Original Research

Journal of Advanced Veterinary Research
(2024) Volume 14, Issue 7, 1150-1155

Synergism between *Saccharomyces cerevisiae* probiotic and rosemary nano-emulsion: Effect on broiler chicken meat quality and shelf life

Asmaa M. Abd-Elrahman[1], Salwa M. Hafez[2], Ghada H, Ali[1], Mohamed A. Kandeil[3], Abdelrahim H.A. Hassan [4,5*]

[1]*Biochemistry, Nutritional Deficiency Diseases and Toxicology Department, Animal Health Research Institute, Beni-Suef Lab. (AHRI), Agriculture Research Center (ARC), Egypt.*
[2]*Food Hygiene Department, Animal Health Research Institute, Beni-Suef Lab (AHRI), Agriculture Research Center (ARC), Egypt.*
[3]*Biochemistry Department, Faculty of Veterinary Medicine, Beni-Suef University, Egypt.*
[4]*School of Biotechnology, Nile University, Giza, 12588, Egypt.*
[5]*Department of Food Safety and Technology, Faculty of Veterinary Medicine, Beni-Suef University, Egypt.*

ARTICLE INFO

Recieved: 29 June 2024

Accepted: 21 August 2024

***Correspondence:**

Corresponding author: Abdelrahim H.A. Hassan
E-mail address: abdelrahim@nu.edu.eg

Keywords:

Broilers, Deteriorative indices, Microbiology, Probiotics, Rosemary, Shelf life.

ABSTRACT

Although several studies have investigated the effect of either probiotic feed additives or postmortem meat treatment on the quality of obtained chicken meat, the impact of combined treatment with probiotic feed additives along with meat dipping in essential oil nano-emulsion on meat shelf-life is barely examined. Therefore, this study investigated the effect of combined treatment with *Saccharomyces cerevisiae* yeast (SCY) and rosemary oil nano-emulsion (RNE) on the quality and shelf-life of chilled broiler meat. The experimental part consisted of adding SCY as a feed additive to broiler ration and/or dipping the resulting chicken meat in RNE 10% for 24 hours. Afterward, chicken meat from different treated groups, as well as the control one was refrigerated (4±1°C) and periodically examined on 0, 3rd, 7th, and 9th days of storage. The obtained results revealed significant reductions in total colony, Total *Enterobacteriaceae*, total *Staphylococci*, and total fungal counts of the SCY+RNE-treated group were reduced by about 79.4%, 34%, 72.8% and 32.5% as compared to control, respectively ($p<0.001$). While of RNE-treated group, they were decreased by about 71.6%, 16.5%,14.4% and 26% as compared to control, respectively ($p<0.001$). Whereas of the SCY-treated group, they were reduced about 57.2%, 59%, 28.5% and 24.7% as compared to control, respectively ($p<0.001$). Additionally, meat spoilage indicators (pH, TBA-RS, TVBN) came in harmony with the microbiological results. As control group samples had the highest values of pH, TBA-RS, and TVBN, followed by the RNE-treated group and the SCY-treated group. On the other hand, the SCY+RNE-treated group showed the lowest pH, TBA-RS, and TVBN levels ($p<0.001$). These results confirm that treatment with SCY alone, RNE alone, and SCY+RNE prolonged the shelf-life of broiler chicken meat. To conclude, the addition of SCY as a probiotic additive to chicken feed in combination with meat dipping in RNE has a potential synergistic favorable effect on chicken meat quality and shelf-life.

Introduction

Improving the shelf-life of meat is considered significant from the economic and nutritional points of view. The antioxidant capacity of meat including chicken meat depends mainly on the activities of endogenous reducing enzymes at early postmortem time (Serpen *et al*., 2012). This capacity decreases when muscular cells lose their homeostasis, consequently, the formation of free radicals is increased leading to meat spoilage and deterioration. Moreover, meat storage stability depends on several internal factors, such as metal catalysts, fatty acid profile, pH, and the presence of other inhibitors (Xiong, 2000; Min and Ahn, 2005).

Lipid and protein deterioration is considered the priority issue affecting meat quality characteristics (Ahn *et al*., 2009; Lund *et al*., 2011). In the meat industry, they have extensively used synthetic antioxidants, yet, because of the negative impression regarding the use of synthetic antioxidants; natural antioxidants (NA) are considered a suitable replacement (Velasco and Williams, 2011). Moreover, several studies reported possible carcinogenic and toxicological effects of the synthetic antioxidants that are used in human foods (Kumar *et al*., 2015). Therefore, recently consumers have preferred food that is free of any chemical additives.

Essential oils (EOs) are aromatic and volatile lipophilic plant extracts with antimicrobial and antioxidant properties which could be a suitable alternative to synthetic chemical preservatives (Prakash *et al*., 2015). The direct incorporation of EOs in the polymeric matrix has disadvantages, including volatilization of EOs during water evaporation due to the coarseness of its droplets and the creation of porous structures in the coating (Tastan *et al*., 2016). Therefore, the application of EOs in the form of nano-emulsions with tiny droplet sizes has many advantages including, high stability, enhancement of physicochemical properties, and improved biological properties by increasing the specific surface area and hence lowering the required amounts of the active component (Mushtaq e.al., 2023). Nano-emulsions are emulsions with particle sizes of 2–200 nm produced by high-energy or low-energy emulsification methods (McClements and Rao, 2011; Meneses *et al*., 2019). The application of essential oil nano-emulsions is an emerging technique that is used in the food industry to prolong the shelf-life, manage food safety concerns, and replace synthetic preservatives (Amin, 2013).

Rosemary is a plant source of bioactive substances containing phenolic compounds with antioxidant properties such as diterpenes, carnosic acid, carnosol, and rosmarinic acid (Chao *et al*., 2020). Nano encapsulation form of these phenolic compounds avoid exposure to oxygen, heat, humidity and light to protect them to achieve a higher antioxidant activity (Duarte and Larroza, 2019; Rashidaie *et al*., 2019). Achieving increased stability, protection, controlled release and reducing the possible adverse impact on the organoleptic properties in meat and meat products (Duarte and Larroza, 2019). Rosemary (*Rosmarinus officinalis* L.) has a wide antimicrobial range (El Bayomi *et al*., 2021). It was added to food because of its palatability, safety for consumers, and delaying effect on fat rancidity in meat (Yu *et al*., 2002; Jongberg *et al*., 2013). There is no limitation on using rosemary essential oil and its extract in meat products as it is generally recognized as safe (GRAS) (European Food Safety Authority, EFSA 2008). Moreover, rosemary extract and its essential oils were reported as potential antioxidant substances that are widely used in the food industry (Balentine *et al*., 2006; Hussain *et al*., 2010). Accordingly, the application of rosemary nano-emulsion (RNE) could be a promising approach to hinder the deterioration of chicken meat.

