

# Identifying the composition of meat preparations

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## Abstract

This paper describes the composition of four products from the group meat preparations and four products from the group partially heat-treated meat preparations. Their composition was determined microscopically with the use of histological staining. The level of damage to skeletal muscle was determined using Calleja staining. Hematoxylin-Eosin staining was used for the description of product microstructure. Evidence of bone tissue was acquired using Alizarin Red staining. Polysaccharides were determined using PAS Calleja staining. The materials of animal origin detected were skeletal muscle, MSM and connective tissue. The vegetable materials detected were starches, native spices, hydrocolloids and breadcrumbs. The composition of the products was also compared with the legislative requirements. The results show a lack of conformity with the valid legislation in the group meat preparations in view of the raw materials of animal origin used. No such lack of conformity was found in the partially heat-treated meat preparations.

*Hamburgers, histological analysis, MSM, nuggets, skeletal muscle damage*

## Introduction

Meat products are undoubtedly highly popular products among Czech consumers. Consumers are interested in traditional meat products such as frankfurters, salamis, sausages and tinned meats. Meat preparations and partially heat-treated meat preparations have, however, been increasing in popularity in recent times. Primarily it is the result of the easy and quick preparation of these products. The consumer can prepare a final dish in a short period of time using these products as they already contain seasonings, salt and salting mixes and require merely heat treatment for product finalisation. These products can be divided into the groups, to which the following conditions apply, in accordance with the Czech legislation (Table 1).

Table 1. Groups of meat preparations in the Czech legislation (Decree No. 326/2001)

Meat preparations	Meat has not been heat treated, its internal cellular structure and its properties of fresh meat have been preserved; other foodstuffs, seasonings or additives have been added to it. Intended for heating or other culinary treatment before consumption. Products made from minced meat with the addition of more than 1% kitchen salt by weight are also considered meat preparations.
Partially heat-treated meat preparations	Partially heat-treated meat or combinations of meats, additives, auxiliary substances and perhaps aromatics. Intended for heat treatment.

The possibilities for the use of raw materials in their production are wide-ranging. The ingredients used must, however, fulfil conditions of food safety, must be permitted for use in foodstuffs and must be stated on the product package labelling (Decree No. 113/2005).

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The basic raw material for the production of meat preparations and partially heat-treated meat preparations is pork and beef meat, and less often poultry meat. Pork and beef is also used for the production of partially heat-treated meat preparations, though poultry and fish are also commonly used.

The aim of this study was to investigate whether the conditions stipulated in the Czech legislation are observed in meat preparations and partially heat-treated meat preparations, and to determine the constituent parts commonly used in their production.

### Materials and Methods

Four samples of meat preparations made of a mixture of pork and beef meat (hamburger mixes A, B, C and D) and four samples of partially heat-treated meat preparations (3 products containing poultry meat – chicken nuggets E, F and G – and 1 sample containing fish meat – fish nuggets H) were used for the purpose of evaluation. The samples were purchased at random on the retail network in the Czech Republic.

Four separate samples from each consumer package were evaluated. Calleja staining was used to determine the level of damage to skeletal muscle. Hematoxylin-Eosin staining was used for the description of product microstructure. Bone tissue was determined using Alizarin Red staining. Polysaccharides were determined using PAS Calleja staining. Samples were processed using the paraffin sections technique for all histological staining. The samples were sliced on an RM2255 rotary microtome (Leica, Germany) to a section thickness of 4 µm with an overcutting of 50 µm between sections, and to 500 µm for detection of bone fragments. Analysis of histological preparations was performed on an Eclipse E220 microscope (Nikon, Japan) at magnifications from 100 to 600. Histological staining was performed in accordance with accredited method SOP-01 and internal histochemical methods.

