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MICROBIOLOGICAL AND PHYSICOCHEMICAL EVALUATION OF PORK LEG TREATED WITH ORGANIC ACIDS AND/OR STEAM UNDER PRESSURE IN THE CONTROL OF SURFACE CONTAMINATION BY *Salmonella* Typhimurium

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ABSTRACT

Asymptomatic carrier pigs are the main risk factor for carcass contamination during the slaughter. Several post-slaughter prevention programs have been tested, such as organic acids and steam under pressure. These alternatives show low cost and high efficiency. This study tested the use of organic acid solutions and steam, in isolated tests and its associations. This experiment also evaluated the physical-chemistry features of the pork. Forty pork legs were contaminated with *Salmonella* Typhimurium DT 177 and subsequently divided into 4 treatments: immersion in physiological solution for 5 seconds (control, T1); immersion in physiological solution with 1000 ppm of organic acids for 5 seconds (T2); sprinkling of steam under pressure (4 bar) at 140 ºC (T3); and T2 after T3 (T4). An area of 100 cm^2 was sampled through superficial swabs for *Salmonella* counts by Most Probable Number method (MPN). Aspect, color, consistency, smell, and levels of fat, protein, pH, and moisture were also evaluated before and after each treatment. The use of steam associated to the immersion in organic acid solution showed better efficiency for reduction of superficial contamination (decreasing 100% of counts) but the better effectiveness was observed through the decreasing of 0.8 log of MPN at skin and 0.77 log of MPN at muscle by using the physiological solution (T1) and the organic acid solution (T2), respectively. The steam treatment associated with the organic acid solutions (T4) decreased the pH and increased moisture of pork legs, although it did not mischaracterize the quality (within required parameters for human consumption). All the other treatments did not change physical-chemistry features

KEYWORDS: organic acids; physicochemistry; pork leg; *Salmonella* sp.; steam.

AVALIAÇÃO MICROBIOLÓGICA E FÍSICO-QUÍMICA DE PERNIS SUÍNOS TRATADOS COM ÁCIDOS ORGÂNICOS E/OU VAPOR NO CONTROLE DA CONTAMINAÇÃO SUPERFICIAL POR *Salmonella* Typhimurium

RESUMO

Suínos portadores assintomáticos são o principal fator de risco para a contaminação de carcaças durante o processamento industrial. Diferentes formas de prevenção e controle têm sido testadas no pós abate, dentre elas o uso de vapor e/ou ácidos orgânicos, que podem ser alternativas de baixo custo e alta eficiência. No presente estudo, testou-se o uso de solução de ácidos orgânicos e aplicação de vapor sob pressão, usados isoladamente ou em associação. Como poucas pesquisas trazem informações sobre o fato desses tratamentos promoverem ou não mudanças nas características físico-químicas da carne suína, o presente experimento se propôs também a avaliar as possíveis alterações físico-químicas de pernis submetidos a esses tratamentos. Porções de pernil foram contaminadas artificialmente com *Salmonella* Typhimurium DT 177 e, posteriormente, divididas em quatro tratamentos: imersão em solução fisiológica por 5 segundos (controle, T1); imersão em solução fisiológica acrescida de ácidos orgânicos 1000 ppm por 5 segundos (T2); aspersão de vapor sob pressão de 4 bar à temperatura de 140 ºC (T3); e T3 seguido por T2 (T4). Após o tratamento, uma área de 100 cm^2 foi amostrada por meio de *swab* para quantificação da *Salmonella* residual pela técnica do número mais provável (NMP). Foram avaliados também o aspecto, coloração, consistência e odor, antes e após cada tratamento dos pernis, bem como análises físico-químicas visando à determinação do percentual de lipídeos, proteínas, pH, umidade e voláteis, também antes e após cada tratamento. Observou-se que o uso de vapor associado à imersão em solução de ácidos orgânicos a 1000 ppm apresentou melhor eficiência na redução das contaminações superficiais (100% de frequência de redução), porém a melhor eficácia foi observada através da redução de 0,8 log base 10 do NMP na pele e 0,77 log base 10 do NMP na musculatura através do uso de solução fisiológica (T1) e solução de ácidos orgânicos (T2), respectivamente. Os resultados obtidos revelaram que os tratamentos utilizados não interferiram nos atributos físico-químicos, como aspecto, cor, odor e consistência. O tratamento de vapor associado aos ácidos orgânicos diminuiu o pH e aumentou o teor de umidade e voláteis da carne, porém não descaracterizou a qualidade físico-química da carne suína, que permaneceu dentro de seus padrões ideais, apta ao consumo humano.