Additionally, probiotics are a significant feed additive that has the potential to increase antimicrobial resistance and reduce enteric diseases in poultry and subsequent contamination of poultry products (Patterson and Burkholder, 2003). Probiotics are microorganisms, such as fungi and

ISSN: 2090-6277/2090-6269/

bacteria, living in nature and have beneficial effects on the host (Khan *et al*., 2011). The most widely used type of yeast, *Saccharomyces cerevisiae* (SC), has been reported to improve feed quality and the performance of animals, as well as work as an immune stimulant to the host. The cell wall of SC contains chitin, mannan, and glucagon, working as immune stimulants and prebiotic sources (Li and Gatlin, 2003). Moreover, the yeast cell wall contains 1,3~1,6 D glucagon and mannan-oligosaccharides which act as natural growth promotors (van Leeuwen *et al*., 2005; Ghosh *et al*., 2007). In addition, the SC cell wall can minimize the toxic effect of aflatoxins in poultry through biodegradation (Parlat and Oguz 2001). Consequently, the addition of SC to broiler chicken ration could enhance the quality, safety, and stability of the resulting meat.

Despite there are several studies have examined the effect of probiotics as feed additives on the quality of meat products, to our knowledge, the investigation of probiotic application as ration additives along with meat treatment by essential oil nano-emulsion on the quality and shelf-life stability of poultry meat is very limited.

Therefore, the current study aimed to investigate the impact of the combined use of SC as a probiotic feed additive and RNE as a food additive on the microbiological and physicochemical status, as well as the shelf-life of obtained broiler meat, in comparison with individual use of each treatment and control (untreated) group.

Materials and methods

Probiotics

Saccharomyces cerevisiae is a probiotic that contains live yeast cells (20 billion CFU/g), active dry yeast (*Saccharomyces cerevisiae* Sc 47 (Allgau vet German. Allgau yeast, Germany). It is used in a dose of 2.5 billion CFU/Kg ration (Maksimović *et al*. 2022).

Rosemary nano-emulsion 10%

The nano-emulsion was prepared in the Nanomaterial's Research and Synthesis Unit (Animal Health Research Institute, Doki, Giza, Egypt) by mixing rosemary oil (10 ml) and Tween 80 (10 ml) for half an hour in a homogeneous blender (1500 watts), then 80 ml of distilled water was slowly added to the mixed oil phase with continuous mixing (Rao and McClements, 2011; Sorour *et al*. 2021). Characterizing the nano-emulsion and measuring electrical conductivity zeta potential (surface charge), and both size droplet and distribution (polydispersity indexes PDI) of micro-emulsion using Microtrac FLEX (12.0.1.0) Instrument was done.

Bird rearing and application of probiotics

Thirty-one-day-old Ross broiler chicks were obtained from Ommat Arab poultry breeder company, Giza, Egypt. They were incubated on Sacrolyte g/L water for 12 hours alternately with florfenicol 10% antibiotics ml/L water for the other 12 hours for three successive days. Chicks were reared under optimum conditions at a temperature range started at 35–37°C then decreased gradually till reached 25–23°C on the 42nd day, and relative humidity of about 50%. They received local standard ration composed of yellow corn, soybean meal, fish meal, corn oil, Glutin, Di-calcium phosphate, limestone ground, and NaCl. Starter ration for 0–19-day old chick encompassed of crude protein 23%, crude fat 4.41%, crude fiber 2.42%, representative energy (Kcal/Kg diet) 3050, Calorie/Protein ratio 132.61, D-L-methionine 0.65%, L-lysine 1.49%, calcium 1.06%, total phosphorous 0.36%. Grower ration for 20- 35 day old chick composed of crude protein 21%, crude fat 5.42%, crude fiber 2.4%, RE (Kcal/Kg diet) 3115, calorie/Protein ratio 148.33, D-L-methionine 0.55%, L-lysine 1.19%, calcium 1.06%, total phosphorous 0.35% and finishing ration for the last week contained crude protein 17%, crude fat 6.43%, crude fiber 2.35%, RE (Kcal/Kg diet) 3265, Calorie/Protein ratio 186.57, D-L-methionine 0.42%, L-lysine 1%, calcium 1.06%, total phosphorous 0.33%) and water for one week and then divided on the 8th day into two equal groups each of 15 chicks. Group A was used as a control which received water and a standard ration till the end of the experiment on the 42nd day. Group B was fed the same ration as the control group but mixed with SC probiotics at 2.5 billion CFU/Kg ration and free water till the end of the experiment. All birds were slaughtered, and thigh and breast muscles were collected. The ethical approval of this experimental design was obtained from the Institutional Animal Care and Use Committee (ARC- IACUC), Animal Health Research Institute, Agriculture Research Center, Egypt, with the number ARC, AHRI, 32, 24 before starting the experiment.

Poultry meat treatment with rosemary nano-emulsion and grouping

The collected breast and thigh muscles either from Group A or Group B were divided into two parts. The first part from each group was dipped in sterile distilled water for 24 hours before being chilled at 4±1°C for up to 9 days. While the second part was dipped in rosemary nano-emulsion 10% for 24 hours before being chilled as in part one. Accordingly, four groups of meat samples (breast and thigh muscles) were obtained, individually refrigerated, and then periodically collected at various time intervals on 0, 3rd, 7th, and 9th days of storage for microbiological and physicochemical analyses. Group I was considered as control which was not exposed to either SC probiotics or RNE. Group II was exposed to SC probiotics only, whereas Group III was exposed to RNE only, and Group IV was exposed to both SC probiotics and RNE treatments.

Poultry meat examination

Physicochemical analysis

The treated samples were analyzed for keeping quality indices by measuring pH, Total volatile basic nitrogen (TVBN) and thiobarbituric acid reactive substances (TBA-RS) according to the Egyptian Organization for Standardization and Quality control (EOS, 2006).

Microbiological examination

Samples were examined at 0, 3, 7, and 9 days of chilling at 4°C, where 25g of each sample was blended with 225 ml of 0.1% peptone water for 1-2 minutes in a sterile blender jar. Further decimal serial dilutions were prepared for testing. Determination of total colony count was done according to USDA (2011), total *Enterobacteriaceae* count (ISO, 2001), total fungal count according to FAO (1992). total *Staphylococci* count and Staphylococcus aureus count (USDA, 2011), where morphological examination of the suspected colonies was applied (Cruickshank *et al*., 1975) then biochemical identification (MacFaddin, 2000) and examination for catalase activity test, oxidase test, growth at 10% NaCl, detection of Arginine decarboxylase (ADH), bile esculent test, mannitol test, detection of hemolysis, coagulase test, thermostable nuclease test "D-Nase activity" (Lachia *et al*., 1971) and fermentation of sugars. Finally, isolation of *Salmonella* spp. (ICMSF, 1978) where smears of suspected colonies were stained with Gram stain and examined morphologically for staining characters. Presumptive *Salmonella* colonies were then subjected to initial screening tests using triple sugar iron agar (TSI), lysine iron agar (LIA), urea broth (Merck) and lysine decarboxylase. All biochemical tests were performed at 37°C for 18–24 hours including citrate utilization, indol production test, methyl red, urea hydrolysis, and Voges- Proskauer (Andrews and Hammack, 1998).