### Results and Discussion

Various kinds of meat are used in the production of meat preparations and partially heat-treated meat preparations, and this may include the use of mechanically separated meat (MSM). MSM does not, however, meet the stipulated criteria for the use of the term meat (Kameník and Pospíech 2012). Alizarin Red histochemical staining was used to detect the presence of MSM. An analysis was also performed to determine the level of damage to skeletal muscle based on the change to the stainability of the muscle fibres of skeletal muscle and muscle protein itself. A semi-quantitative evaluation with Calleja staining was used for this purpose (Sifre et al. 2009). The results are summarised in Table 2.

The results of evaluation of muscle tissue indicate that 3 samples (B, C and D)

Table 2. The presence of bone tissue and the level of damage to skeletal muscle

Sample	Number of bone fragments [cm <sup>2</sup> ]	Level of damage to skeletal muscle
Meat preparation		
Hamburger mix A	0.32	+++
Hamburger mix B	9.55	++
Hamburger mix C	7.05	+
Hamburger mix D	38.46	+++
Partially heat-treated meat preparation		
Poultry nuggets E	1.36	++
Poultry nuggets F	0	+
Poultry nuggets G	0.07	+
Fish nuggets H	0.41	++
Pearson correlation coefficient	R = 0.51	(p < 0.05)

+ Minor damage, ++ severe damage, +++ extremely severe damage to muscle fibres

were judged to be positive for the use of MSM according to the histological analysis, while another 3 samples (A, E and H) can be classified to be dubious and 2 samples (F and G) were negative according to the methodology of Pospiech et al. (2013). The level of damage to skeletal muscle shows that there is just a weak correlation of  $R = 0.51$  between the degree of damage to skeletal muscle and the number of bone fragments which was not statistically significant. The reason for these differing results is probably the processing of meat preparations and partially heat-treated meat preparations which leads to damage to muscle fibres independently of the use of MSM. Partially heat-treated meat preparations, in particular, are heat treated (pre-fried) and subsequently frozen. Both these processes lead to certain morphological changes to muscle tissue (Šimoniová et al. 2013) which may be erroneously considered changes caused by separation. The presence of bone fragments is not, however, influenced by these changes.

The knowledge of other ingredients used in the production of the product is also important to the assessment of the quality of the final product. Their presence affects the resultant textural, physical and sensory properties. The most important additives that can be studied with the use of microscopic methods are plant additives such as seasonings, starches and other hydrocolloids. A summary of the results of these analyses is given in Table 3.

Table 3. Plant constituents present in the samples

Sample	Seasoning	Fibre	Plant protein	Textured plant protein	Starch	Other hydrocolloids	Other plant additives	Other plant structures detected
Meat preparation								
Hamburger mix A	+				+	+		
Hamburger mix B	+				+	+		
Hamburger mix C	+				+	+		
Hamburger mix D	+				+	+		
Partially heat-treated meat preparation								
Poultry nuggets E	+				+		breadcrumbs	palisade cells
Poultry nuggets F		+			+		breadcrumbs	
Poultry nuggets G	+		+		+		breadcrumbs	
Fish nuggets H	+				+		breadcrumbs	palisade cells

+ Positive histological result

A number of conclusions can be drawn from this study into the presence of plant constituents. The seasonings used in these groups of meat products are generally in their native state. Sample F was the only sample in which no plant tissues typical of the use of seasonings were found. In this case, seasonings can be assumed to have been used in the form of seasoning extracts that cannot be detected by histological techniques. Fibre and plant proteins were only used in these products in isolated cases (F and G). Starch was the additive used most in both groups (Plate I, Fig. 1). The principal reason for its wide use is its low purchase price, along with its effect on texture properties, its water binding capacity and its use as a fat substitute (Desmond et al. 1998). Other hydrocolloids are used for the same reason as starch. The most important reason for their use is that they serve as binding agents, texture stabilisers and fat substitutes (Osburn and Keeton 1994).

