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EFFECT OF RADIATION, HYDROGEN PEROXIDE AND CHLORINE ON BACTERIAL DECONTAMINATION OF BROILER CARCASSES

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ABSTRACT

Eighty random samples of broiler chicken carcasses were collected from an automatic poultry slaughtering plant in Dakahlia Governorate just after washing in the chiller and divided into four groups each group consists of 20 chicken carcasses which subdivided into two subgroups (ten carcasses for each subgroup). The 1st group left as a control. The 2nd group decontaminated with 25ppm chlorine (first subgroup), the other subgroup decontaminated with 50ppm chlorine for three minutes. The results were reduced significantly when compared with corresponding control statistically for both concentrations. The 3rd group were decontaminated with 1% hydrogen peroxide (first subgroup) and the other subgroup decontaminated with 2% hydrogen peroxide for two minutes Where the Aerobic Plate Counts (APCs), Most Probable Number (MPN) count and the counts of *Staph.aureus* were reduced significantly when compared with corresponding controls ($P<0.05$). The 4th group decontaminated with gamma rays first subgroup irradiated with 2Kilo Gray(KGy) and the other subgroup irradiated with 3KGy. The irradiation results of APCs were reduced significantly ($P<0.05$) after irradiation with 2KGy and 3KGy, MPN count were reduced significantly ($P<0.05$) after irradiation by 2KGy and no growth after irradiation with 3KGy and the counts of *Staph.aureus* were reduced significantly ($P<0.05$) after irradiation with 2KGy and no growth after irradiation by 3KGy.

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Key words: *Broiler, irradiation, Staph.aureus, hydrogen peroxide.*

INTRODUCTION

Poultry meat is a nutritious food consumed all over the world because of its relatively low cost and low fat content, however it is highly perishable with a relatively short shelflife even where it is kept under refrigeration. Although poultry constitute an excellent source of high quality and easily digested protein it is liable to transmit different types of potentials to consumers Zahra, (2001). Many attempts were applied to reduce the microbial findings and level of poultry meat contamination through using variable chemicals like sodium hypochlorite which resulted in significant reduction in the number of microorganisms, Ismail *et al.* (2001). One of the newly emerging technologies to ensure microbiological safety of meat is radiation processing Kanatt *et al.* (2005). The safety and efficacy of irradiation in food preservation has been thoroughly demonstrated world wide Pelezar *et al.* (1997). The radiation doses required to inactivate 90% of the common foodborne pathogens associated with meat and poultry are in the range of 1-4KGy, Thayer *et al.* (1993). Contamination of poultry carcasses with *Staph. aureus* usually occurs through handling by human, if such products are left without refrigeration for several hours or cooled slowly in refrigerator, growth of *Staph.aureus* and enterotoxin formation may occur, growth of *Staph.aureus* in the product is favoured by lack of competitive bacteria which are destroyed by heat, Bryan (1980). The aerobic plate counts (APCs), Most Probable Number of Coliforms (MPN) and counts of *Staph.aureus* in chicken carcasses collected from different poultry shop were $3.38x10^6 \pm 1.02x10^6$, $2.58x10^4 \pm 0.9x10^4$ and $9.95x10^3 \pm 1.56x10^3$cfu/gm respectively, Morshdy *et al.* (2008). While the counts were 6.1 ± 0.1, 2.7 ± 0.1 and 3.7 ± 0.1 $\log_{10}$ cfu/gm respectively in fresh whole chicken broiler carcasses, Mira; Eskandar, (2007) also Mahmoud; Hammouda (2006) examined thigh and breast and found that APCs and *Staph.aureus* count were $1.4x10^6 \pm 4x10^5$ and $6x10^5 \pm 2x10^4$ while *Staph.aureus* count were $8.9x10^3 \pm 0.3x10^3$ and $2.7x10^3 \pm 1.7x10^4$ respectively. May (1974) mentioned that final rinsing of commercially slaughterd broilers with 25 ppm chlorine resulted in significant reduction in TVCs., Whyte *et al.* (2001) proved that final