Statistical analysis

Preliminary statistical analysis was carried out using Graph Pad In-Stat software (version 3, ISS-Rome, Italy). Unless differentially specified,

groups of data were compared with one-way analysis of variant (ANOVA) followed by Tukey-Kramer (TK) Multiple Comparison post-test. The data as indicated are reported in tables and figures as mean±standard error (SEM). Values of P ≤ 0.05 were regarded as significant.

Results

SCY and RNE reduced the deterioration indicators of broiler chicken meat

The obtained results showed that pH in SCY+ RNE-treated group were 6.05±0.029, 5.9±0.026, 5.95 ±0.05 and 6.003 ±0.003, in RNE-treated group were 6.2 ±0.011, 5.89±0.018, 6.187±0.0088 and 6.26±0.011, in SCY-treated group were 5.74±0.063, 5.89±0.012, 6.177±0.0088 and 6.32±0.011 and in control group were 5.94±0.026, 5.68 ±0.017, 6.19±0.0088 and 6.34±0.0088 on 0, 3rd, 7th and 9th day respectively there is a highly significant decrease (p<0.001) in pH in SCY+ RNE-treated group, RNE-treated group and SCY-treated group in compare with control (Figure 1).

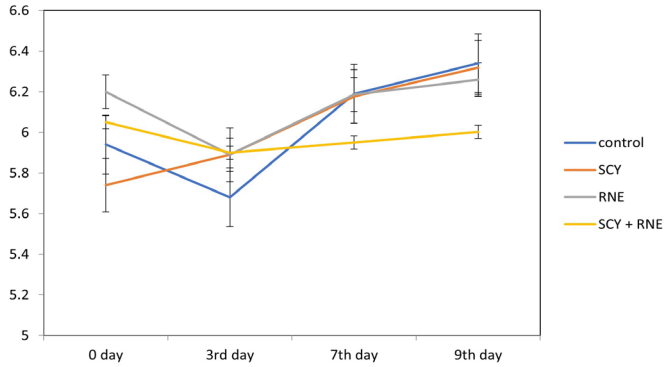

Figure 1. pH changes in broiler chicken meat exposed to *Saccharomyces cerevisiae* yeast (SCY) and rosemary nano-emulsion (RNE) 10% either individually or together at different time intervals during chilled storage at 4±1°C. Data are represented by means±standard errors of at least 3 replicates (n≥3).

Figure 2 shows the changes in TVBN of treated and untreated broiler chicken meat at different time intervals during refrigerated storage at 4±1°C. The obtained results showed that the changes in TVBN of SCY+ RNE-treated group were 8.77±0.082, 9.87±0.067, 12.62±0.35 and 18.73±0.088, in RNE-treated group were 9.67±0.234, 11.34±0.081, 13.46±0.087 and 18.8±0.04, in SCY-treated group were 8.77±0.1, 11.55±0.39, 15.25±0.15 and 18.76±0.06 and in Control group were 11.67±0.285, 12.97±0.23, 16.48±0.057 and 21±0.1 on 0, 3rd, 7th and 9th day respectively there is a highly significant decrease at (p<0.001) in the TVBN changes of SCY+ RNE-treated group, RNE-treated group and SCY-treated group in compare with control.

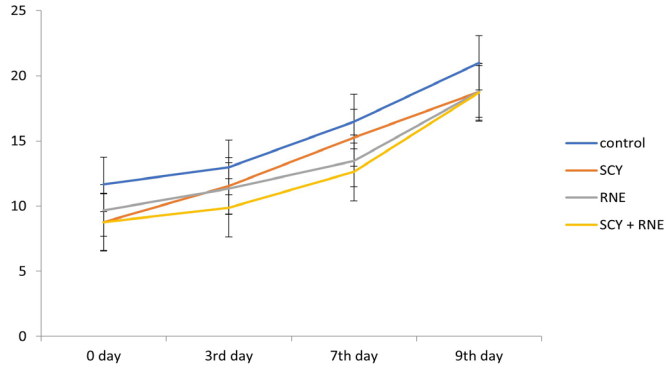

Figure 2. The changes in TVBN of broiler chicken meat exposed to *Saccharomyces cerevisiae* yeast (SCY) and rosemary nano-emulsion (RNE) 10% either individually or together at different time intervals during chilled storage at 4±1°C. Data are presented as means±standard errors of at least 3 replicates (n≥3).

Figure 3 shows the changes in TBA of treated and untreated broiler chicken meat at different time intervals during refrigerated storage at 4±1°C. The changes in TBA of SCY+ RNE-treated group were 0.114±0.025, 0.126±0.007, 0.148±0.005 and 0.1716±0.005, of RNE-treated group were 0.218±0.018, 0.277±0.0021, 0.383±0.0035 and 0.5746±0.014, of SCY-treated group were 0.187±0.013, 0.226±0.018, 0.391±0.007 and 0.4756±0.0088 and of control group were 0.241±0.014, 0.475±0.028, 0.803±0.042 and 0.892 ±0.011 at 0, 3rd, 7th and 9th day respectively. There is a highly significant decrease at (p<0.001) in the TBA changes of SCY+ RNE-treated group, RNE-treated group and SCY-treated group in compare with control.

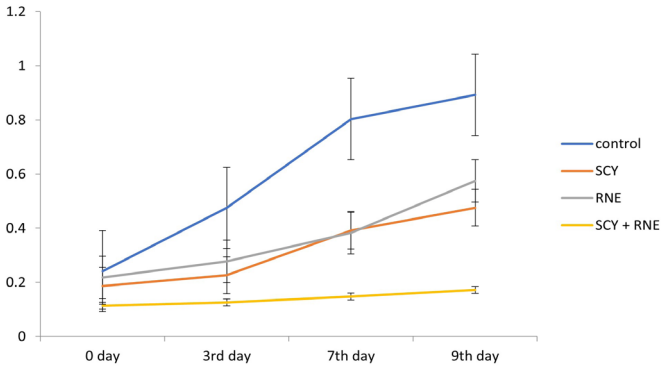

Figure 3. The changes in TBA of broiler chicken meat exposed to *Saccharomyces cerevisiae* yeast (SCY) and rosemary nano-emulsion (RNE) 10% either individually or together at different time intervals during chilled storage at 4±1°C. Data are presented as means±standard errors of at least 3 replicates (n≥3).

SCY and RNE improved the microbiological quality of broiler chicken meat

Table 1 summarizes the TCC of treated and untreated broiler chicken meat at different time intervals during refrigerated storage at 4±1°C. The obtained results showed that TCC in SCY+ RNE-treated group were $4.3x10^3$±44,$0.9x10^4$±57.74, $6.3x10^4$±120.2 and $8.3x10^4$±136.4, while in RNE-treated group were $5x10^3$±57.7, $1.2x10^4$±88.2, $7.6x10^4$±66.67 and 1.26x 10^5±120.2, and in SCY-treated group were $5.3x10^3$±16.7, $1.26×10^4$±115.5, $8.3x10^4$±66.67 and $2.3x10^5$±233.3 on 0, 3rd, 7th and 9th day, respectively. On the other hand, they were $7×10^3$±76.4, $3.6×10^4$±333.33, 1.373.5 in the control group on 0, 3rd, 7th and 9th day, respectively. There was a significant decrease (p<0.001) in TCC in SCY+ RNE-treated group, RNE-treated group, and SCY-treated group in comparison with the control group.

