Effect of Nano-Curcumin on Staphylococcus aureus, Escherichia coli, and Bacillus cereus inoculated in chicken kofta

Rasha Elsabagh (Rasha.alsbagh@fvtm.bu.edu.eg)
Benha University
Elsayed M. Abd-Elaaty
Animal Health Research Institute
Wael K. Elfeil
Suez Canal University
Mohamed S. Diab
New Valley University

Research Article

Keywords: antimicrobials, chicken kofta, nano-Curcumin, nanotechnology, meat safety

Posted Date: April 19th, 2022

DOI: https://doi.org/10.21203/rs.3.rs-1549939/v1

Abstract

Food safety is the major issue in the food industry. Curcumin is a natural antimicrobial that has nutritional and health benefits. Nanotechnology can improve its bioavailability and application in food. Thus, this study was designed to evaluate the antimicrobial effect of nano-curcumin on some food poisoning bacteria, especially those of zoonotic importance, such as *Escherichia coli*, *Staphylococcus aureus*, and *Bacillus cereus*, *in vitro* and inoculated in chilled chicken kofta. Nano-curcumin showed antimicrobial effects on the microbes under study *in vitro*, which were confirmed using transmission electron microscopy (TEM).

The microbiological quality and physicochemical and sensory attributes of chilled chicken kofta was evaluated at 4°C for 27 days. Nano-curcumin reduced the count of *E. coli* from 5.73 to 1.09 log cfu/g, the count of *S. aureus* from 5.73 to 1.02 log cfu/g, and the count of *B. cereus* count from 5.87 to 3.13 log cfu/g. Nano-curcumin increased PH, TBA and TVB-N values over the storage period. Moreover, it enhanced the sensory attributes and shelf life of chicken kofta until the 27^{th} day of storage. Nano-curcumin is considered a novel antimicrobial and antioxidant agent for meat products.

Introduction

Food safety is the major issues in food industry, as it has severe and life-long implications to public health if food contaminated with pathogenic microorganisms is consumed [1]. Although, Chicken meat is one of the most preferable foods as it is a good source of protein and cheaper than red meat, it spoils rabidly as it contains high protein and water contents, which encourages the growth of spoilage and pathogenic microorganisms within a short period of time [2]. This food contamination caused mainly by zoonotic bacteria transmits from animals to humans and impact human health; the most common foodborne zoonotic bacterial pathogens are *Escherichia coli* (*E. coli*), and Staph-toxin [3]. One of the most important food-borne pathogens is *E. coli*, as it considers the main cause of outbreaks every year [1]; it causes Hemolytic– Uremic syndrome and intestinal infections. Another food poisoning bacteria is *Staphylococcal aureus* (*S. aureus*) that has rapid onset, violent vomiting and nausea with or without diarrhea [4]. *S. aureus* produces staphylococcal enterotoxin (SE) that is responsible for most of staphylococcal food poisoning [5]. Also *Bacillus cereus* (*B. cereus*) which is an opportunistic pathogen can cause food poisoning as a result of consuming foods containing bacteria or toxins [6].

Meat products with functional ingredients considered a demand in meat industry to prevent the risk of diseases and to enhance health conditions [7]. Great efforts are being made in the food industry for improving hygiene and increasing the shelf life of meat products through preventing the growth and multiplication of food-borne pathogens [8]. Also the most common problem that faced meat industry is the resistance of foodborne pathogens to multiple chemical antimicrobials. So, modern technique must be used to solve this problem through using natural plants as curcumin which is considered as herbal medicine and nutritional supplement that used as spice and coloring agent in food.

Nanotechnology can be applied throughout different aspects of the food chain processing to improve food safety and quality control and increase food shelf life [8]. Nano-particles and Nano-emulsion at a diameter 10

to 200 nm are powerful for lipophilic compounds to enhance flavor, antioxidant, and antimicrobial properties [9].

The main bioactive compound of turmeric is Curcumin. It has antimicrobial effects various species of Gram-negative and Gram-positive bacteria [10]. Moreover, it has many health benefits such as anti-diabetic, anti-inflammatory, anti-cancer, and antioxidant activities.

However, curcumin has many limitations as low water solubility, degradation during processing, low bioavailability [11]. Nanotechnology can improve its application by increases the solubility, dispersion and bioavailability of curcumin [12]. Moreover, it has antimicrobial activity against different types of foodborne pathogens. It enhances the microbiological quality, physical, chemical composition and sensory properties of chilled chicken fillets for 12 days [2].

The aim of this work was to (1) Evaluate the antibacterial activity of Nano-curcumin against certain pathogenic microorganism (*E. coli*, *S. aureus* and *B. cereus*) (2) Assess antioxidant activity of Nano-curcumin (3) Enhance sensory attributes and shelf life of chicken Kofta stored at 4^{o}C using Nano-curcumin.

