FUNCTIONAL AND MORPHOLOGICAL CHANGES IN POLTURY FILLETS DURING ACID MARINATING WITH SODIUM LACTATE

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Abstract – The objective of this study was to evaluate the effect of 48 h acid marinating with 2% sodium lactate (SL) and 2% sodium chloride (NaCl) on poultry fillets. Muscles (24 h *post mortem*) were immersed in brine solution (2:1) and stored at 0 ± 4°C. The weight gain of the samples (M12 – marinated meat sample after 12 h soaking; M24 - marinated meat sample after 24 h soaking; M48 - marinated meat sample after 48 h soaking) increased significantly (p > 0.05) with 7.67 %, 10.79% and 19.98% resp. Meat pH decreased in M12 (6.39) and M24 (6.29), compared to the control sample (6.44) but in M48 (6.45) increased again. The brine pH was changed during 48 h marinating as follows: 5.88, 6.15, 6.19, 6.17. A little decline in water holding capacity (WHC) was found at the first 12 h of marinating (C - 15.82%, M12 - 14.97%), but after that WHC significantly increased to 19.73% (M24) and 20.80% (M48). Compared to control sample, in acid treated meat swelling in muscle fibers occur and connective tissue was found clearly disordered. The observed changes increased with the time of marinating. While sample preparation in light microscopy with Mayer`s haematoxylin is used for morphological observation of muscle tissue, Van-Gieson`s picrofuchsin stained samples clearly differentiated connective tissue from muscle tissue. It may be used effective to explore the morphological changes during marinating.

Key Words – Meat structure, Weight gain, Light microscopy, Haematoxylin, Van-Gieson`s picrofuchsin

I. INTRODUCTION

Marinating is commonly used method which involves injection, tumbling or soaking to disperse in the muscle tissue solutions contained water, salt and other ingredients. There are several types of marinating solutions according to added ingredients. The first are alkaline marinade solutions contained salt-phosphate mixtures, the second are acid solutions contained organic acids or their salts. The third type is water-oil emulsions contain salt, sugar, vinegar or citric acid and other supplements, but the information about this type of marinating in available literature is scarce.

The comparatively high pH during alkaline marinating increases water holding capacity due to protein extraction and shifting of the pH from the muscles proteins isoelectric point [1] and contributed to the dissociation of the actomyosin complex.

The acid marinade solutions most often include sodium lactate, potassium lactate, sodium citrate, and combinations of potassium lactate and diacetate. Offer and Knight [2] and [3] suggested that tenderness in acid marinated meat may be caused by changes in the connective tissue. According to Pearson [4] the irreversible changes occur at pH below 4.5, after marinating with weak organic acids/their salts, causing decrease in functional activity and WHC.

It is clear that the changes in the muscle and connective meat tissue depended of the marinating type and the marinade ingredients. There are many studies about alkaline marinating, but the impact of SL and NaCl marinating on muscle tissue characteristics in poultry has not been thoroughly investigated.

Therefore, the aim of this study is to identify the functional and morphological changes occurring in poultry fillets during acid marinating with sodium lactate, through appropriate methods which demonstrate changes in muscle and connective tissue.

II. MATERIALS AND METHODS

Poultry

The poultry fillets were purchased by the "Gradus" Ltd, Stara Zagora, district Stara Zagora (Bulgaria). The meat temperature was kept around 4 °C and at the 24 h *post mortem* (pH 6.4) poultry fillets was used in the experiment.

Marinade solution.

Sodium lactate was purchased by the "Teokom" Ltd, Sofia (Bulgaria).

Commercial packages sodium chloride was purchased from a local supermarket. Acid marinade solution (2% SL and 2% NaCl, aqueous solution) was prepared the day before conducting the experiments and stored at 4°C prior to use.

Seven samples of approximately 40 g each were cut from poultry breasts, placed in plastic box together with marinade solution (ratio solution: sample = 2: 1) and stored at 4 °C for 48 h. The samples were measured at 12, 24 and 48 h and labeled as follows: control sample C - not treated with acid marinade solution; M12 – acid marinated meat sample after 12 h soaking; M24 - acid marinated meat sample after 24 h soaking; M48 - acid marinated meat sample after 48 h soaking.