The total *Enterobacteriaceae* counts of treated and untreated broiler chicken meat at different time intervals during refrigerated storage at 4±1°C were shown in Table 2. The obtained results showed that Total *Enterobacteriaceae* counts in SCY+ RNE-treated group were $2.3x10^3$±16.67, $6x10^3$±57.7, $8.3x10^3$±33.33 and $5x10^4$±577.35, in RNE-treated group were $3.3x10^3$±60.1, 6.6x 10^3 ±88.2, $8.3x10^3$ ±33.33 and $6.6x10^4$ ±60.2, in SCY-treated group were $3.3x10^3$±44.1, $7.3x10^3$ ±44.1, $9.3x10^3$±72.65 and $2.1x10^4$±88.2 and in Control group were $4.3x10^3$±16.67, $7.6x10^3$±120.2, $1.3x10^4$ ±76.4 and $7.6x10^4$±88.2 on 0, 3rd, 7th and 9th day, respectively. There is a highly significant decrease (p<0.001) in Total *Enterobacteriaceae* counts in SCY+ RNE-treated group, RNE-treated group, and SCY-treated group in comparison with control.

Table 3 shows the Total fungal counts of treated and untreated broiler chicken meat at different time intervals during refrigerated storage at 4±1°C. Total fungal counts in SCY+ RNE-treated group were $2x10^3$±28.9, $4.6x10^3$±33.33, $9x10^3$±57.35 and $5.9x10^4$±440.1, in RNE-treated group were $3.3x10^3$±60.1, $6x1036x10^3$±50, $9.3x10^3$±16.67 and $6.3x10^4$±333.3, in SCY-treated group were $3.6x10^3$±44.1, $5x10^3$±57.7, $1.6x10^3$ ±33.33 and $7.3x10^4$±333.3 and in Control group were $4.3x10^3$±44.1, $7.3x10^3$±33.33, $2.6x10^4$±72.65 and 7.3x104±166.67 on 0, 3rd, 7th and 9th day respectively there is a highly significant decrease at (p<0.001) in Total fungal counts in SCY+ RNE-treated group, RNE-treated group and SCY-treated group in compare with control.

Staphylococci counts of treated and untreated broiler chicken meat at different time intervals during refrigerated storage at 4±1°C were presented in Table 4. The obtained results showed that in Total *Staphylococci* counts SCY+ RNE-treated group were $1x10^3$±5.8, $2.6x10^3$±44.1, $5.3x10^3$±16.67 and $1.3x10^3$±132.3, in RNE-treated group were $1.6x10^3$±33.3, $4x10^3$±57.7, $6.6x10^3$±44.1 and $2x10^4$±86.6, in SCY-treated group were $1.6x10^3$±44.1, $3.3x10^3$±44.1, $6x10^3$±57.7 and $1.6x10^4$±33.33 and in Control group were $2.3x10^3$±16.67, $4.3x10^3$±33.3, $8x10^3$±76.4 and $2.3x10^4$±16.67 on 0, 3rd, 7th and 9th day respectively, there is a highly significant decrease at ($p<0.001$) in Total *Staphylococci* counts in SCY+ RNE-treated group, RNE-treated group and SCY-treated group in compare with control.

Regarding the incidence of *Salmonella* and Staphylococcus aureus in broiler chicken meat exposed to *Saccharomyces cerevisiae* yeast (SCY) and/or rosemary nano-emulsion (RNE) 10% at different time intervals during chilled storage at 4±1°C, it was failed to detect them throughout the storage duration (data not shown).

Discussion

Lipid oxidation of meat is one of the most important factors affecting shelf life and one of the decisive factors in the consumer's purchase decision of a product. So, the use of natural products that have an antioxidant capacity such as SMY and RNE can overcome and delay the oxidation process. Accordingly, they prolong shelf life and improve the quality of meat.

Our results in Figures 1, 2 and 3, confirm the antioxidant effect of SCY and RNE, where there was amelioration to the deteriorative characters (pH, TVBN and TBARS) all over the 9 days of storage in in SCY+ RNE-treated, RNE-treated and SCY-treated groups. Rosemary has a potential effect to inhibit the lipid oxidation of food, by eliminating free radicals and the chain reaction of metal ions such as Fe2 is terminated,

Table 1. Total colony counts (CFU/g) of broiler chicken meat exposed to *Saccharomyces cerevisiae* yeast (SCY) and rosemary nano-emulsion (RNE) 10% either individually or together at different time intervals during chilled storage at 4±1°C.

Treatment groups	Day of chilled storage			
	0 day	3rd day	7th day	9th day
Group I (Control)	$7×10^3$±76.4^a	$3.6×10^4$±333.33^a	$1.3×10^5$±881.9^a	$6×10^5$±5773.5^a
Group II (SCY-treated)	$5.3x10^3$±16.7^c	$1.26×10^4$±115.5^b	$8.3×10^4$±66.67^c	$2.3×10^5$±233.3^c
Group III (RNE-treated)	$5×10^3$±57.7^b	$1.2×10^4$±88.2^b	$7.6×10^4$±66.67^b	$1.26×10^5$±120.2^b
Group IV (SCY+ RNE-treated)	$4.3×10^3$±44^d	$0.9×10^4$±57.74^c	$6.3×10^4$±120.2^d	$8.3×10^4$±136.4^d

Data are presented as means±standard errors of at least 3 replicates (n≥3). Different small letters superscripts ($^{a, b, c, d}$) within the same column indicate high significant differences between means at ($P < 0.001$).

Table 2. Total *Enterobacteriaceae* counts (CFU/g) of broiler chicken meat exposed to *Saccharomyces cerevisiae* yeast (SCY) and rosemary nano-emulsion (RNE) 10% either individually or together at different time intervals during chilled storage at 4±1°C.

Treatment groups	Day of chilled storage			
	0 day	3rd day	7th day	9th day
Group I (Control)	$4.3x10^3$ ±16.67^a	$7.6x10^3$±120.2^a	$1.3x10^4$±76.4^a	$7.6x10^4$±88.2^a
Group II (SCY-treated)	$3.3x10^3$±44.1^b	$7.3x10^3$±44.1^b	$9.3x10^3$±72.65^c	$2.1x10^4$±88.2^c
Group III (RNE-treated)	$3.3x10^3$ ±60.1^b	$6.6x10^3$±88.2^a	$8.3x10^3$±33.33^b	$6.6x10^4$±60.2^b
Group IV (SCY+ RNE-treated)	$2.3x10^3$±16.67^c	$6x10^3$±57.7^c	$8.3x10^3$±33.33^b	$5x10^4$ ±577.35^d

Data are presented as means±standard errors of at least 3 replicates (n≥3). Different small letters superscripts ($^{a, b, c, d}$) within the same column indicate high significant differences between means at ($P < 0.001$).

Table 3. Total fungal counts of broiler chicken meat exposed to *Saccharomyces cerevisiae* yeast (SCY) and rosemary nano-emulsion (RNE) 10% either individually or together at different time intervals during chilled storage at 4±1°C.