Material And Methods

Bacterial strains:

Referenced pathogenic bacterial strains*: E. coli* (lot no: 020090, Des: NCTC: 12241 and ATCC: 25922)*, S. aureus* (lot no: 460074, Des: NCTC: 10788 and ATCC: 6538)*,* and *B. cereus* (lot no: 02900402, Des: NCTC: 10400 and ATCC: 6633) were obtained from Media Unit, Food Hygiene Department, Animal Health Research Institute, Dokki, Giza, Egypt.

The pathogenic strains were prepared and adjusted to obtain a population of 6 $\log_{10}$ CFU/mL.

Preparation of nano-curcumin (NC)

Curcumin powder from *Curcuma longa* (turmeric) (diferuloylmethane, (E,E)-1,7-bis(4-Hydroxy-3-methoxyphenyl)-1,6-heptadiene-3,5-dione, natural yellow 3) was purchased from Sigma-Aldrich (CAS 458-37-7).

The nanoparticles were produced at the National Research Center, Physics Division, according to A Khataee, S Fathinia and M Fathinia [13] with some modifications. Briefly, powder was ground using a Moulinex grinder (Model MC300, France) and then crushed using a high-energy planetary ball-mill (Model PM 2400, Iran) at 320 rpm/2 h rotation speed to prepare NC. The ball-milling process was applied under cold atmospheric conditions (25°C and 1 atm.). Finally, the homogenous NC was measured using transmission electron microscopy (TEM).

All processes used for producing NC were controlled to keep its antioxidant and antibacterial properties.

Assessment of *in vitro* antimicrobial activity of NC against different food poisoning bacteria

The disk diffusion method was used according to N Padmavathy and R Vijayaraghavan [14] to assess the inhibitory range of NC at different concentrations (i.e., 2, 5, and 10 ppm) against each inoculated pathogenic bacterium: *E. coli*, *S. aureus*, and *B. cereus* at a concentration of 10^6 CFU/mL.

Evaluation of the antimicrobial property of NC using TEM

The morphology and size of NC and its biocide effect on *E. coli*, *S. aureus*, and *B. cereus* were analyzed using TEM with negative staining of phosphor tungstic acid (PTA 1%). Nano-emulsions were diluted according to K Kaur, R Kumar and S Mehta [15]. The nanoemulsion samples were left for 3 h to dry. The grid was analyzed using TEM (JEOL JEM 1400, USA) with a working voltage of 200 kV at the TEM Unit of Cairo University Research Park, Egypt.

Assessment of the antimicrobial effect of NC on food poisoning bacteria inoculated into chicken kofta

- **Preparation of chicken kofta**

Chicken kofta was prepared according to the method described by [16]. Chicken kofta prepared from minced chicken meat (72%) and refined wheat flour (7%), and the quantities of oat flour (8%), casein (2.5%), and hydrogenated fat (7.5%) were optimized.

Before the experiment, the meat was surface treated with ultraviolet light (UV) for 15 min for each side to minimize background micro-flora according to MK Morsy, R Elsabagh and V Trinetta [17].

- **Challenge study**

The prepared mixture was divided into six groups and then inoculated with the prepared cultured bacteria adjusted at 10^6 bacteria as follows: 1st group: *E. coli*-inoculated samples (EC); 2nd group: *E. coli*-inoculated kofta treated with NC (10 ppm) (NCEC); 3rd group: *S. aureus*-inoculated samples (SA); 4th group: *S. aureus*-inoculated samples treated with NC (10 ppm) (NCSA); 5th group: *B. cereus*-inoculated samples (BS); and 6th group: *B. cereus*-inoculated samples treated with NC (10 ppm) (NCBS).

After inoculation, the samples were maintained at room temperature for 15 min to allow for cell attachment and stuffed into a sterile polyethylene casing. The samples were kept at 4°C ± 1°C for 27 days, and on days 0, 3, 6, 9, 12, 15, 18, 21, 24, and 27, the remaining microbial populations were analyzed. This experiment was repeated in triplicates to obtain the mean values for statistical analysis (n = 3).

- **Microbiological assay**

The samples were aseptically opened, and approximately 10 g from each sample was transferred into 90-mL buffered peptone water 0.1% (BPW; Biolife) and then stomached (model G-560E, Bohemia) for approximately 1 min. Ten-fold serial dilutions were prepared, and then, 1 mL was spread plated over EMB (Biolife) according to ISO21150 [18] for *E. coli,* paired parker (LO, Biolife) according to ISO6888-1 [19] for *S. aureus*, and *B. cereus* Agar Base-MYP (BC-MYP, Biolife) with polymyxin B sulfate supplement (Code 4240001) and egg yolk emulsion (Code 42111601) following [20] for *B. cereus*. The obtained colonies were counted after 24 h from incubation at 37°C and expressed as log10 CFU/gm.