PALAVRAS-CHAVE: ácidos orgânicos; análises físico-químicas; pernil suíno; *Salmonella* sp.; vapor.

INTRODUCTION

Pork is rich in essential nutrients, and it is the most consumed animal protein worldwide, contributing to the achievement of a balanced diet. It has characteristic flavor and tenderness, besides being a source of vitamins and minerals (ODA, 2004).

In the growing market for pork production, agribusinesses seek to ensure increased production combined with profitability and food security for the consumer. There are many management tools available for pathogen control in production systems such as GMP (Good Manufacturing Practice) and HACCP (Hazard Analysis and Critical Control Points). Nevertheless, pathogens causing food poisoning, such as *Salmonella enterica*, can still be isolated in meat products in Brazil (FERRAZ et al., 2004; SPRICIGO et al., 2008). Brazilian law determines the absence of *Salmonella* sp. (ANVISA, 2001), but the production systems of pork and its derivatives still face problems of contamination of both carcasses in slaughterhouses and field produced swine. This information corroborates FERRAZ’s et al. (2004) conclusions about the importance of slaughtered *Salmonella* carrier swine and the contamination of the final product. Several studies have evaluated methods of microbiological control in meat (CONTE & MARIN, 2003), but with different methods and divergent results. Many alternatives for the treatment of swine carcasses after slaughter are being studied, for instance, the use of organic acids, the use of hot water in the form of steam and the association between thermal and chemical treatments.

Due to the deleterious effects that microbial contamination can cause to meat, and considering that treatments that can be used to decontaminate carcasses should not promote changes in their physical and chemical characteristics, the present study aimed at (a) finding an alternative to the surface control of *Salmonella* Typhimurium in artificially contaminated pork legs, evaluating the use of organic acids and steam under pressure, in both isolated and associated treatments, and at (b) assessing possible changes in their physicochemical traits.

MATERIAL AND METHODS

This study was divided into two stages, the first one was the microbiological evaluation of legs previously contaminated by *Salmonella* Typhimurium phagotype DT 177 and submitted to four treatments at the Bacteriology Laboratory of the Animal Microbiology Diagnostic Centre (CEDIMA). The second was the carry-out of physical and chemical analyzes of legs submitted to three treatments, at the Laboratory of Food Science, both from the Center of Agroveterinary Science (CAV / UDESC) from June to November 2008.

We used a total of 40 legs from a swine abattoir under Federal Inspection of the State of Santa Catarina. The legs were cooled and divided into four parts, with an average weight of about

3.5 kg each. The swine batch used was composed entirely of females, aged between 160 and 180 days.

For the first stage, the previously contaminated legs were submitted to four different treatments:

- T1: immersion in saline for 5 seconds;
- T2: immersion in acid saline solution at 1,000 ppm (organic acids solution with citric acid as the main constituent) for 5 seconds;
- T3: spraying of steam under pressure (4 BAR) at 140 °C for 15 seconds;
- T4: T3 follwoed by T2.

For the second stage, legs were submitted to three different treatments:

- T 1.1: immersion of the legs in organic acids solution (ascorbic acid, citric acid and lactic acid) at 1%, diluted in one liter of distilled water;
- T 2.1: spraying of steam by sprinkling generated by a device of the brand Vaporex®, with temperature between 120 and 140 °C, and operating pressure of 4 BAR for 15 seconds;
- T 3.1: T 2.1 followed by T 1.1.