The use of hydrocolloids has also been recorded in fish preparations (Park 1996), though this was not confirmed by our study, with only starch and breadcrumbs being found in the products (E, F, G and H). Palisade cells found (E and H – Plate I, Fig. 2) generally testify to the use of pulses (Horn 1987), though their presence in a refined form (protein extracts) was not confirmed in these two samples. Breadcrumbs were used primarily as a coating material (Plate II, Fig. 3) for partially heat-treated meat preparations, though they were found in the mixture itself in products F and G (Plate II, Fig. 4, Plate III, 5). Histological examination cannot, however, distinguish conclusively whether breadcrumbs or wheat flour were involved. Flour being added to the mixture rather than breadcrumbs cannot be ruled out. The reason for the use of flour or breadcrumbs in the mixture is their effect on the functional properties of the mixture. Mention has, however, also been made of the action of wheat flour as an antioxidant (Ulu 2004).

The presence of basic raw materials of animal origin, which might affect the quality of the product or give an indication of the source and quality of the ingredients used, was also analysed by microscopic methods. The results of these analyses are summarised in Table 4.

Table 4. The presence of constituents of animal origin

Sample	Skeletal muscle	Smooth muscle	Connective tissue	Veins	Nervous tissue	Skin	Fat tissue	Animal protein isolates	Other organs
Meat preparation									
Hamburger mix A	+	+	+		+				
Hamburger mix B	+		+						cartilage
Hamburger mix C	+	+	+	+			+		cartilage
Hamburger mix D	+		+				+		feathers
Partially heat-treated meat preparation									
Poultry nuggets E	+	+	+	+	+	+	+		spleen, cartilage
Poultry nuggets F	+	+	+	+	+	+	+		heart, cartilage
Poultry nuggets G	+		+			+			
Fish nuggets H	+		+				+		

+ Positive histological result

A number of conclusions can also be drawn from the analysis of raw materials of animal origin. Skeletal muscle and connective tissue were found in all the samples. The degree of damage to skeletal muscle differed, however, as can be seen in Table 2. Veins (C, E and F), smooth muscle (A, C, E and F) and peripheral nerves (A, E and F) were also detected in the samples. All the given animal constituents can be considered normal parts of meat (Decree No. 326/2001) and their presence is to be expected in products in which skeletal muscle is used. A large amount of collagen connective tissue in a product may, however, be caused by its addition in the form of collagen isolates or for the reason that skin emulsions, tendons or beef knuckle has been used (Zarkadas et al. 1995). High collagen connective tissue content was found on a semi-quantitative basis in samples B, C and D. The large amount of collagen connective tissue may also have been caused by the use of MSM in this case (Pospiech et al. 2010). The absence of veins, nervous tissue and fat is not caused by their actual absence, but is the result of the intense comminution of raw materials which means that they can no longer be identified microscopically. Animal protein extracts (isolates, concentrates) were not confirmed in any of the samples.

The presence of skin was confirmed in samples E, F and G. This was poultry skin in all cases. Its presence was also declared by the manufacturer either directly as poultry skin or as poultry MSM. Other organs were also found in two cases (E and F) – spleen and heart tissue. Their finding is associated with the use of poultry MSM. This finding does not, however, represent any risk, and other organs may also be found if poultry MSM is used. Cartilage was also found in samples B, C, E and F. This was poultry cartilage in all cases. This indicates the use of poultry meat in hamburger mixes B and C (Plate III, Fig. 6). The finding of feathers (Plate IV, Fig. 7) in sample D also indicates the use of poultry as a raw material. In view of the presence of large amounts of bone tissue in the samples (B, C and D) (Plate IV, Fig. 8), the source can be assumed to have been poultry MSM.

### Conclusions

The results of this study indicate that MSM (samples B, C and D) was used in the group of meat preparations and that the use of MSM was suspected in one case (sample A). These findings point to the use of MSM in these products. Only four samples were tested and this does not represent the entire retail market. These findings do, however, show that the supervisory bodies should devote attention to this group of meat products. No conflict with the valid legislation was found in the group of partially heat-treated meat preparations. The range of ingredients used in these products is extremely wide. The principal animal material used is skeletal muscle, MSM and connective tissue. The principal plant materials used are starches, native seasonings, hydrocolloids and breadcrumbs.