Physicochemical evaluation

Physicochemical evaluations were applied according to AOAC [21]**.** pH values were monitored using a digital pH-meter (model P107, Consort, Belgium), total volatile base nitrogen TVB-N (N/100 g of sample). Meanwhile, TBARS (MDA kg^{-1}) was evaluated using spectrophotometry (CE 599 Universal, USA).

Sensory evaluation

Sensory assessment for chicken kofta was evaluated under controlled conditions of temperature and humidity by seven well-trained panelists working at the Food Hygiene and Control Department of the Animal Health Research Institute. The criteria used as the basis of the descriptive organoleptic assessment (i.e., color, odor, and texture) using the triangle test and hedonic rating system. The scale points were used in the evaluation as follows: (9: excellent, 8: very very good, 7: very good, 6: good, 5: medium, 4: fair, 3: poor, 2: very poor, and 1: very very poor) [22].

Statistical analysis:

Data on bacteriological and physicochemical properties and sensory attributes were tested for normality and homogeneity. The values were expressed as means ± standard errors of the mean.

Concerning the results of the effect of NC on food poisoning bacteria, the results were statistically analyzed using Student's t-test according to R Steele and J Torrey [23].

To know whether the "P" value is significant or not, the calculated "P" value was compared with the tabulated "P" value at the level of degree of freedom at $P \leq 0.05$ from the tables.

Results And Discussion

Foodborne pathogens are considered a major threat to international public health safety [24]. Natural antimicrobials along with nanotechnology are considered a novel tool for controlling these foodborne pathogens in the meat industry [17]. The Scientific Committee for Food of the European Community approved curcumin (number E100) in the list of additives, as it has an antioxidant, a flavoring agent, and natural colorant [25]**,** with broad antimicrobial effects in various foods.

Table 1. Assessment of the antimicrobial effect of nano-curcumin on *E. coli, S. aureus, and B. cereus* using the disk diffusion method (*in vitro*).

Pathogenic bacteria	Nano-curcumin (NC)		
	2 ppm	5 ppm	10 ppm
E. coli **(EC)**	4 ± 0.13	12 ± 0.10	15± 0.14
S. aureus **(SA)**	5± 0.11	13 ± 0.10	18 ±0.15
B. cereus **(BS)**	ND*	10 ± 0.23	13 ±0.11

ND*: Not detected

The results in Table 1 showed the antibacterial effect of different concentrations of NC (2, 5, and 10 ppm) on Gram-negative bacteria (*E. coli*), Gram-positive bacteria (*S. aureus*), and spore-forming bacteria (*B. cereus*); the diameter of the inhibition zone varied depending on NC concentration. NC at 10 ppm showed the broadest zone of inhibitions with diameters of 15, 18, and 13 mm for *E. coli*, *S. aureus*, and *B. cereus*, respectively. Similar results were reported regarding the inhibitory effects of NC on the growth of *S. aureus in vitro* [26]. RK Basniwal, HS Buttar, V Jain and N Jain [27] have proven that NC is more effective against Gram-positive than against Gram-negative bacteria. Furthermore, RS Pandit, SC Gaikwad, GA Agarkar, AK Gade and M Rai [28] have reported that NC exhibits *in vitro* antibacterial effects on *E. coli* and *S. aureus* with diameters of 12 and 15 mm, respectively.

TEM was used to evaluate the size of NC and its morphology. Moreover, TEM was used to evaluate the antimicrobial effects of NC on the structure and morphology of the pathogenic bacteria under study.

Figure 1 shows the normal structure of NC; nanoparticles were highly homogeneous and uniformly distributed with a spherical appearance and a nano-size of 90:120 nm. [2] reported that NC has a spherical shape.

TEM illustrated the antimicrobial effects of NC on the morphological structure of the pathogenic bacteria under study.

Morphological assessment showed that *E. coli* had a rod shape with intact cell walls. This normal morphology of *E. coli* was altered when the bacteria was treated with NC. NC-treated *E. coli* cells showed pores in the bacterial cell wall, and an electromagnetic field was observed between NC and *E. coli*.

NC had the same effects on *S. aureus*, as it altered the normal structure of the bacteria. Many pores were observed in the bacterial cell wall, and NC arranged around and inside the bacterial cell. *S. aureus* bacterial cell wall became corrugated and denatured. Meanwhile, *S. aureus* had a normal morphology with intact cell wall and homogenous cell structure.

The normal and organized cell wall morphology of *B. cereus* was altered following NC treatment. It showed pores in the bacterial cell wall, and curcumin nanoparticles were entrapped in and around its cell wall.

Table 2. Antimicrobial effect of nano-curcumin on the inoculated food poisoning bacteria in chicken kofta.