To carry out the first stage of the experiment (microbiological evaluation) we used 40 swine legs with skin that were artificially contaminated by immersion in sterile physiological solution containing 10^3 UFC/mL of *Salmonella* Typhimurium – phage-type DT177. We divided the legs into four blocks according to the treatments and carried out ten replications for each. The legs were grouped into four blocks for the appropriate treatments, and 10 repetitions were performed for each treatment. Fifteen minutes after the contamination in solution with *Salmonella* Typhimurium, we collected samples from the surface of 100 cm^2 of skin and muscles of each leg, using sterile swabs. The legs were submitted to specific treatments and new samples were collected 15 minutes after the carry-out of each treatment, in order to evaluate the variation of the amount of bacteria per surface area.

To quantify *Salmonella*, we inoculated the swabs in 100 ml sterile buffered peptone water (BPW). We homogenized each solution and transferred the liquid phase to BPW tubes, according to the Most Probable Number (MPN) method per 100 mL for series of three tubes with inocula of 10; 1 and 0.1 mL (BRAZIL, 2003). Diluted samples were incubated for 20 hours at 37 °C and the isolation of *Salmonella* sp. was conducted by the means of Rappaport-Vassiliadis broth (42 °C for 24 hours) and XLD agar (37 ºC for 24 hours) (KICH et al., 2004). The results were expressed as MPN/100 cm^2 of sampled surface.

We tested three treatments to carry the second stage of the experiment (physicochemical analyses). We used ten legs per treatment, with double samples collected before and after each treatment in order to compare the results.

For carrying out the aspect, color, consistency, odor and physicochemical analyses, we collected approximately 500 grams of meat, before each treatment (as control group) and immediately after each treatment (T 1.1, T 2.1, T 3.1). These portions of meat were taken from different regions of the leg that did not reach great vessels or adipose tissues, according to BRAZIL (1999). After collection, we homogenized these portions of meat using a food multiprocessor.

These analyses were performed before and after each treatment, in order to verify if any of the treatments would alter these characteristics. We carried out a cooking test to complete the analisis and, after evaluating the odor, we boiled the material for another five minutes to observe the meat visual aspect (BRAZIL, 1999). The physicochemical analises aimed at determining the percentage of lipids, protein, pH and moisture, were conducted according to the official methodology described by BRAZIL (1999).

For statistical analysis of the microbiological assessment, we calculated frequencies of samples that reduced the MPN of *Salmonella* sp. after treatment compared to the initial MPN. These frequencies were compared using Fisher's exact test. As for the physicochemical analysis, we evaluated the results by means comparisons by *t* test for paired data, comparing before and after each treatment (SAS, 1999).

RESULTS AND DISCUSSION

By analyzing the results obtained in MPN in the skin, in the first step (microbiological evaluation), we observed a significant effect ($p < 0.05$) of the treatment in all four cases. The treatment with acid (T2) was the least effective in reducing contamination frequency and the simultaneous use of steam and acid (T4) was the most effective (Table 1). Figure 1 shows the mean of MPN and their standard errors before and after each treatment, and the greatest reductions of MPN were observed with the use of saline (T1) (0.8 log base 10), and acid solution (T2) (0.77 log base 10) in the skin and muscles, respectively. It is noteworthy that treatments with the physiological solution (T1) and steam (T3) showed intermediate results and did not differ significantly (p> 0.05) from the other two treatments.

The reduction of MPN in the treatment with saline (T1) can be attributed to the fact that water has a physical effect on the removal of bacterial cells of *Salmonella* typhimurium on the surface of swine carcasses. This confirms the effect of the mechanical action of the liquid on the removal of bacterial cells superficially deposited on tissues. CUTTER (1999) reported the same in a experimental study of surface contamination by *Salmonella* Typhimurium in bovine carcasses. Thus, the mechanical action of liquids during the washing of carcasses before cooling and storage can be a useful measure in the control of *Salmonella* sp. deposited on the surface of the carcasses.