rinsing of poultry carcasses with 25ppm chlorine reduce TVCs from 4.98±0.38 to4.52±0.24 and Enterobacteriaceae from 3.37±0.31 to 3.16±0.16 log_{10}cfu/gm. Commercial chlorine chiller on poultry carcasses during processing reduce APCs, *Staph.aureus* from $6.2x10^4$ and $1.4x10^4$ to. $2.4x10^4$ and $6x10^3$cfu/gm respectively Gelis; Kabul (2006), while Karen *et al.* (2010) mentioned that significant reduction in APCs and Coliforms by 50 ppm chlorine rinsing for three minutes the reduction were 0.4 and 0.21 log_{10} cfu/gm respectively. Hydrogen peroxide is highly unstable and breaksdown into water and single oxygen molecule which is strong oxidizing and disinfecting agent Black *et al.* (2008), also several investigators concluded that chlorine and hydrogen peroxide were most frequently used in commercial poultry processing as antimicrobials due to their availability, low cost and efficacy Bolder, (1997); Hugas; Tsigarida (2008); Northcutt; Jone (2004), also Mostafa (2010) concluded that Hydrogen peroxide 0.1% in chiller reduce APCs, Coliforms and *Staph.aureus* counts by 97.3%,72.01% and 94.9 % respectively and similar results were recorded by EL-said *et al.* (2002). Oliveira *et al.* (2009) mentioned that gamma radiation 1.5 and 3KGy on chicken breast reduce APCs from 3.0±0.01 to less than 1.0 and total Coliforms reduced from 2.9±0.3 to 0.3±0.4 and *Staphaureus* to less than 0.5 log_{10} cfu/gm respectively. Also Min *et al.* (2007) reported that no viable cells were detected after exposure of inoculated chicken thigh and breast to 2KGy radiation. Irradiation with air and vacuum reduce mesophiles from 4 and 3.8 to 3.3 and 2.7 log_{10}cfu /gm at 2KGy and 1.7 and 1.6 log_{10} cfu/gm at 3KGy while Coliforms reduced from 2.2 and 3 log_{10} cfu/gm to 0.6 and 0.5 log_{10} cfu/gm by 2KGy and not detected at 3KGy respectively Mantilla *et al.* (2011). The results recorded by Mohamed *et al.* (2008) on fish fillets exposed to radiation on APCs, Coliforms and *Staph.aureus* were in control samples 6.4,2.68 and 2.38log_{10} cfu/gm while at 2KGy were 2.6, less than 3 and less than 2log_{10} cfu/gm and at 3KGy were 2.1, less than 3 and less than 2 log_{10} cfu/gm respectively.

Therefore this study focused on the effect of Chlorine, Hydrogen peroxide and Irradiation on bacterial population, Coliforms and *Staph.aureus* in whole chicken carcasses from a local poultry plant with comparison between the three methods.

MATERIALS and METHODS

Total number of 80 broiler chicken carcasses from an automatic poultry slaughtering plant in Dakahlia, Egypt were collected after complete preparation (slaughtering, scalding, defeathering and evisceration), just afterwashing in the chiller where they divided into four groups. The first group [control group] were placed in a chiller containing cold water for 60 minutes and left as a control.

The second group were divided into two subgroup (each one ten carcasses) one of them rinsed with 25ppm chlorine (sodium hypochlorite 14% vol./vol.) and the other one rinsed with 50 ppm chlorine for three minutes then sampling.

The third group were also divided into two subgroup (each oneten carcasses) one of them rinsed with 1% hydrogen peroxide and the other one rinsed with 2% hydrogen peroxide for two minutes.

The fourth group divided into two subgroups (each one ten carcasses), 250 gm from each carcass were packed in a sterile polyethylene bags heat sealed then irradiated at National Center for Radiation Research and Technology (NCRRT) Nasr City, Cairo. The irradiation source was Cobalt 60 irradiation model ISS LEDDVATED. The dose rate was established using alanine transfer dosimeter and variation in the absorption of irradiation dose was minimized by placing the samples within a uniform area of the irradiation field. One subgroup exposed to 2KGy and the other subgroup exposed to 3KGy of gamma rays. After irradiation 25gm of each exposed samples were homogenized with 225ml of 0.1% peptone water in a stomacher for 2.5 minutes at 3000rpm followed by ten fold serial dilution in 0.1% peptone water Each of the prepared samples were examined for enumeration of its microorganisms content as follows.

1-Total Colony Count:
0.1 ml of each dilution was evenly spread over the dry surface of standard plate count agar using a sterile bent glass spreader only plates contain 30-300 colonies were counted as the total colony per gram of samples according to APHA (1985).

2-*Coliform Count:*
Using most probable number technique (MPN) according to ICMSF (1978).

3- *Staph. Aureuscount:*
Using Baired Parker agar medium with eggyolk tellurite emulsion APHA (1992).