Groups	Zero day	3rd day	6th day	9th day	12th day	15th day	18th day	21st day	24th day	27th day
EC	5.90 ± 0.03	6.41 ± 0.05	6.92 ± 0.02	7.43 ± 0.03	7.90 ± 0.05	8.44 ± 0.07	8.76 ± 0.03	8.85 ± 0.03	9.31 ± 0.03	9.42 ± 0.10
NCEC	5.73 ± 0.03*	4.43 ± 0.06***	4.25 ± 0.03***	3.84 ± 0.03***	3.19 ± 0.04***	2.82 ± 0.04***	2.25 ± 0.06***	1.90 ± 0.01***	1.46 ± 0.06***	1.09 ± 0.01***
SA	5.97 ± 0.01	6.00 ± 0.30	6.84 ± 0.05	7.32 ± 0.03	7.92 ± 0.04	8.47 ± 0.03	8.91 ± 0.04	8.94 ± 0.03	9.51 ± 0.03	9.86 ± 0.03
NCSA	5.73 ± 0.08*	4.38 ± 0.03**	4.63 ± 0.27**	3.93 ± 0.03***	3.47 ± 0.03***	2.73 ± 0.03***	2.25 ± 0.05***	1.34 ± 0.03***	1.16 ± 0.03***	1.02 ± 0.01***
BC	5.96 ± 0.01	6.93 ± 0.03	7.72 ± 0.06	7.95 ± 0.02	8.23 ± 0.02	8.42 ± 0.03	8.84 ± 0.03	9.46 ± 0.03	9.91 ± 0.05	9.97 ± 0.01
NCBC	5.87 ± 0.03*	5.78 ± 0.03***	5.64 ± 0.01***	5.32 ± 0.07***	4.81 ± 0.01***	4.60 ± 0.00***	4.24 ± 0.07***	3.66 ± 0.02***	3.33 ± 0.02***	3.13 ± 0.02***

Data are presented as means ± standard error).

(* represents statistical significance at $p < 0.05$. ** represents statistical significance at $p < 0.01$. *** represents statistical significance at $p < 0.001$).

The antimicrobial effects of NC on *E. coli*, *S. aureus*, and *B. cereus* experimentally inoculated into chicken kofta stored at 4°C for 27days are shown in Table 2.

The count of *E. coli* was gradually increased in the untreated group during the storage period (27 days) from 5.90 to 9.42 log cfu/g. A significant difference in *E. coli* count was observed between the untreated and NC-treated samples. NC decreased the count of *E.coli* from 5.73 to 1.09 log cfu/g at the end of the experiments. NC has a great antibacterial effect on *E. coli* [28]**.** *E. coli* is a foodborne pathogen responsible for sepsis, urinary tract infections, and neonatal meningitis. Moreover, [29] proved the antibacterial effect of NC on *E. coli*.

Concerning the antimicrobial effect of NC on *S. aureus*-inoculated into chicken kofta during the storage period at 4°C, the count of *S. aureus* increased in the control sample during the storage period and reached 9.86log cfu/g after 27 days. A significant difference in the count of *S. aureus* was observed between the untreated and NC-treated groups as it decreased from 5.73 to 1.02 log cfu/g. Similar results were reported by [28] and [30] who have proven the antibacterial effect of NC on *S. aureus*.

The effect of NC on the count of *B. cereus*-inoculated into chicken kofta during the storage period at 4°C is shown in Table 2. The count of *B. cereus* increased in the control sample during the storage period (27 days)

from 5.96 to 9.97 log cfu/g, which was significantly different from that in NC-treated samples as it decreased from 5.87 to 3.13 log cfu/g. This confirmed the antimicrobial effect of NC on Gram-positive bacteria, such as *B. cereus*, and this finding agrees with those of [27] and [10].

Antioxidant role of NC and its effect on physicochemical parameters and shelf life time of chicken kofta

pH

As shown in Figure 2, there were significant differences ($P \leq 0.05$) between the treated and untreated samples during cold storage at 4°C. The pH values were acceptable at zero day and then increased, which became unacceptable at the 12^{th} day of storage, whereas the pH values in the NC-treated group became unacceptable at the 27^{th} day of storage. The pH values increased due to the bases of alkaline volatile (e.g., ammonia and tri-methylamine) formed by microbial enzymes **[31].** pH is used as an indicator of freshness to evaluate meat quality [32]. Bacterial growth during cold storage of meat increases pH values [6]. This increase in pH leads to the spoilage of untreated chicken kofta in the 12^{th} day of cold storage. Meanwhile, those treated with NC showed a delay in the rise of pH values, indicating the antibacterial effect of NC.

TVB-N

The total volatile basic-nitrogen (TVB-N) is an essential indicator of changes in the quality of meat. As shown in Figure 3, the TVB-N values in the control group differed significantly from those in the NC-treated group ($p \leq 0.05$). The maximum permissible limit for TVB-N according to [33] is 20 mg/100mg. Untreated samples spoiled on the 9^{th} day of storage, whereas NC-treated samples spoiled on the 20^{th} of storage for *E. coli*-inoculated samples and 24^{th} day of storage for *S. aureus*- and *B. cereus*-inoculated samples. TVB-N is a toxic compound produced by enzymatic degradation of bacteria, and its content gradually accumulates in spoiled meat [34]. Moreover, [35] proved that TVB-N content is the percentage of alkaline substances, such as ammonia, which are produced due to the decomposition of proteins in meat, so TVB-N content is an important indicator of meat freshness. It was found that TVB-N increased in untreated samples throughout the storage period as bacterial count increased, whereas the antibacterial effect of NC decreased the formation of toxic and volatile compounds, resulting in spoilage delays.