Water holding capacity was measured according Grau & Harnm [5] procedure, with some modifications (area has been calculated using the computer program Autocad).

Weight gain was determined using method described by Dragoev [6].

Light microscopic observation: Raw meats treated with or without salt-lactate solution were observed under light optical microscope (Olimpus BX41TF, Japan) at magnification 1000x. For light microscopic observation, the samples were cut into thin sections of approximately 5µm. The specimens were stained with Mayer`s hematoxylin and with Van Gieson's picrofuchsin for muscle and connective tissue observation resp.

III. RESULTS AND DISCUSSION

Functional changes

The determined result showed that pH in the poultry fillets decreased at first 24 h but in M48 sample increased again (Fig.1).

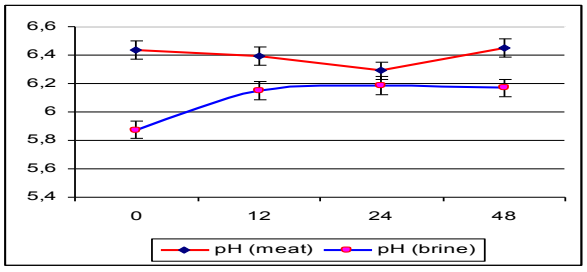

Figure 1. Poultry meat and brine pH changes in during 48 h marinating.

The opposite changes occurred in marinade solution.The possible reason for the observed changes is SL, NaCl and water penetration from solution to the meat and diffusion of muscle extract in to the brine. The weight gain significantly increased in acid marinated meat compared to untreated control and strong correlation between marinating time, marinade uptake and weight gain exist (Fig. 2). A little decrease in sample M 12 WHC was found (C - 15.82%, M12 - 14.97%), may be contributed by changes in pH, but after that WHC significantly increased to 19.73% (M24) and 20.80% (M48) (Fig. 3).

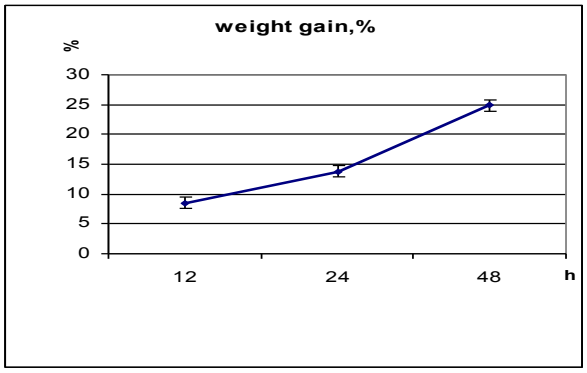

Figure 2. Weight gain changes during 48 h marinating.

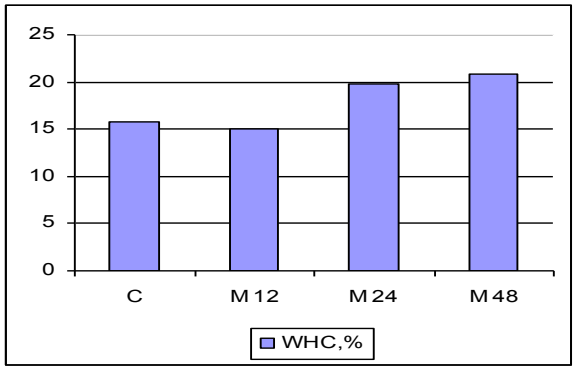

Figure 3. WHC changes during 48 h marinating.

Morphological analysis

During acid marinating with 2% SL and 2% NaCl, significant changes in morphological structure of poultry fillets occur (Fig. 4 - 11) demonstrated in light microscopy at 1000 x magnification. Miofibrilar structure in control sample (Fig. 4, 9) was found intact and with proper arrangement. After 12 h, due to presence of NaCl and SL from marinade solution the myofibrillar grid opened and the amount of water held in muscle increases. As a result, the muscle fibers swelled and their diameter increased.