Treatment groups	Day of chilled storage			
	0 day	3rd day	7th day	9th day
Group I (Control)	$4.3x10^3$±44.1^a	$7.3x10^3$±33.33^a	$2.6x10^4$±72.65^a	$7.3x10^4$±166.67^a
Group II (SCY-treated)	$3.6x10^3$ ±44.1^c	$5x10^3$±57.7^c	$1.6x10^3$±33.33^c	$7.3x10^4$±333.3^a
Group III (RNE-treated)	$3.3x10^3$±60.1^b	$6x10^3$±50^b	$9.3x10^3$±16.67^b	$6.3x10^4$±333.3^b
Group IV (SCY+ RNE-treated)	$2x10^3$±28.9^d	$4.6x10^3$±33.33^d	$9x10^3$±57.35^d	$5.9x10^4$ ±440.1^c

Data are represented by means±standard errors of at least 3 replicates (n≥3). Different small letters superscripts (a, b, c, d) within the same column indicate high significant differences between means at ($P < 0.001$).

Table 4. Total *Staphylococci* count in broiler chicken meat exposed to *Saccharomyces cerevisiae* yeast (SCY) and rosemary nano-emulsion (RNE) 10% either individually or together at different time intervals during chilled storage at 4±1°C.

Treatment groups	Day of chilled storage			
	0 day	3rd day	7th day	9th day
Group I (Control)	$2.3x10^3$±16.67^a	$4.3x10^3$±33.3^a	$8x10^3$±76.4^a	$2.3x10^4$±16.67^a
Group II (SCY-treated)	$1.6x10^3$±44.1^b	$3.3x10^3$±44.1^c	$6x10^3$±57.7^c	$1.6x10^4$±33.33^c
Group III (RNE-treated)	$1.6x10^3$ ±33.3^b	$4x10^3$±57.7^b	$6.6x10^3$±44.1^b	$2x10^4$±86.6^b
Group IV (SCY+ RNE-treated)	$1x10^3$ ±5.8^c	$2.6x10^3$±44.1^c	$5.3x10^3$±16.67^d	$1.3x10^3$ ±132.3^d

Data are represented by means±standard errors of at least 3 replicates (n≥3). Different small letters superscripts (a, b, c, d) within the same column indicate significant differences between means at ($P < 0.001$).

reducing the activated oxygen molecules rate of formation (Afonso *et al*., 2013; Raškovic *et al*., 2014; Aminzare *et al*., 2019; Rashidaie *et al*., 2019).

Rosemary is a natural antioxidant widely used for food conservation (Feng *et al*., 2016). Yeast having manno-oligosaccharides or mannose which confirmed by Lyons (2002) and Oyofo *et al*. (1989). mannanoligosaccharides and β-glucans content of *Saccharomyces cerevisiae* yeast make them used as a growth promoter and immune stimulant (Celik *et al*., 2000; Çelýk *et al*., 2003). Also, it has antimicrobial properties and other health-related benefits through the maintenance of the intestinal biostructure (Wang and Gu, 2010; Zhang, L. *et al*., 2016). Also, it prevents some specific intestinal pathogens, produce various nutrients, improve the chicken intestinal metabolism (Zarei *et al*., 2018), and enhances the general performance as well as improve the local and systemic immunity (Korver, 2012; Wang *et al*., 2017).

Additionally, data in Tables 1, 2, 3 and 4 showed the beneficial effects of adding probiotics (SCY) to chicken ration (SCY), dipping chicken meat in rosemary nano-emulsion (RNE) and a combination of both treatments (SCY+RNE) on chicken meat quality and shelf life, where the great reduction in microbial counts was achieved SCY+RNE, followed by RNE and finally SCY.

The antioxidant and antimicrobial activities of rosemary extract (RE) is linked with their contents of phenolic diterpenes such as carnosic acid, carnosol, rosmanol, rosmaridiphenol and rosmariquinone, ursolic acid, and caffeic acid (Aruoma *et al*., 1992). Moreover, several research studies have reported RE as an antimicrobial food additive because of the phenolic constituents and their ferrous ion-chelating effect (Zhang, H. *et al*., 2016). Moreover, adding RE or their essential oils has an impact retarding microbial growth of both Gram-positive and Gram-negative bacteria in different meat systems (Liu *et al*., 2009; Jiang *et al*., 2011). The antimicrobial mechanism of these plant extracts and their essential oils was explained by their effect on the bacterial cell membrane integrity and permeability (Ojeda-Sana *et al*., 2013).

Probiotic supplementation improved birds' immunity, which is proved by the results in samples of the SMY-treated group revealing significantly lower microbial counts as compared to control group, such findings agree with the results reported by Sarwar et. al. (2023) and Ayanwale et. al. (2006). Moreover, Cuenca et. al. (2022) explained the mode of action of SCY as probiotics, where it has shown the capacity to cross the gastric barrier, multiplying and colonizing the intestine. When added to the diet of monogastric animals, it favors the development of the gastrointestinal microbial flora and stimulates immunity and microvilli; it inhibits the action of microbial toxins and increases an antagonistic effect against pathogenic microorganism.

Samples of the SMY+RNE group showed the lowest microbial counts due to the double effect of using SC probiotics as feed additives and RE nano-emulsion as food additives, leading to prolonging the shelf life, where TCC did not reach 10^5 CFU/g as shown in Table 1.

Regarding the results of the effect of probiotic (*Saccharomyces cerevisiae*) on microbial properties of chicken meat, a significant reduction of microbial counts was observed in the RNE-treated group (Tables 1, 2, 3, and 4), as compared to the control (untreated) group.

Additionally, probiotics have been shown to inhibit pathogenic bacteria through different mechanisms including maintaining normal intestinal microflora by competitive exclusion and antagonism, altering metabolism by increasing digestive enzyme activity and decreasing bacterial enzyme activity and ammonia production, inhibiting the action of microbial toxins and exerting an antagonistic effect against pathogenic micro-organisms, improving feed intake and digestion and finally stimulating the immune system (Kabir *et al* 2005; Condoy *et al* 2021; Castro and Rodríguez 2005).

Both total colony count (TCA) and *Enterobacteriaceae* count (EC) are used to evaluate food contamination, manufacturing system hygiene, quality control, and food safety (American Public Health Association, 1984). It is recommended that the flesh total colony count TCC should not exceed 106/g wet weight (ICMSF, 1998). Whereas the Egyptian Organization for Standardization (EOS, 2005) recommended that TCC and *Enterobacteriaceae* count should not be more than 10^5/g, and 10^3/g, respectively.

Higher TCCs were reported by Bailey *et al*. (2000) (4.62 and 6.87 $\log_{10}$/g on 0 and 7th day of chilling at 4°C, respectively) and Mahmoud *et al*. (2020) (7.6×10^5 and 4.9×10^6 CFU/g for breast and thigh samples, respectively). Moreover, higher counts of *Enterobacteriaceae* were recorded by Mahmoud *et al*. (2020) who reported 1.6×10^5CFU/g and 3.6×10^5 CFU/g for breast and thigh samples, respectively, while lower *Staphylococci* counts (6.3×10^2CFU/g in breast and 2.5×10^3 CFU/g in thigh samples) and total fungal counts (1.1×10^3 CFU/g in breast and 4.7×10^3 CFU/g in thigh samples).