TBA

Fatty acid oxidation forms malonaldehyde that can be demonstrated as TBA values. According to A Abdel-Hamied, A Nassar and N El-Badry [36], these aldehydes are characterized by a rancid flavor, which speeds the rate of lipid oxidation. The maximal permissible limit was 0.9 mg malonaldhyde/kg [33]. The data presented in Figure 4 showed that the TBA values (mg malonaldhyde/kg) of untreated samples differed significantly ($P \leq 0.05$) from those of NC-treated samples. From the data, it was observed that the TBA values of all groups ranged between 0.08 and 0.11 mg malonaldhyde/kg on zero day. The highest TBA values were observed in *E. coli*-inoculated samples on the 9^{th} day of storage. Meanwhile, the lowest TBA values were observed in NC-treated *S. aureus*-inoculated samples. There are many scientific proofs on the ability of curcumin on living cells to trap free radicals, such as reactive oxygen and nitrogen species, through several

means, thus manifesting its antioxidant property [11]. NC has anti-lipid peroxidation effects, which prevent the formation of malonaldehyde [37].

Effects of NC on the sensory attributes of chilled chicken kofta.

The acceptability (i.e., odor, color, and texture) of chicken kofta during storage at 4°C is shown in Figure 5. The results showed significant differences ($p \leq 0.05$) between treated and control samples as control samples spoiled on the 9th day of storage and continue to deteriorate throughout the storage period. The data revealed that the group inoculated with *E. coli* and *S. aureus* spoiled on the 12th day of cold storage, while samples inoculated with *B. cereus* spoiled on 15th day of cold storage. The shelf life of chicken kofta extended till 21th day of cold storage in *E. coli*-inoculated group treated with NC. While, group inoculated with *S. aureus* treated with NC spoiled at 21th day of cold storage. While group inoculated with *B. cereus* treated with NC spoiled on the 24th day of cold storage. Lipid oxidation and protein degradation lead to changes in color, odor, and texture during storage **[38]**. This improvement occurred due to the natural antimicrobial and antioxidant activities of curcumin [39]. These activities of curcumin increase due to its nanostate that provides continuous release and high solubility and stability, which subsequently enhance its bioactivity, functionality, and quality and the sensory attributes of food [2].

Conclusions

Nanotechnology is one of the most hopeful technologies for improving food safety and quality. NC showed a broad-spectrum antimicrobial effect on zoonotic foodborne bacteria, such as *E. coli*, *S. aureus*, and *B. cereus*, in chicken meat products, which was proved using TEM. Moreover, NC showed an antioxidant effect that improved sensory attributes and shelf life time and delayed the microbial deterioration of chilled chicken kofta stored at 4°C to reach the 27th days of storage. NC is considered a novel antimicrobial, antioxidant, and natural additive in meat products.

Declarations

Author contribution

RE, EA and MSD: Conceptualization. RE, EA, MSD and WKE: Methodology. EA, WKE and RE: Validation. RE, MSD and WKE: Formal analysis. RE, EA and MSD: Investigation. RE, EA and MSD: Resources. RE, EA and WKE: Data curation.RE, EA, MSD and WKE: Writing – original draft preparation.MSD, RE and WKE Writing – review and editing. RE and EA: Visualization. RE, MSD and WKE: Supervision. All authors reviewed the manuscript.

Consent to Participate

All authors interpreted the data and approved the final version.

Ethical approval

The present study did not involve either humans or animals as an experimental setup. The experiment was conducted at Animal Health Research Institute, Egypt.

Data availability

All the data used and/or analyzed during the current study are available from the corresponding author on reasonable request

Declaration of competing interest

The authors declare that there are no conflicts of interest.

Funding:

There is no fund project for this manuscript

Acknowledgments

The authors express their sincere gratitude to DR\ Mohebat A. Abd El-Aziz M.A, Food Control, Animal Health Research Institute, Agriculture research center, for providing some materials and help in statistical analysis.