Regarding the muscle, treatments with saline (T1) and steam (T3) only decreased the counting in approximately 40% of samples, whereas the two other treatments (T2 and T4) were more effective, reducing the count in 100% of the replicates (Table 1).

By analyzing the results of treatment with saline (T1), the frequency of counting reduction was higher in skin than in muscle. This can be explained by the fact that the skin surface is more uniform, presenting less irregularities for contaminats deposit. This argument can also justify the lower effect of treatment (T3) on muscle than on skin. Moreover, the surface irregularities can induce higher fluid retention and therefore lower homogeneous distribution of the heat conveyed by steam.

Although it is possible to infer that the treatment with the acidic solution (T2) can induce mechanical removal of salmonella as well as saline (T1), results regarding the muscles showed greater frequency reduction at an average log of (MPN) of *Salmonella* typhimurium than tresults regarding the skin. Therefore, we can state that the acid solution used in (T2) has a more effective action on muscles than on skin.

In this study, the isolated actions of treatments with steam under pressure (T3) and acidic solution (T2) were not satisfactory, and the low number of repetitions can be one of the reasons for the lack of clarification of the separate effects of each treatment. This study did not allow us to define the organic acids as effective chemical agents in isolated treatments to control *Salmonella* Typhimurium in superficial contamination of swine carcasses. Thus, we would suggest a new study with new design for more conclusive information.

NETTEN et al. (1995) found better results for the microbiological control by chemical and physical synergistic action, as GUERRERRO et al. (1995) also observed through the combination of organic acids and low temperatures. These results and the reduction of MPN at 100% of repetitions (T4), on both skin and muscle, suggest that high temperature and pressure steam spraying associated with the chemical and mechanical action of the acid solution justify the better performance of treatment (T4) in the microbiological control of *Salmonella* Typhimurium on the surface of swine legs.

TABLE 1. Percentage of swine leg samples which showed reduction in the count of the Most Probable Number (MPN) of *Salmonella* after each treatment for decontamination on both skin and muscle, withdrawing the samples with counts below 3MPN.

Treatament	Skin	Muscle
Saline (T1)	87.5% (7/8)^{ab}	44.4% (4/9)^{b}
Organic Acids (T2)	50.0% (4/8)^{b}	100.0% (10/10)^{a}
Steam (T3)	75.0% (6/8)^{ab}	44.4% (4/9)^{b}
Steam+Acid (T4)	100.0% (10/10)^{a}	100.0% (9/9)^{a}
Fisher’s exact test (P)	0.0486	0.0018

Percentages followed by different letters in columns differ significantly by Fisher's exact test (p <0.05).