Samples treated with RNE showed lower total colony counts than those reported by Elzamzamy (2014). On the other hand, Al-Hijazeen, (2018) stated higher counts (4.15, 4.15, 7.11, 7.60 and 8.10 log CFU/g at 40C on 0, 2, 4, 6, and 8, respectively). Total *Enterobacteriaceae* counts in the current study revealed higher counts than those reported by Elzamzamy (2014). On the other hand, Al-Hijazeen 2018 reported higher counts in the treated samples during chilling at 4°C.

Table 3 showed similar counts of Staphylococcus to those reported by Elzamzamy (2014) in the case of the RNE-treated group. On the contrary, total fungal counts displayed in Table 4 revealed higher values than the results reported by Elzamzamy (2014).

In the current study, *Salmonella* was not detected in treated and untreated samples, on the contrary, Bailey *et al*. (2000) determined counts of 1.54 and 1.51 $\log_{10}$/g on 0 and 7th day of chilling at 4°C. While Mahmoud *et al*. (2020) reported *Salmonella* in 9% of the examined breast and thigh samples.

Conclusion

Adding *Saccharomyces cerevisiae* as probiotic additives to poultry feeds and then dipping meat in rosemary nano-emulsion improved meat quality and prolonged the shelf life of poultry meat. Further investigation on the cytotoxicity of rosemary nano-emulsion and sensory attributes of treated poultry samples is recommended.

Conflict of interest

The authors declare that they have no conflict of interest.

References

Afonso, M.S., Mara de, O.S.A., Carvalho, E.B.T., Rivelli, D.R., Rogero, M.M., Lottenberg, A.M., Torres, R.R.P., Barros, S.B.M., Mancini, F.J., 2013. Phenolic compounds from Rosemary (*Rosmarinus officinalis* L.) attenuate oxidative stress and reduce blood cholesterol concentrations in diet-induced hypercholesterolemic rats. Nutrition & Metabolism 10, 1-9. https://doi.org/10.1186/1743-7075-10-19

Ahn, D.U., Nam, K.C., Lee, E.J., 2009. Lipid oxidation and flavor.In: M. Du and R. J. McCormick (Eds.), Applied muscle biology and meat science Boca Raton, CRC Press, Chap. 12, pp. 227-246.

Al-Hijazeen, M.,,2018. Evaluate antibacterial activity of rosemarinus officinalis linn. Extract and originum syriacum l. essential oil using raw chicken meat. Carpathian Journal of Food Science and Technology 10, 132-145.

American Public Health Association, 1984. Compendium of methods for the microbiological examination of foods, 2d ed. APHA, Washington, DC.

Amin, R.A., 2013. Screening of antibacterial activity of cinnamon, clove and rosemary essential oils against common food borne pathogens in minced beef. Benha Veterinary Medical Journal 25, 151-164.

Aminzare, M., Hashemi, M., Ansarian, E., Bimkar, M., Hassanzad, A.H., Reza, M.M., Daneshamooz, S., Raeisi, M., Jannat, B., Afshar, A., 2019. Using natural antioxidants in meat and meat products as preservatives: A review. Adv. Anim. Vet. Sci. 7, 417-426.

Andrews, W.H., Hammack. T.S., 1998. *Salmonella*. In Bacteriological Analytical Manual Revision A, Chapter 5. Gaithersburg, MD, AOAC International.

Aruoma, O. I., Halliwell, B., Aeschbach, R., Loliger, J., 1992. Antioxidant and pro-oxidant properties of active rosemary constituents: carnosol and carnosic acid. Xenobiotica 22, 257-268. https://doi.org/10.3109/00498259209046624

Ayanwale, B.A., Kpe, M., Ayanwale, V. A., 2006. The effect of supplementing *Saccharomyces cerevisiae* in the diets on egg laying and egg quality characteristics of pullets. Journal of Poultry Science 5, 759-63. https://doi.org/10.3923/ijps.2006.759.763

Bailey, J. S., Lyon, B.G., Lyon, C.E., Windham, W.R., 2000. The Microbiological Profile of Chilled and Frozen Chicken. Journal of Food Protection 63, 1228-1230. https://doi.org/10.4315/0362-028X-63.9.1228

Balentine, C.W., Crandall, P.G., O'Bryan, C.A., Duong, D.Q., Pohlman, F.W., 2006. The pre-and post-grinding application of rosemary and its effect of lipid oxidation and color during storage of ground beef. Meat Science 73, 413-421. https://doi.org/10.4315/0362-028X-63.9.1228

Castro, M., Rodríguez, F., 2005. Yeasts: probiotics and prebiotics that improve animal produc-