### References

- Decree of the Ministry of Agriculture No. 326/2001 Coll. on products of animal origin. In: Sbírka zákonů (In Czech)
- Decree of the Ministry of Agriculture No. 113/2005 Coll. on labelling of food. In: Sbírka zákonů (In Czech)
- Desmond EM, Troy DJ, Buckley JD 1998: The effects of tapioca starch, oat fibre and whey protein on the physical and sensory properties of low-fat beef burgers. *LWT - Food Sci Technol* **31**: 653-657
- Horn D 1987: Zum Nachweis pflanzlicher Eiweisszubereitungen in fleischerzeugnissen mit histologischen untersuchungsverfahren. *Fleischwirtschaft* **67**: 616-618 (In German)
- Kamenik J, Pospiech M 2012: Strojně oddělené maso - legislativní požadavky a způsob detekce. *Maso* **23**: 6-10 (In Czech)
- Osburn WN, Keeton JT 1994: Konjac Flour Gel as fat substitute in low-at prerigor fresh pork sausage. *J Food Sci* **59**: 484-489
- Park JW 1996: Temperature-tolerant fish protein gels using konjac flour. *J Muscle Foods* **2**: 165-174
- Pospiech M, Tremlová B, Řezáčová Lukášková Z, Randulová Z 2010: Mikroskopie masných výrobků. *Maso* **21**: 11-12 (In Czech)
- Pospiech M, Tremlová B, Eliášová M, Talandová M 2013: Optimalizace výpočtu obsahu kostní tkáně v histologických řezech. *Maso* **24**: 25-28 (In Czech)
- Sifre L, André B, Coton JP 2009: Development of a system to quantify muscle fibre destructuration. *Meat Sci* **81**: 515-522
- Šimoniová A, Pospiech M, Škorpilová T, Rohlík BA, Tremlová B, Pipek P 2013: Metody detekce zmrazení/rozmrazení masa: histologie a enzymová aktivita. In: Sborník přednášek a posterů XLIII. Lenfeldovy a Höklový dny, Brno: Veterinární a farmaceutická univerzita Brno, **63**: 255-258 (In Czech)
- Ulu H 2004: Effect of wheat flour, whey protein concentrate and soya protein isolate on oxidative processes and textural properties of cooked meatballs. *Food Chem* **87**: 523-529
- Zarkadas CG et al. 1995: Assessment of the protein quality of beefstock bone isolates for use as an ingredient in meat and poultry products. *J Agric Food Chem* **43**: 77-83