References

1. FDA: **Outbreaks of Foodborne Illness**. In: *https://wwwfdagov/food/recalls-outbreaksemergencies/outbreaks-foodborne-illness.* 2020.
2. Abdou ES, Galhoum GF, Mohamed EN: **Curcumin loaded nanoemulsions/pectin coatings for refrigerated chicken fillets**. *Food Hydrocolloids* 2018, **83**:445-453.
3. EFSA: **The European union one health 2018 zoonoses report**. *EFSA J* 2019, **17**:5926-6202.
4. Argudin MA, Mendoza MC, Rodicio MR: **Food poisoning and Staphylococcus aureus enterotoxins**. *Toxins (Basel)* 2010, **2**(7):1751-1773.
5. Book BB: **Foodborne Pathogenic Microorganisms and Natural Toxins Handbook**. *Center for Food Safety and Applied Nutrition (CFSAN)< http://vm cfsan fda gov/~ mow/intro htm* 2006.
6. Abd El-Aziz MA, Ibrahim HM, EL-Roos NA, Anis B, Elsabagh R: **NANO TECHNOLOGICAL ENHANCEMENT OF MEAT BALLS QUALITY**. *Proceedings on Engineering* 2020, **2**(3):323-332.
7. Sirini N, Roldán A, Lucas-González R, Fernández-López J, Viuda-Martos M, Pérez-Álvarez J, Frizzo LS, Rosmini MR: **Effect of chestnut flour and probiotic microorganism on the functionality of dry-cured meat sausages**. *LWT* 2020, **134**:110197.
8. Baltić MŽ, Bošković M, Ivanović J, Dokmanović M, Janjić J, Lončina J, Baltić T: **Nanotechnology and its potential applications in meat industry**. *Tehnologija mesa* 2013, **54**(2):168-175.
9. Rao J, McClements DJ: **Food-grade microemulsions, nanoemulsions and emulsions: fabrication from sucrose monopalmitate & lemon oil**. *Food hydrocolloids* 2011, **25**(6):1413-1423.
10. da Silva AC, de Freitas Santos PD, do Prado Silva JT, Leimann FV, Bracht L, Goncalves OH: **Impact of curcumin nanoformulation on its antimicrobial activity**. *Trends in Food Science & Technology* 2018,

72:74-82.

11. Rafiee Z, Nejatian M, Daeihamed M, Jafari SM: **Application of different nanocarriers for encapsulation of curcumin**. *Critical reviews in food science and nutrition* 2019, **59**(21):3468-3497.
12. Abbas S, Karangwa E, Bashari M, Hayat K, Hong X, Sharif HR, Zhang X: **Fabrication of polymeric nanocapsules from curcumin-loaded nanoemulsion templates by self-assembly**. *Ultrasonics Sonochemistry* 2015, **23**:81-92.
13. Khataee A, Fathinia S, Fathinia M: **Production of pyrite nanoparticles using high energy planetary ball milling for sonocatalytic degradation of sulfasalazine**. *Ultrasonics sonochemistry* 2017, **34**:904-915.
14. Padmavathy N, Vijayaraghavan R: **Enhanced bioactivity of ZnO nanoparticles—an antimicrobial study**. *Science and technology of advanced materials* 2008.
15. Kaur K, Kumar R, Mehta S: **Formulation of saponin stabilized nanoemulsion by ultrasonic method and its role to protect the degradation of quercitin from UV light**. *Ultrasonics sonochemistry* 2016, **31**:29-38.
16. Prasad B, Rashmi M, Yashoda K, Modi V: **Effect of casein and oat flour on physicochemical and oxidative processes of cooked chicken kofta**. *Journal of Food Processing and Preservation* 2011, **35**(3):359-368.
17. Morsy MK, Elsabagh R, Trinetta V: **Evaluation of novel synergistic antimicrobial activity of nisin, lysozyme, EDTA nanoparticles, and/or ZnO nanoparticles to control foodborne pathogens on minced beef**. *Food control* 2018, **92**:249-254.
18. ISO21150: **Cosmetics. Microbiology - Detection of Escherichia coli.** 2006.
19. ISO6888-1: **Horizontal methods for the enumeration of coagulase-positive Staphylococci (Staphylococcus aureus and other species), Technique using Baird-Parker medium** 2003.
20. ISO7932: **Horizontal method for the enumeration of presumptive Bacillus cereus–colony-count technique at 30°C**. 2004.
21. AOAC: **Official methods of analysis of the association of official analytical chemists**. In. Edited by Horwitz, 17th edn: Washington, DC; 2005.
22. STANDARD B, ISO B: **Sensory analysis—Methodology—General guidance for establishing a sensory profile**. 2003.
23. Steele R, Torrey J: **Principles and procedures of statistics: a biometrical approach. New York: McGraw & Hill**. *Inc p* 1980, **593**.
24. Manea L, Buruleanu L, Rustad T, Manea I, Barascu E: **Overview on the microbiological quality of some meat products with impact on the food safety and health of people**. In: *2017 E-Health and Bioengineering Conference (EHB): 2017*. IEEE: 105-108.
25. EFSA A: **Panel (Panel on Food Additives and Nutrient Sources added to Food), 2010. Scientific Opinion on the re-evaluation of curcumin (E 100) as a food additive. EFSA Journal 2010; 8 (9): 1679. 46 pp**. In.; 2010.
26. Krausz AE, Adler BL, Cabral V, Navati M, Doerner J, Charafeddine RA, Chandra D, Liang H, Gunther L, Clendaniel A: **Curcumin-encapsulated nanoparticles as innovative antimicrobial and wound healing agent**. *Nanomedicine: Nanotechnology, Biology and Medicine* 2015, **11**(1):195-206.