tion.Corpoica. Ciencia y Tecnología Agorpecuaria 6, 26-38. https://doi.org/10.21930/rcta.vol6_num1_art:33
Celik, I., Oguz, H., Demet, O., Donmez, H.H., Boydak, M., Sur, E., 2000. Efficacy of polyvinylpolypyrrolidone in reducing the immunotoxicity of aflatoxin in growing broilers. British Poultry Science 41, 430-439. https://doi.org/10.1080/713654954
Çelýk, K.., Denlý, M., Savas, T., 2003. Reduction of toxic effects of aflatoxin B1 by using baker yeast (*Saccharomyces cerevisiae*) in growing broiler chicks' diets. Revista brasileira de zootecnia 32, 615-619. https://doi.org/10.1590/S1516-35982003000300013
Chao, S.X., Canellas, E., Wrona, M., Becerril, R., Nerin, C., 2020. Comparison of two antioxidant packaging based on rosemary oleoresin and green tea extract coated on polyethylene terephthalate for extending the shelf life of minced pork meat. Food packaging and shelf life 26, 1-9. https://doi.org/10.1016/j.fpsl.2020.100588
Condoy, M. C., ChaucaGuamán, J., Sinchire, C.G., Sigüencia, H. S., 2021. *Saccharomyces cerevisiae* as an alternative to antibiotics as a growth promotor in animal feed. International Journal of Probiotics and Prebiotics. 16, pp. x-x.Cruickshank, R., Duguid, J., Marmion, B., Swain, R.H., 1975. Medical Microbiology,12th Ed., Edinburg, London and New York.
Cuenca, M., Chauca, J., García, C., Sigüencia, H., 2022. *Saccharomyces cerevisiae* as a replacement alternative to growth-promoting antibiotics in animal feed. Archivos de Zootecnia 71, 62-69. https://doi.org/10.21071/az.v71i273.5612
Duarte, F.C., Larroza, N.I., 2019. Oil nanoencapsulation: development, application, and incorporation into the food market. Nanoscale Research Letters 14, 1-13. https://doi.org/10.1186/s11671-018-2829-2
EOS, 2005. Egyptian organization for standardization and Quality control. Standard specification No1090, for chicken meat.
EOS, 2006. Methods of analysis and testing for meat and meat products. Determination of thiobarbituric acid (TBA). Egyptian organization for standardization EOS, NO 63-10 (TBA).
EOS, 2006. Methods of analysis and testing for meat and meat products. Determination of hydrogen ion concentration (pH). Egyptian organization for standardization EOS, NO 63-11 (pH).
EOS, 2006. Methods of analysis and testing for meat and meat products. Determination of total volatile nitrogen (TVB-N). Egyptian organization for standardization EOS, NO 63-9 (TVB -N).
El Bayomi, R.M., Elsobky, A.M., Khater, D.F., El-Atabany, A.I., 2021. Effect of cinnamon and wheat germ essential oils on the chemical and bacteriological quality of oriental sausage. Journal of Animal Health Production 9, 49-55. https://doi.org/10.17582/journal.jahp/2021/9.s1.49.55
Elzamzamy, F.M., 2014. Effect of Rosemary extract on microbiological, chemical and sensorial properties of chilled chicken meat. Middle East Journal of Applied Sciences 4,142-150.
EOS, 2005. Egyptian Organization for Standardization and Quality Control for poultry and chilled rabbit chicken carcasses. EOS, No. 1651,
EFSA, 2008. Scientific Opinion of the Panel on Food Additives, 541 Flavourings, Processing Aids and Materials in Contact with Food (AFC). Use of rosemary extracts as 542 a food additive. The EFSA Journal 721, 1-29. https://doi.org/10.2903/j.efsa.2008.721
Feng, L., Shi, C., Bei, Z., Li, Y., Yuan, D., Gong, Y., Han, J., 2016. Rosemary extract in combination with polylysine enhance the quality of chicken breast muscle during refrigerated storage. International Journal of Food Properties 19, 2338-2348. https://doi.org/10.1080/10942912.2015.1130053
FAO, 1992. Manual of Food Quality Control, Food and Agriculture Organization of the United Nations, Roma. Microbiological analysis part 4, pp. 131.
Ghosh, H.K., Halder, G., Samanta, G., Paul, S.K.., Pyne, S.K., 2007. Effect of dietary supplementation of organic acid and mannan oligosaccharide on the performance and gut health of Japanese quail (Coturnix coturnix japonica). Asian J. Poult. Sci. 1, 1-7. https://doi.org/10.3923/ajpsaj.2007.1.7
Hussain, A.I., Anwar, F., Chatha, S.A.S., Jabbar, A., Mahboob, S., Nigam, P.S., 2010. *Rosmarinus officinalis* essential oil: antiproliferative, antioxidant and anti-bacterial activities. Brazilian Journal of Microbiology 41, 1070-1078. https://doi.org/10.1590/S1517-83822010000400027.
ICMSF, 1998. International Commission on Microbiological Specifications for Foods. .Microorganisms in Foods. St Edmunds Bury Press, Suffolk, pp. 461-472.
ICMSF, 1978. Microorganisms ecology of food. Toronto Univ Press. Toronto, Ontario, Canada.
ISO 21528-3, 2001. International Organization for Standardization Microbiology of Food and Animal Feeding Stuffs-Horizontal method for the detection and enumeration of *Enterobacteriaceae*- Colony count technique. Geneva.
Jiang, Y., Wu, N., Fu, Y., Wang, W., Luo, M., Zhao, C.J., Liu, X., 2011. Chemical composition and antimicrobial activity of the essential oil of rosemary. Environmental Toxicology and Pharmacology 32, 63-68. https://doi.org/10.1016/j.etap.2011.03.011
Jongberg, S., Torngren, M.A., Gunvig, A., Skibsted, L.H., Lund, M.N., 2013. Effect of green tea or rosemary extract on protein oxidation in Bologna type sausages prepared from oxidatively stressed pork. Meat Science 93, 538-546. https://doi.org/10.1016/j.meatsci.2012.11.005
Kabir, S.M.L., Rahman, M.M., Rahman, M.B., 2005. Potentiation of probiotics in promoting microbiological meat quality of broilers. J. Bangladesh Soc. Agric. Sci. Technol. 2, 93-96.
Khan, S.H., Yousaf, B., Mian, A.A., Rehman, A., Farooq, M.S., 2011. Assessing the effect of administering different probiotics in drinking water supplement on broiler performance, blood biochemistry and immune response. Journal Applied Animal Research 39, 418-28. https://doi.org/10.1080/09712119.2011.623783
Korver, D.R., 2012. Implications of changing immune function through nutrition in poultry. Animal Feed Science and Technology 173, 54-64. https://doi.org/10.1016/j.anifeedsci.2011.12.019
Kumar, Y., Yadav, D.N., Ahmad, T., Narsaiah, K.., 2015. Recent trend in the use of natural antioxidants for meat and meat products. Comprehensive Reviews in Food Science and Food Safety 14, 796-812. https://doi.org/10.1111/1541-4337.12156
Lachia, R., Genigeogis, C., Hoeprich. P., 1971. Meta chromatie agar- diffusion methods for detecting Staphylococcal nuclease activity. Appl. Microbiol. 21, 585-587. https://doi.org/10.1128/am.21.4.585-587.1971
Li, P., Gatlin, D.M., 2003. Dietary brewer's yeast and the prebiotic GrobioticTM AE influence growth performance, immune responses and resistance of hybrid striped bass (Morone chrysops M. saxatilis) to Streptococcus iniae infection. Aquaculture 4, 445-456. https://doi.org/10.1016/j.aquaculture.2003.08.021
Liu, D.C., Tsau, R.T., Lin, Y.C., Jan, S.S., Tan, F.J., 2009. Effect of various levels of rosemary or Chinese mahogany on the quality of fresh chicken sausage during refrigerated storage. Food Chemistry 117, 106-113. https://doi.org/10.1016/j.foodchem.2009.03.083