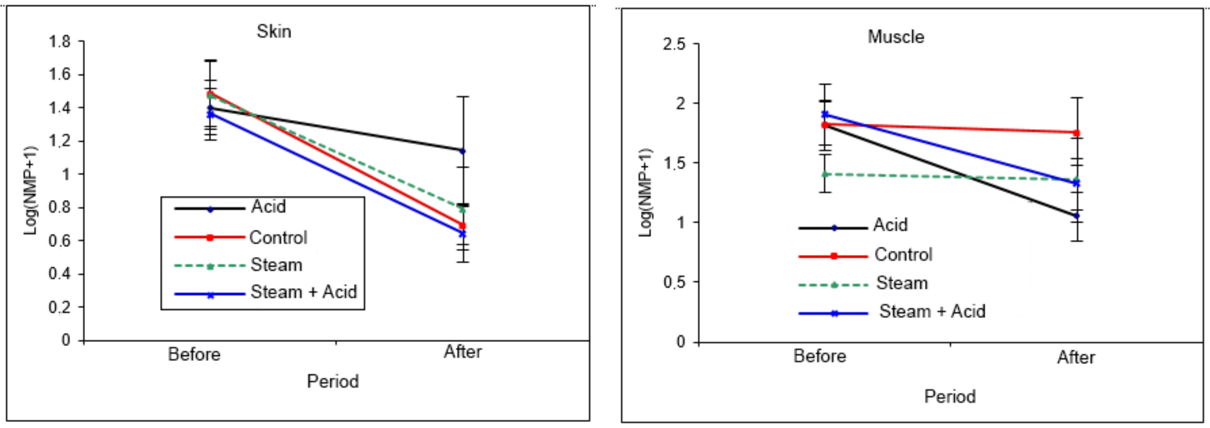

FIGURE 1. Means and standard errors of the Most Probable Number (MPN) of Salmonella sp. before and after each treatment to decontaminate the skin and muscle of swine legs. MPN transformed in logarithmic scale, withdrawing the samples with scores below 3 MPN.

For the second phase of the experiment (physicochemical evaluations), the results showed that the treatments did not affect the features appearance, color, odor and consistency of legs treated. These results confirm what some authors have found in other species (BELL et al., 1986; DUBAL et al., 2004), because, as consumer relies mainly on visual appearance to buy meat preoducts, it is essencial that the meat appearance is close to optimal, expected and desirable levels. Maintaining the stability of meat color during distribution, storage and marketing is also necessary. Thus, it is important that the treatments applied on the flesh aimed at reducing the surface microbial load does not change color or alter organoleptic characteristics.

According to BRAZIL (1999), the desirable features regarding pork appearance are uniformity, lack of blood, stains and slime. The cut surface of pork must show good marbling quality, without flabbiness and exudate. Pork must present uniform color between pink and red, a small layer of white, firm and compact fat, and a mild, pleasant and characteristic odor *(sui generis)*.

The results of this experiment confirm GOLDBACH & ALBAN's (2006), who studied swine carcasses treated with hot water, and found that changes in meat color may occur but are transient and disappear after cooling. GILL et al. (1999) tested hot water on bovine carcasses and verified that the treatment did not cause losses in meat appearance. JAMES et al. (2000) evaluated and tested the use of steam and hot water on lamb carcasses and verified no changes in meat appearance.

None of the proposed treatments altered the cooking test, regarding meat smell and consistency. This is an additional proof based on changes in consistency, odor and taste in food occurred in early stages of decomposition, enhanced by heating the sample. Ammonia, hydrogen sulfide or rancid odor are easily identified (BRAZIL, 1999).

Treatments T 1.1 and T 2.1 did not alter the physicochemical properties of meat (Table 2). VASCONCELOS et al. (2002) studied sheep carcasses treated with acetic acid and did not find any changes in meat pH after treatment. However, treatment T 3.1 decreased meat pH and increased moisture content, but meat did not present uncharacteristic featuers and was suitable for human consumption. These alterations could be transient as shown by some studies.

We observed that treatment T 3.1 decreased the pH, which is probably due to the increased penetration power of the acid by the concomitant steam application, and the fact that the analyzes were performed immediately after the treatment application. ÖZDEMIR et al. (2006) treated bovine carcasses with hot water followed by lactic acid and found that the pH declined after the treatment; however, after storaging the carcass under refrigeration at 4 °C for 5 days, the pH returned to normal. According to FIORUCCI et al (2002), lactic acid causes a small, temporary reduction in meat pH, but the pH returns to normal levels from 48 to 72 hours after application.

Treatment T 3.1 increased the moisture content of the meat, presumably due to its association, altering the water holding capacity (WHC) of the meat. According to SILVA & SOARES (2001), the use of substances such as organic acids on meat can change its property of retaining water. Thus, steam heat may have increased capacity of the meat absorbing the organic acid solution (decreasing the pH), reducing the water retention capacity of the meat and increasing its moisture content. KOZEMPEL et al. (2003) reported that steam is able to open the pores of the meat surface, supporting the argument that it increased the capacity of absorbing organic acids.