27. Basniwal RK, Buttar HS, Jain V, Jain N: **Curcumin nanoparticles: preparation, characterization, and antimicrobial study**. *Journal of agricultural and food chemistry* 2011, **59**(5):2056-2061.
28. Pandit RS, Gaikwad SC, Agarkar GA, Gade AK, Rai M: **Curcumin nanoparticles: physico-chemical fabrication and its in vitro efficacy against human pathogens**. *3 Biotech* 2015, **5**(6):991-997.
29. Roy S, Rhim J-W: **Preparation of carbohydrate-based functional composite films incorporated with curcumin**. *Food Hydrocolloids* 2020, **98**:105302.
30. Sandikci Altunatmaz S, Yilmaz Aksu F, Issa G: **Antimicrobial effects of curcumin against L. monocytogenes, S. aureus, S. Typhimurium and E. coli O157: H7 pathogens in minced meat**. *Veterinarni Medicina* 2016.
31. Badee A, Moawd R, ElNoketi M, Gouda M: **Improving the quality and shelf-life of refrigerated chicken meat by marjoram essential oil**. *J Appl Sci Res* 2013, **9**:5718-5729.
32. Liu S, Zhai R, Peng H: **Non-destructive Detection of the pH Value of Cold Fresh Pork Using Hyperspectral Imaging Technique**. In: *International Conference on Computer and Computing Technologies in Agriculture: 2015*. Springer: 266-274.
33. EOS: **Minced Meat Mixed With Soybean Protein. (Egyptian Organization for Standardization and Quality)E.S: 2097/**. 2005.
34. Cheng J-H, Sun D-W, Wei Q: **Enhancing visible and near-infrared hyperspectral imaging prediction of TVB-N level for fish fillet freshness evaluation by filtering optimal variables**. *Food analytical methods* 2017, **10**(6):1888-1898.
35. Sun Y, Zhang M, Adhikari B, Devahastin S, Wang H: **Double-layer indicator films aided by BP-ANN-enabled freshness detection on packaged meat products**. *Food Packaging and Shelf Life* 2022, **31**:100808.
36. Abdel-Hamied A, Nassar A, El-Badry N: **Investigations on antioxidant and antibacterial activities of some natural extracts**. *World Journal of Dairy & Food Sciences* 2009, **4**(1):1-7.
37. Ghalandarlaki N, Alizadeh AM, Ashkani-Esfahani S: **Nanotechnology-applied curcumin for different diseases therapy**. *BioMed research international* 2014, **2014**.
38. Sirocchi V, Devlieghere F, Peelman N, Sagratini G, Maggi F, Vittori S, Ragaert P: **Effect of Rosmarinus officinalis L. essential oil combined with different packaging conditions to extend the shelf life of refrigerated beef meat**. *Food chemistry* 2017, **221**:1069-1076.
39. Assadpour E, Mahdi Jafari S: **A systematic review on nanoencapsulation of food bioactive ingredients and nutraceuticals by various nanocarriers**. *Critical reviews in food science and nutrition* 2019, **59**(19):3129-3151.