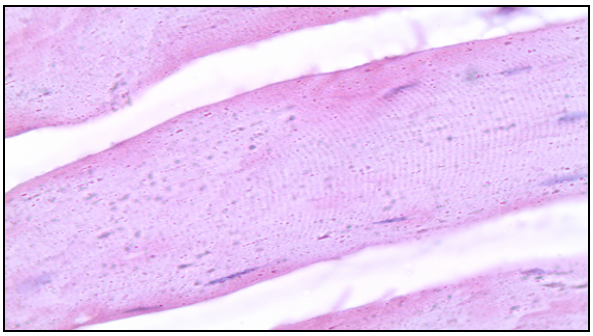

Figure 4. Control sample, longitudinal section (*LS*) of poultry fillets (24 h *post mortem*), hematoxylin stained, 1000x

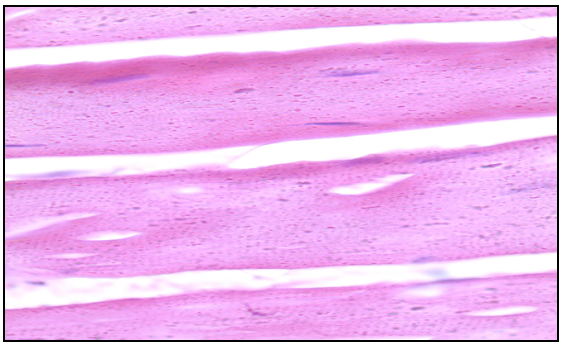

Figure 5. Sample M12 *(LS),* longitudinal section, hematoxylin stained, 1000x

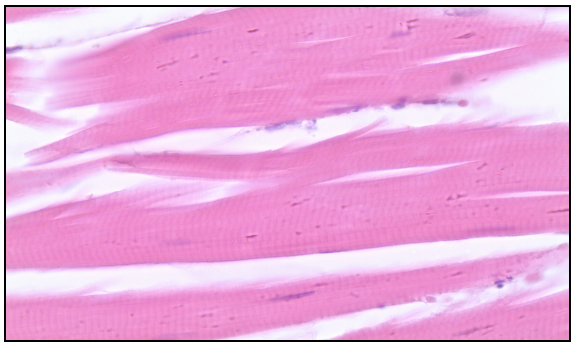

Figure 6. Control sample (untreated meat) transverse section of poultry fillets (7 d post mortem), stained with hematoxylin, 1000x4

Muscle tissue changes in M12 sample (Fig. 5) are similar to changes in control sample after 7 days storage at 0-4°C (Fig. 6). Fiber swelling, gaps formations and marinade penetration may be increased tenderness and juiciness of acid marinated poultry fillets.

After 24 h acid marinating with 2% SL and 2% NaCl partial fragmentation and gaps formation

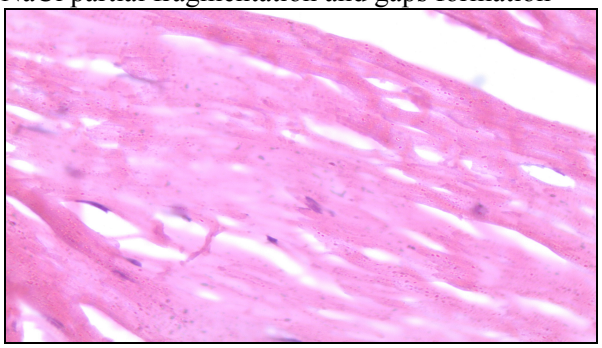

Figure 7. Sample M24, (*LS*), hematoxylin stained, 1000x

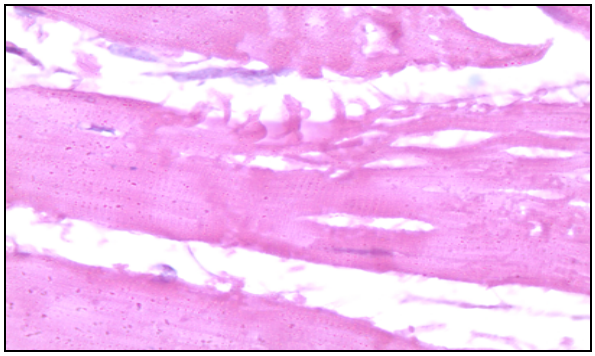

Figure 8. Sample M48, (*LS*), hematoxylin stained, 1000x

between myofibrils occur (Fig. 4, 7), which was a sign of major structural changes in the myofibrils. Marinating for 48 h. disrupted the myofibrillar structure to the largest extent (Fig. 8, 11).

