

Smyth, A. B., McCord, A., & O'Neill, E. 1998, Heat-induced gelation properties of chicken breast muscle salt soluble proteins when mixed with b-lactoglobulin or an a-lactalbumin enriched protein fraction. *Meat Science*, **48**, 135–147.

Steffe, J.F., 1996 – Rheological methods in food process engineering (2nd ed.) East Lansing Michigan: Freeman Press.

Vardhanabhuti, B., Foegeding E. A., McGuffey, M. K., Daubert, C. R., Swaisgood, H. E., 2001 – Gelation properties of dispersions containing polymerized and native whey protein isolate. *Food hydrocolloids, (***15***) 165-175.*

Xiong, Y .L. 1994, Myofibrillar protein from different muscle fiber types: Implications of biochemical and functional properties in meat processing. *Critical reviews in food science and nutrition,* **34***, 293 - 320.*