Biochemical Identification of *Staph.aureus*;
Identification of *Staph.aureus* according to APHA (1992); films were prepared from the pure culture of isolated organisms stained with Gram's stain and examined microscopically for G+ve cocci.

The tests for identification were motility, catalase test, mannitol test and coagulase test (tube method).

RESULTS

Table 1: Statistical analytical results for effect of chlorine on decontamination process.

Microbial count log mean±S.E.	control	after decontamination with 25ppm chlorine	after decontamination with 50 ppm chlorine
APC	5.73 ±3.23	5.71±3.08*	5.62±3.57*
MPN	3.74±1.45	3.69±1.07*	3.55±1.44*
Staph.aureuscount	3.62±1.59	3.58±1.17*	3.6±1.1*

APC=aerobic plate count & MPN=most probable number of coliforms & *Staph.aureus*=staphylococcus aureus count, NG=no growth.

- Means results of decontamination are significantly different ($P<0.05$)

Table 2: Statistical analytical results for effect of hydrogen peroxide on decontamination process.

Microbial countlog mean±S.E.	control	after decontamination with 1%H_2O_2	after decontamination with 2%H_2O_2
APC	5.73 ±3.23	5.34±2*	5.20±2.47*
MPN	3.74±1.45	3.53±1.46*	3.30±1.04*
Staph.aureuscount	3.62±1.59	3.44±1*	3.30±1.07*

Table 3: Statistical analytical results for effect of gamma rays irradiation on decontamination process.

Microbial countlog mean±S.E.	Control	after decontamination with 2KGy	after decontamination with 3KGy
APC	5.73 ±3.23	4.92±2.11*	4.80±0.1*
MPN	3.74±1.45	2.87±0.1*	NG*
Staph.aureus count	3.62±1.59	2.93±0.01*	NG*

DISCUSSION

The sanitary evaluation of slaughtered birds is based mainly on its microbial findings and level of contamination. Many investigators proved that more than 70 % of poultry carcasses are contaminated and the microbial count being more than $3x10^5$cfu/ml, Tamblyn *et al.* (1997).

Table (1) shows that the mean log_{10}cfu/gmvalues of APCs in examined control samples, after decontamination with 25ppm and 50ppm chlorine were 5.73 ±3.23,5.71±3.08 and 5.62±3.57 log_{10}cfu/gm. while MPN count were 3.74±1.45, 3.69±1.07 and 3.55±1.44log_{10}cfu/gm and the counts of *Staph.aureus* were 3.62±1.59,3.58±1.17 and 3.6±1.1 log_{10}cfu/gm respectively, the results in control samples nearly in accordance with Bryan (1980), Tamblyn *et al.* (1997); Mahmoud, Hammouda (2006); Mira, Eskandar (2007) and Morsh-dy *et al.* (2008). The higher microbial load could be attributed to the technique of evisceration and poor hygienic levels. The reduction were more than those reported by Whyte *et al.* (2001) and similar to Gelis, Kabul (2006); Stopforth *et al.* (2007) and Karen *et al.* (2010) the difference in reduction may be due to the final processing and rinsing before decontamination process. The results were reduced significantly when compared statistically with corresponding controls ($P<0.05$).

Table (2): The results of decontamination with hydrogen peroxide (H_2O_2) declared that. The mean log cfu value of APCs were 5.73±`3.23,5.34±2, and 5.20±2.47 log_{10} cfu/gm after decontamination with hydrogen peroxide 1% and 2% respectively while MPN count were 3.74±1.45,3.53±1.46 and 3.30±1.04 log_{10}cfu/gm and the counts of *Staph.aureus* were 3.62±1.59,3.44±1 and 3.30±1.07 log_{10}cfu/gm respectively similarly with those recorded by Bolder (1997); Northcutt, Jones (2004); Black *et al.* (2008) and Hugas; Tsigarida (2008), higher results recorded

by EL-Said *et al.* (2002) and Mostafa (2010), while the reduction after 2%H_2O_2 were similar to Northcutt, Jones (2004) and lower than the results recorded by Mostafa (2010). The results were reduced significantly when compared with corresponding controls ($P<0.05$).