Morphological changes were confirmed by the results obtained for weight gain (Fig. 2) and WHC (Fig.3).

After morphological observation from Van-Gieson strained samples, some deformations in connective tissue occur. While connective tissue in C sample (Fig. 9) was with regular arrangement, in M24 (fig. 10) and M48 (Fig. 11) samples looks disordered.

Figure 9. Control sample (untreated meat) transverse section of poultry fillets (24 h *post mortem),* Van - Gieson`s stained, 1000x

In similar research (*m. Biceps femoris*) with immunohistochemical methods, Sultana *et al*. [3] found that the connective tissue in the control samples was intact and in the form of thin collagen fibers. According to Balcerzac *et al*. [4] observed deformations are due to the myofibril swelling rather than protease activity. Acid marinating with 2% SL and 2% NaCl for 48 h disrupted the connective tissue to the largest extent.

The picrofuchsin used for Van-Gieson sample preparation, stained muscle tissue in yellow and connective tissue bright red colour clearly differentiated muscle and connective tissue.

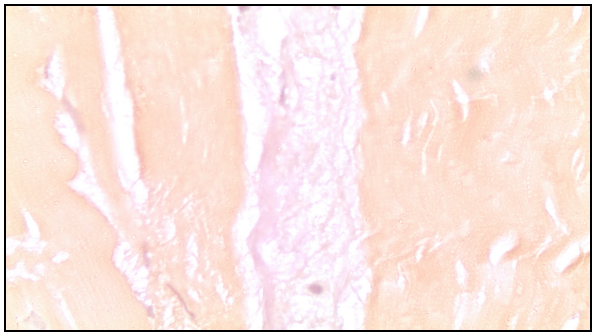

Figure 10. Sample M24, longitudinal section, Van - Gieson`s stained 1000x

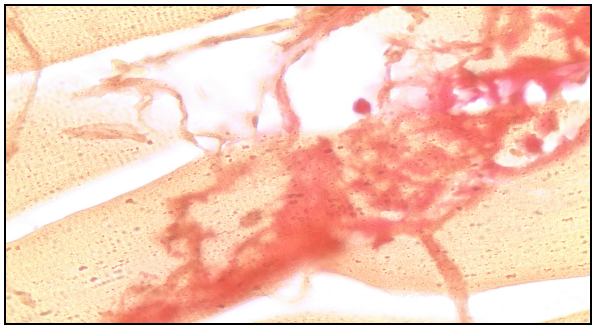

Figure 11. Sample M48, longitudinal section, Van - Gieson`s stained 1000x

IV. CONCLUSION

This study concludes that during 48 h marinating with 2% SL and 2% NaCl strong correlation between marinating time, marinade uptake and weight gain exist.

After 12 h soaking, the myofibrillar grid is opened and the amount of water held in muscle increases due to presence of NaCl and SL form marinade solution. The muscle fibers swelled and their diameter increased. Changes in connective and muscle tissue increased as follows: M12 <M24<M48. After 12 h 2% SL and 2% NaCl treatment muscle tissue are similar to changes in control sample after 7 d storage at 0-4°C.

Morphological observation of samples M24 and M48 showed that the muscle and connective tissue was disrupted to the largest extent, therefore optimal time for this kind of marinating is 12 h.

While sample preparation for light microscopy with Mayer`s haematoxylin is used only for muscle tissue observation, Van-Gieson`s picrofuchsin stained samples clearly differentiated connective from muscle tissue and may be used effective to explore the morphological changes during marinating.

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