Lund, M.N., Heinonen, M., Baron, C.P., Estévez, M., 2011. Protein oxidation in muscle food: a review. Molecular Nutrition & Food Research 55, 83-95. https://doi.org/10.1002/mnfr.201000453
Lyons, T.P., 2002. Navigating from niche markets to mainstream: A feed industry Kakumei. Proceedings of Alltech's 16th Anuual Asia Pacific Lecture Tour, pp. 1-16.
MacFaddin, J.F., 2000. Biochemical tests for identification medical bacteria. Warery Press Inc., Baltimore, Md.
Mahmoud, R., Ibrahim, A., Abdulsalam, S., 2020. Assessment of Microbiological Quality of Imported Broiler Chicken Carcasses Retailed for Sale in Al Beida City, Libya. Damanhour Journal of Veterinary Sciences 4, 16-19. https://doi.org/10.21608/djvs.2020.33638.1019
Maksimović, Ž., Starčević, M., Šefer, D., Janjić, J., Radovanović, A., Radulović, S., Perić, D., Marković, R., 2022. Effects of adding different dosages of *Saccharomyces cerevisiae* in diet on growth performance, carcass characteristics, intestinal morphology, and gut micro flora of broilers. Kafk as Univ Vet Fak Derg. 28, 461-468.
McClements, D.J., Rao, J., 2011. Food- grade nanoemulsions: Formulation, fabrication, properties, performance, biological fate, and potential toxicity. Critical Reviews in Food Science and Nutrition 51, 285-330.
Meneses, A.C., Sayer, C., Puton, B.M.S., Cansian, R.L., Araújo, P. P. H., Oliveira, D., 2019. Production of clove oil nanoemulsion with rapid and enhanced antimicrobial activity against grampositive and gram-negative bacteria. Journal of Food Process Engineering 42, e13209. https://doi.org/10.1111/jfpe.13209
Min, B., Ahn, D.U., 2005. Mechanism of lipid peroxidation in meat and meat products: a review. Food Science and Biotechnology 14, 152-163.
Mushtaq, A., Wani, S.M., Malik, A.R., Gull, A., Ramniwas, S., Nayik, G.A., Ercisli, S., Marc, A., Riaz Ullah, H., Bari, A., 2023. Recent insights into Nanoemulsions: Their preparation, properties and applications. Food Chemistry: X. 18, 100684. https://doi.org/10.1016/j.fochx.2023.100684
Ojeda-Sana, A.M., Van Baren, C.M., Elechosa, M.A., Juárez, M.A., Moreno, S., 2013. New insights into antibacterial and antioxidant activities of rosemary essential oils and their main components. Food Control 31, 189-195. https://doi.org/10.1016/j.foodcont.2012.09.022
Oyofo, B.A., Deloach, J.R., Corrier, D.E., Norman, J.O., Ziprin, R.L., Mollenhauser, H.H., 1989. Effects of carbohydrates on *Salmonella* typhimurium colonisation in broiler chickens. Avian Diseases 33, 531- 534. https://doi.org/10.2307/1591117
Prakash, B., Kedia, A., Mishra, P.K., Dubey, N., 2015. Plant essential oils as food preservatives to control moulds, mycotoxin contamination and oxidative deterioration of Agri- food commodities– potentials and challenges. Food Control 47, 381-391.
Parlat, S.S., Oguz, H., 2001. Biological suppression of aflatoxicosis in Japanese quail (Coturnix coturnix japonica) by dietary addition of yeast (*Saccharomyces cerevisiae*). Research in Veterinary Science 71, 207-211. https://doi.org/10.1053/rvsc.2001.0512
Patterson, J. A., Burkholder, K.M., 2003. Application of prebiotics and probiotics in poultry. production. Poultry Science 82, 627-31. https://doi.org/10.1093/ps/82.4.627
Rao, J., McClements, D.J., 2011. Formation of Flavor Oil Microemulsions, Nanoemulsions and Emulsions: Influence of Composition and Preparation Method. J of Agri. & Food Chem. 59, 5026-5035. https://doi.org/10.1021/jf200094m
Rashidaie, A.S.S., Ariaii, P., Charmchian, L.M., 2019. Effects of encapsulated rosemary extract on oxidative and microbiological stability of beef meat during refrigerated storage. Food Science & Nutrition 7, 3969-3978. https://doi.org/10.1002/fsn3.1258
Raškovic, A., Milanovic, I., Pavlovic, N., Cebovic, T., Vukmirovic, S., Mikov, M., 2014. Antioxidant activity of rosemary (*Rosmarinus officinalis* L.) essential oil and its hepatoprotective potential. BMC Complementary and Alternative Medicine. 14, 1-9. https://doi.org/10.1186/1472-6882-14-225
Sarwar, F., Akhtar, R., Akram, Q., Rizwan, H.M., Naeem, M.A., Azad, A., Rahman, M.S., Saleem, M. M., Anwar, M.N., Zahid, B., Tariq, M., Habib, Z., 2023. Effects of *Saccharomyces cerevisiae* Supplemented Diet on Production Performance, Egg Quality and Humoral Immunity in Black Australorp and Fayoumi Layers. Brazilian Journal of Poultry Science 25, 01-8.
Serpen, A., Gokmen, V., Fogliano, V., 2012. Total antioxidant capacities of raw and cooked meats. Meat Science 90, 60-65. https://doi.org/10.1016/j.meatsci.2011.05.027
Sorour, H.K., Hosny, R.A., Elmasry, D.M.A., 2021. Effect of peppermint oil and its microemulsion on necrotic enteritis in broiler chickens. Veterinary World 14, 483-491. https://doi.org/10.14202/vetworld.2021.483-491
Tastan, Ö., Ferrari, G., Baysal, T., Donsì, F., 2016. Understanding the effect of formulation on functionality of modified chitosan films containing carvacrol nanoemulsions. Food Hydrocolloids 61, 756– 771.
USDA, 2011.Quantitative Analysis of Bacteria in Foods as Sanitary Indicators January 2011.
van Leeuwen, P., Verdonk, J.M.A.J., Kwakernaak, C., 2005. Effects of fructooligosaccharide inclusion in diets on performance of broiler chickens. Confidential Report 5, I00650.
Velasco, V., Williams, P., 2011. Improving meat quality through natural antioxidants. Chilean Journal of Agricultural Research 71, 313-322. https://doi.org/10.4067/S0718-58392011000200017
Wang, Y., Sun, J., Zhong, H., Li, N., Xu, H., Zhu, Q., Liu. Y., 2017. Effect of probiotics on the meat flavor and gut microbiota of chicken. Scientific Reports 7, 1-13. https://doi.org/10.1038/s41598-017-06677-z
Wang, Y., Gu, Q., 2010. Effect of probiotic on growth performance and digestive enzyme activity of Arbor Acres broilers. Research in Veterinary Science 89, 163-167. https://doi.org/10.1016/j.rvsc.2010.03.009
Xiong, Y.L., 2000. Protein oxidation and implications for muscle food quality., Antioxidants in muscle foods, New York, Wiley. pp. 85-111.
Yu, L., Scanlin, L., Wilson, J., Schmidt, G., 2002. Rosemary extracts as Inhibitors of lipid oxidation and color change in cooked turkey products during refrigerated storage. Journal of Food Science 67, 582-585. https://doi.org/10.1111/j.1365-2621.2002.tb10642.x
Zarei, A., Lavvaf, A., Motamedi Motlagh, M., 2018. Effects of probiotic and whey powder supplementation on growth performance, microflora population, and ileum morphology in broilers. Journal of Applied Animal Research 46, 840-844. https://doi.org/10.1080/09712119.2017.1410482
Zhang, H., Wu, J., Guo, X., 2016. Effect of antimicrobial and antioxidant activities of spice extract on raw chicken meat quality. Food Science and Human Wellness 5, 39-48. https://doi.org/10.1016/j.fshw.2015.11.003
Zhang, L., Zhang, L., Zhan, X.A., Zeng, X., Zhou, L., Cao, G., Chen, A.G., Yang, C., 2016. Effects of dietary supplementation of probiotic, Clostridium butyricum, on growth performance, immune response, intestinal barrier function, and digestive enzyme activity in broiler chickens challenged with Escherichia coli K88. Journal of Animal Science and Biotechnology 7, 1-9. https://doi.org/10.1186/s40104-016-0061-4