TABLE 2: Means, standard deviations and descriptive level of probability of t test for paired data for lipids, protein, moisture and pH in relation to different treatments and physicochemical composition of swine meat

Treatament	Period Before	Pr> t * After
Lipids		
Steam (T 2.1)	1.91±0.063	1.91±0.065
Steam + Organic acids (T 3.1)	2.09±0.060	2.09±0.057
Organic acids (T 1.1)	2.09±0.064	2.10±0.061
Protein		
Steam (T 2.1)	22.18±0.159	22.19±0.137
Steam + Organic acids (T 3.1)	21.50±0.178	21.50±0.113
Organic acids (T 1.1)	21.75±0.078	21.74±0.195
Moisture		
Steam (T 2.1)	75.87±0.135	74.90±0.776
Steam + Organic acids (T 3.1)	75.81±0.180	76.28±0.187
Organic acids (T 1.1)	76.08±0.184	75.88±0.335
pH		
Steam (T 3.1)	6.11±0.088	6.11±0.107
Steam + Organic acids (T 2.1)	5.95±0.028	5.88±0.034
Organic acids (T 1.1)	6.12±0.116	6.11±0.131

* Descriptive level of probability of t test ($p<0.05$)

CONCLUSIONS

Using steam associated with the immersion of meat in an organic acids solution at 1000 ppm is an effective action to control pork superficial contamination by *Salmonella* Typhimurium.

Soaking pork in saline produces a mechanical action that results in good efficacy in reducing the MPN of *Salmonella Typhimurium* surface contamination of pork skin.

The treatments did not alter meat characteristics as appearance, color, odor and consistency. Likewise, when applied alone, they did not alter the physicochemical properties of meat (pH, lipid, protein and moisture). Treatment with steaming associated with the use of organic acids caused a slight decrease in pH and an increase in moisture content; however, the meat remained adequate for human consumption, because their color characteristics, odor, flavor and consistency remained unchanged.

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REFERENCES

ANVISA. Agência nacional de vigilância sanitária. **Ministério da Saúde**. Resolução RDC nº12 de 02 de janeiro de 2001.

BELL, M. F., MARSHALL, R. T., ANDERSON, M. E. Microbiological and sensory tests of beef treated with acetic and formic acids. **Journal of Food Protection**, v. 49, n. 3, p. 207-210, 1986.

BRASIL. Ministério da Agricultura Pecuária e Abastecimento. Instrução Normativa nº 20 de 21 de julho de 1999. Oficializa os métodos analíticos físico-químicos para controle de produtos cárneos e seus ingredientes – sal e salmoura. **Diário Oficial da União**, Brasília, 27 de agosto de 1999. Disponível em <*www.**agricultura**.gov.br/arq_editor/file/Aniamal/.../laba* *lac.pdf*> Acesso em 25 jul 2013.

CONTE, A. P. F; MARIN, J. M. Selection of 5-fluorocytosine-resistant mutants from an *Aspergillus niger* citric acid-producing strain. **Brazilian Journal of Microbiology**, São Paulo, v. 34, n. 1, p. 1-4, 2003.

CUTTER, C.N. Combination spray washes of saponin with water or acetic acid to reduce aerobic and pathogenic bacteria on lean beef surfaces. **Journal of Food Protection**, v. 62, n. 3, p. 280-283, 1999.

DUBAL, Z.B., PATURKAR, A. M., WASKAR, V. S., ZENDE, R. J., LATHA, C., RAWOOL, D. B., KADAM, M.M. Effect of food grade organic acids on inoculated *S. aureus, L.monocytogenes, E. coli and S. Typhimurium* in sheep/goat meat stored at refrigeration temperature. **Meat Science**, v. 66, n. 4, p. 817–821, 2004.

FERRAZ, S. M. ; SCHWARZ, P ; CANAL, C. W ; CARDOSO, M. R. I. Prevalência de suínos portadores de Salmonella sp. ao abate e contaminação de embutidos tipo frescal. **Acta Scientiae Veterinariae**, v. 32, n.2, p. 141-147, 2004.