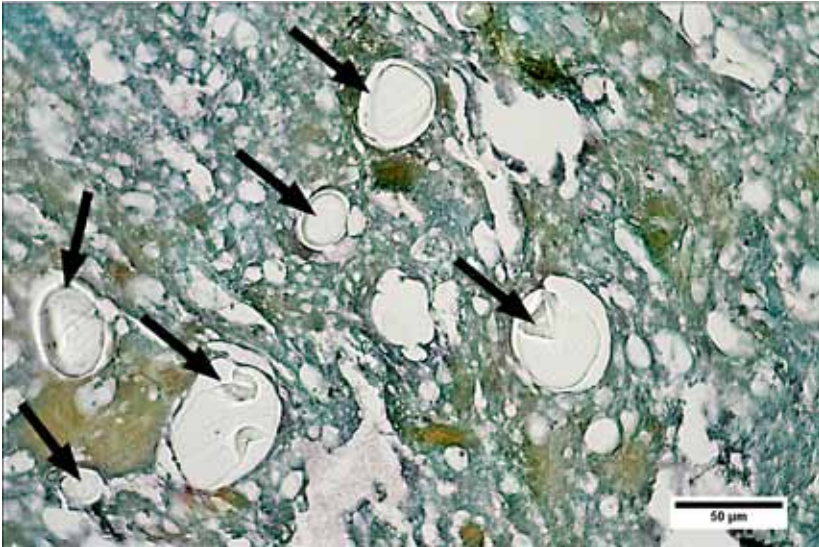


Fig. 1. Hamburger mix A, arrow starch, protein network in green, Calleja

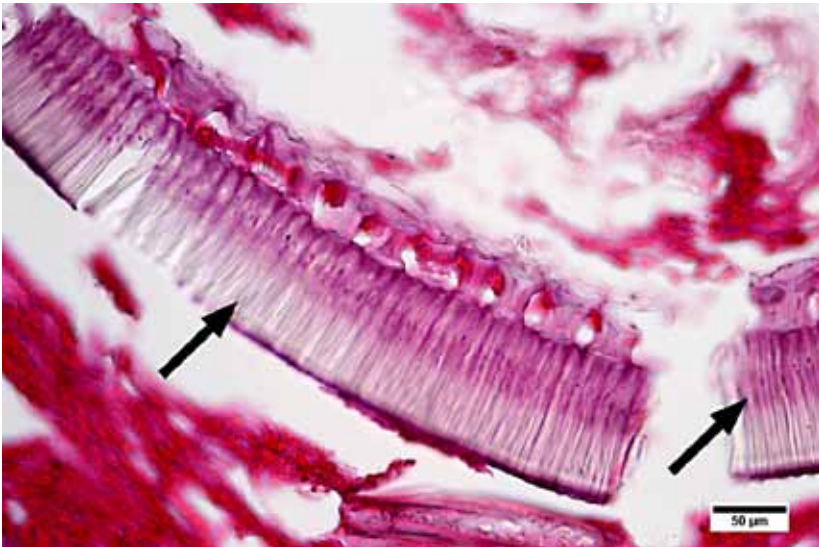


Fig. 2. Poultry nuggets E, arrow palisade cells, Hematoxylin-Eosin



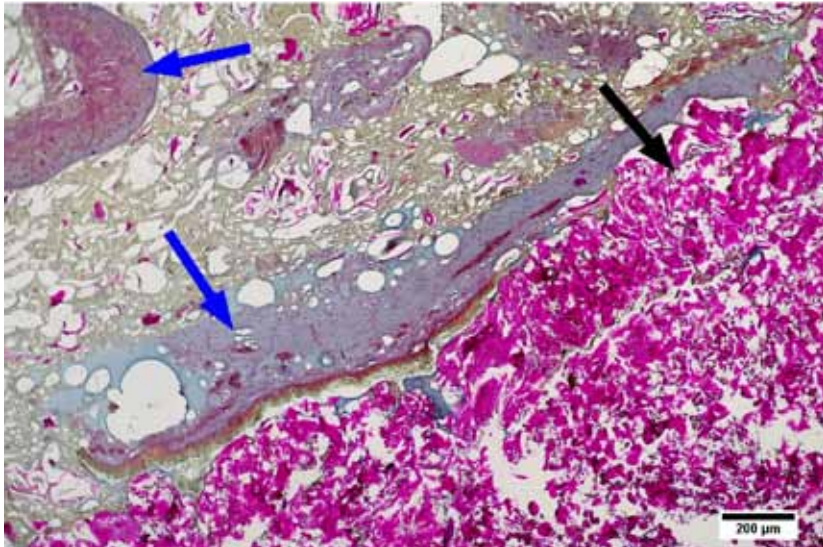


Fig. 3. Poultry nuggets E, arrow breadcrumbs, blue arrow poultry skin, protein network in green, PAS Calleja