The results in Table (3) declared that the mean log cfu value of APCs were 5.73±3.23,4.92±2.11 and 4.80±0.1 log_{10}cfu/gm in examined control samples, after irradiation with 2KGy and 3KGy respectively while MPN count were 3.74±1.45,2.87±0.1 log $_{10}$cfu/gm respectively in control samples and after irradiation with 2KGy and no growth after irradiation with 3KGy and the counts of *Staph.aureus* were 3.62 ±1.59 and 2.93±0.05 log_{10}cfu/gm respectively in examined control samples and after irradiation with 2KGy and no growth after irradiation with 3KGy the reduction after irradiation nearly similar to Mohamed *et al.* (2008); Oliveira *et al.* (2009) and Mantilla *et al.* (2011), while higher reduction recorded by Min *et al.* (2007) the reduction after irradiation in APC in addition to no growth of Coliforms and *Staph.aureus* were similarly to Mohamed *et al.* (2008); Oliveira *et al.* (2009) and Mantilla *et al.* (2011). The effects of irradiation as log viable counts of all the inspected bacteria showed gradual reduction as the irradiation dose increased. The results were reduced significantly when compared with corresponding controls ($P<0.05$).

In conclusion from the aforementioned results declared that chlorine is a bactericidal, viricidal and fungicidal agent can be successfully applied in the food industry and may conistitute an important factor in reducing losses resulting from food spoilage and human infection.

The use of hydrogen peroxide in chiller used for chicken chilling prior to its packaging greatly reduce its bacterial load. It is a power full sanitizer for poultry carcasses, it enhances its meat quality.

Radiation did not affect the sensory character of chicken carcasses and produces no apparent changes in the organoleptic characteristics, it inhibit microbial growth and so extends the shelf life of the product. From the results it was obvious that the microbial reduction was greater after radiation than after hydrogen peroxide and after hydrogen peroxide than after chlorine.

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تأثير الإشعاع وماء الأكسجين والكلور على إزالة التلوث البكتيري لذبائح بدارى التسمين

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اشتملت الدراسة على عدد ثمانين عينة لذبائح بدارى التسمين تم تجميعها من احدى المجازر الآلية بمحافظة الدقهلية بعد انتهاء مراحل الذبح والتنظيف والتجويف والغسيل بالماء البارد مباشرة حيث تم تقسيمها إلى أربعة مجموعات: الأولى: لم يتم معالجتها واستخدمت كدليل عددها عشرون عينة ، الثانية: تم تقسيمها إلى مجموعتين كل منها عشرة عينات حيث تم غسل المجموعة الأولى منها بمحلول الكلور ٢٥ جزء في المليون والمجموعة الثانية تم غسلها بمحلول الكلور ٥٠ جزء في المليون لمدة ثلاث دقائق ، الثالثة: تم تقسيمها إلى مجموعتين كل منها عشرة عينات حيث تم غسلها بمحلول ماء الأكسجين ١ % ومحلول ماء الأكسجين ٢ % لمدة دقيقتين ، الرابعة: تم تعريضها لأشعة جاما بعد تقسيمها إلى مجموعتين كل مجموعة عشرة عينات حيث تم تعريض المجموعة الأولى إلى أشعة جاما ٢ كيلو جراي والمجموعة الثانية تم تعريضها لأشعة جاما ٣ كيلو جراي ، وأظهرت النتائج أن متوسط العد الكلي للميكروبات الهوائية في المجموعة الثانية وكذا العد الاحتمالي للميكروبات المعوية وميكروب المكور العنقودي الذهبيه نسبة نقص طفيف، أما بالنسبة للمجموعة الثالثة فكان العد الكلي للميكروبات الهوائية والعد الاحتمالي للميكروبات المعوية والعد الكلي لميكروب المكور العنقودي به نسبة نقص وفي المجموعة الرابعة والتي تم تعريضها لأشعة جاما فكان العد الكلي للميكروبات الهوائية به نسبة نقص والعد الاحتمالي للميكروبات المعوية به نسبة نقص عند ٢ كيلوجراى وكانت العينات سلبية بعد التعرض لاشاعة جاما ٣ كيلوجراى وكذلك العد الكلي لميكروب المكور العنقودي الذهبى به نسبة نقص عند ٢ كيلوجراى وكانت العينات سلبية بعد التعرض لاشاعة جاما ٣ كيلوجراى مما سبق يتضح أن ماء الكلور هو أضعف المواد المستخدمة في التأثير على الميكروبات بصفة عامة يليه ماء الأكسجين ثم أشعة جاما والتي أظهرت أعلى تأثير دون أن تغير في الصفات الطبيعية للحوم المعرضة للأشعة.