FIORUCCI, A. R., SOARES, M. H. F. B., CAVALHEIRO, E. T. G. Ácidos Orgânicos: dos primórdios da química experimental à sua presença em nosso cotidiano. **Química Nova na Escola**, n° 15, maio/2002; Disponível em <http://www. *qnesc.sbq.org.br/online/qnesc15/v15a02.pdf*>, acesso em 24, jul,2013.

GILL, C. O., BRYANT, J., BEDARD, D. The effects of hot water pasteurizing treatments on the appearances and microbiological conditions of beef carcass sides. **Food Microbiology**, v. 16, p. 281- 289, 1999.

GOLDBACH, S. G., ALBAN, L. A cost-benefit analysis of *Salmonella*-control strategies in Danish pork production**. Preventive Veterinary Medicine,** n 77, p 1-14, 2006.

GUERRERO, I., MENDIOLEA, R., PONCE, E., PRADO, A. Inoculation of Lactic Acid Bacteria on Meat Surfaces as a Means of Decontamination in Semitropical Conditions. **Meat Science**, v. 40, p. 397-411, 1995.

JAMES, C., THORNTON; J. A., KETTENRIGHAM, L., JAMES, S. J. Effect of steam condensation, hot water or chlorinated hot water immersion on bacterial numbers and quality of lamb carcasses. **Journal of Food Engineering**, v. 43, p. 219-225, 2000.

KICH, J. D.; BOROWSKY, L.; SILVA, V.; RAMENZONI, M.; TRIQUES, N.; CARDOSO, M. R. I**.** . Avaliação da atividade antibacteriana de desinfetantes comerciais frente a amostras de Salmonella Typhimurium isoladas de suínos. **Acta Scientiae Veterinarie**, v. 32, n.1, p. 33-39, 2004

KOZEMPEL, M.; GOLDBERG; CRAIG, J. C. Jr. The vacuum/steam/vacuum process. **Food Technology**, v. 57, n. 12, p. 30-33, 2003

NETTEN, P.; MOSSEL, D.A.A.; VELD, H. Lactic acid decontamination of fresh pork carcasses:a pilot plant study, **International Journal of Food Microbiology**, v. 25, p.1-9, 1995

ODA, E. I. Carnes PSE e DFD em aves e suínos: diferenças e semelhanças. **Revista Nacional da Carne**, n. 325, p. 108-113, 2004.

ÖZDEMIR, H.; YILDIRIM, Y., KÜPLÜLÜ, O., KOLUMAN, A., GÖNCÜOGLU, M., INAT, G. Effects of lactic acid and hot water treatments on *Salmonella*Typhimurium and *Listeria monocytogenes* on beef. **Food Control**, v. 17, p. 299–303, 2006.

SAS INSTITUTE (CARY NC). SAS **User Guide:** Statistical Analysis System, release 8.2, 1999; Disponível em <*ww.ufrrj.br/institutos/it/deng/.../**SAS**_o_essencial_da_linguagem.pdf*>, acesso em 24 jul 2013.

SILVA, J. A; SOARES, L. F. Sanitização de Carcaças de Frango com Ácidos Orgânicos Comerciais e Suco de Limão. **Revista TeC Carnes**, v. 3, n. 1, p. 19-26, 2001.

SPRICIGO, D. A.; MATSUMOTO, S. R.; ESPÍNDOLA, M. L.; FERRAZ, S. M. Prevalência, quantificação e resistência a antimicrobianos de sorovares de Salmonella isolados de lingüiça frescal suína. **Ciência e Tecnologia de Alimentos**, v. 28, p. 1-7, 2008.

VASCONCELOS, E. C., ZAPATA, J. F. F., FIGUEIREDO, E. A., BRANCO, M. A. A. C., BORGES, A. S. A microbiota da carcaça e da carne ovina tratada com ácido acético, embalada a vácuo e maturada por 48 dias. **Ciência e Tecnologia de Alimentos**, Campinas, v. 22, n. 3, 2002.

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