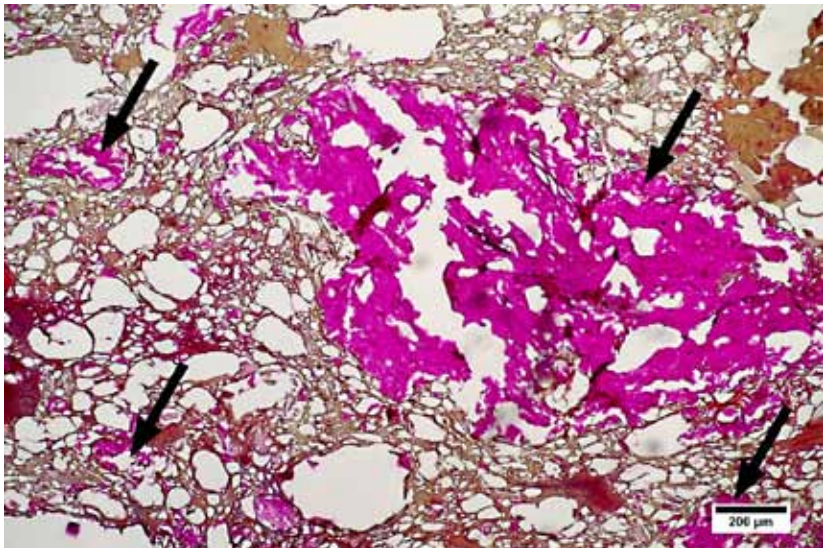


Fig. 4. Poultry nuggets E, arrow breadcrumbs/flour, protein network in green-brown, PAS Calleja

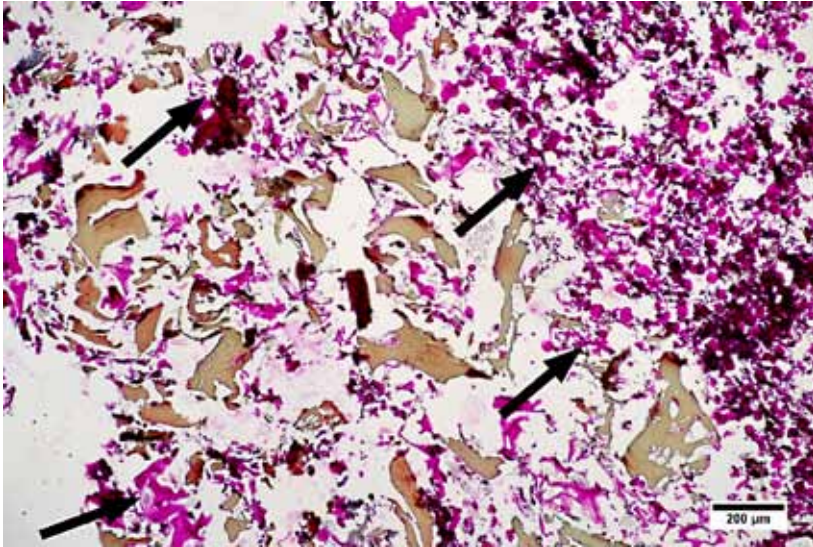


Fig. 5. Poultry nuggets E, arrow bread crumbs/flour, skeletal muscle in green, PAS Calceja

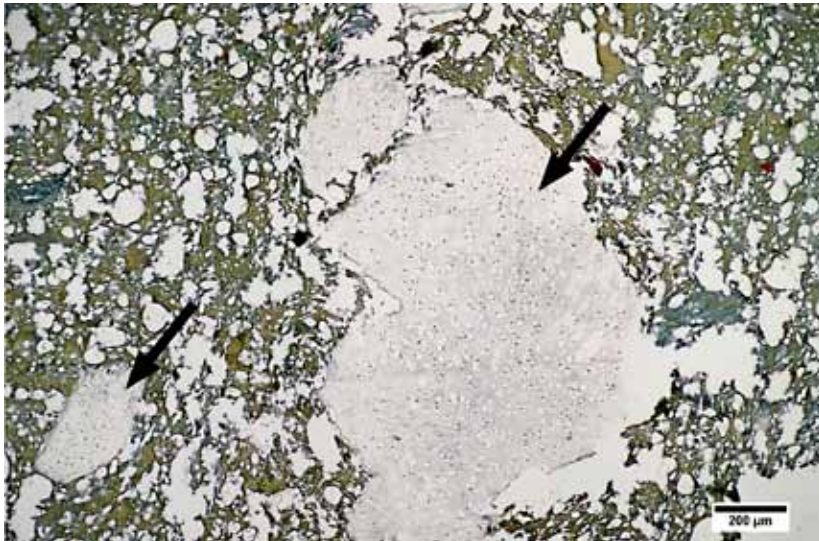


Fig. 6. Hamburger mix B, arrow cartilage, protein network in green, Alizarin Red



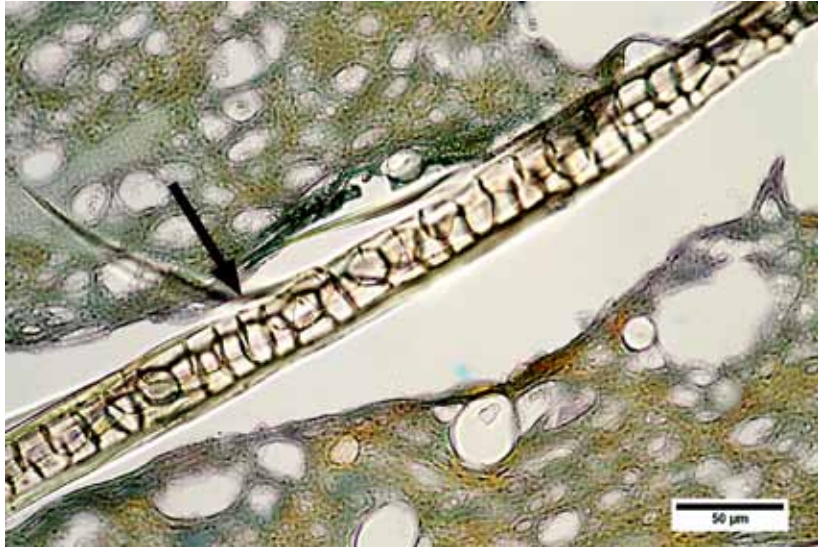


Fig. 7. Hamburger mix D, arrow feather, protein network in green, Alizarin Red

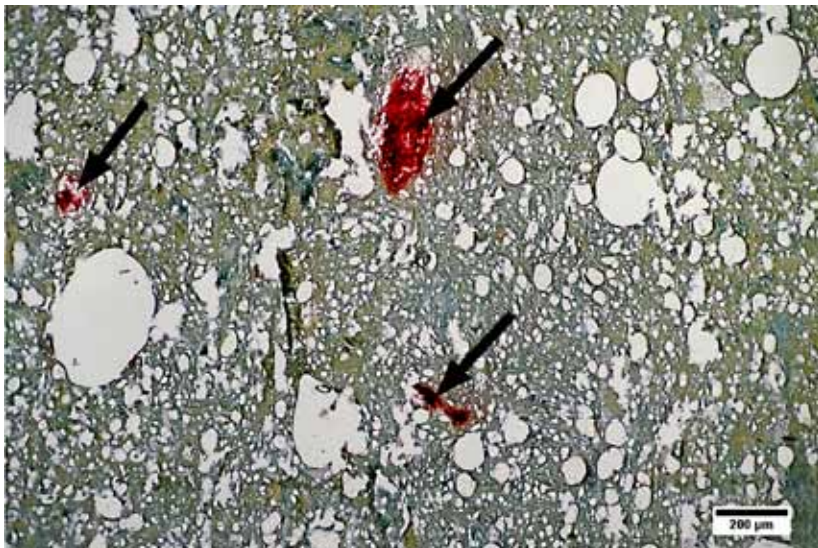


Fig. 8. Hamburger mix C, arrow bone fragments, protein network in green, Alizarin Red