Figures

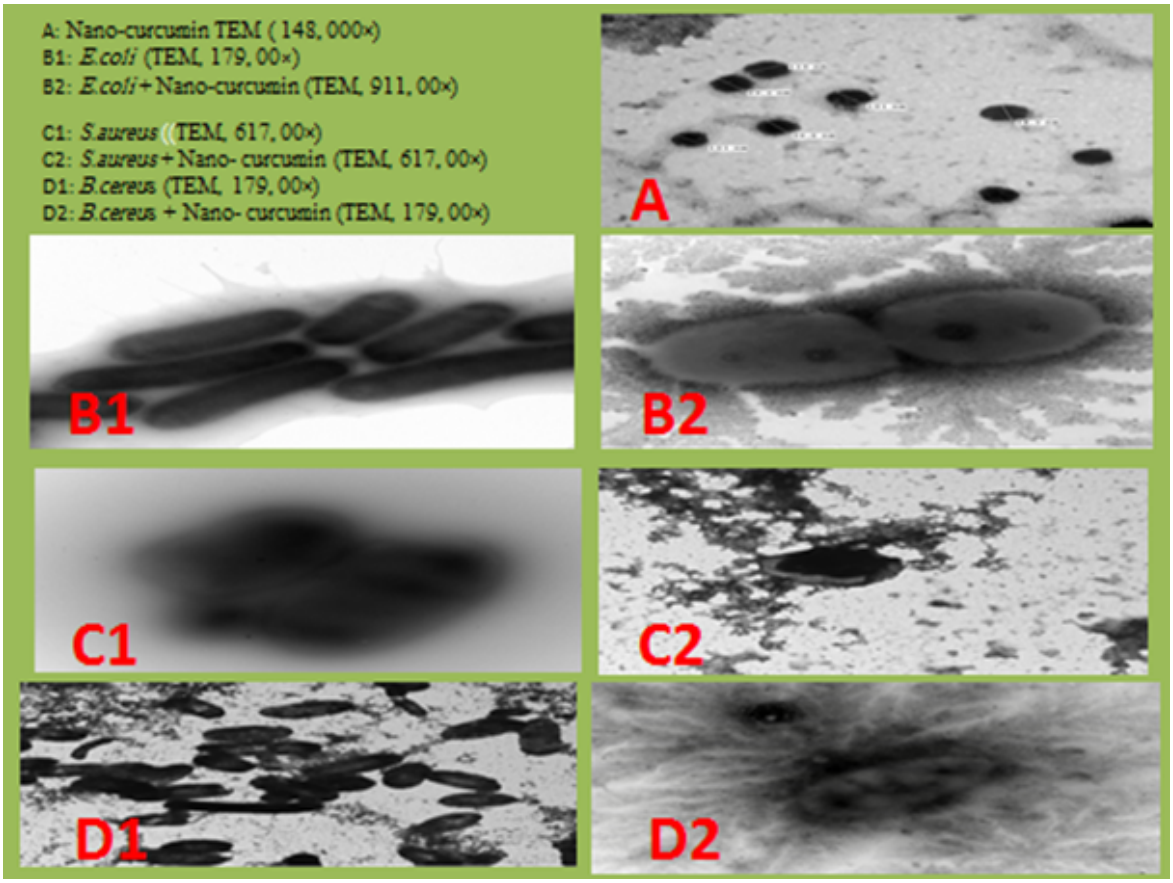

Figure 1

TEM evaluation of the effects of nano-curcumin on the food poisoning bacteria under study.

A: Nano-curcumin with a spherical appearance and size of 90:120 nm (TEM, 148,000×).

B1, C1, D1: Normal bacterial cell structure with organized cell wall for *E.coli* (TEM, 179,000×), *S. aureus* (TEM, 617,000×), and *B. cereus* (TEM, 179,000×). B2, C2, D2: Nano-curcumin effects on *E.coli*, *S. aureus*, and *B. cereus*, showing the accumulation of nano-curcumin inside and around *bacterial cells*.

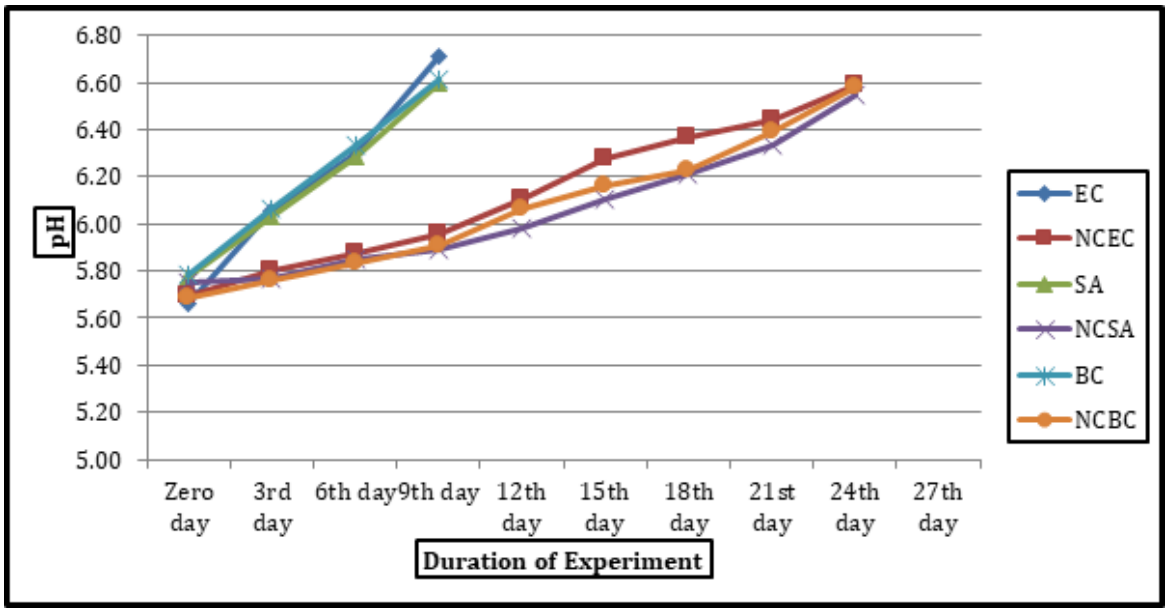

Figure 2

Changes in pH values in chicken kofta inoculated with food poisoning bacteria and treated with nano-curcumin (10 ppm) during cold storage at 4ºC.

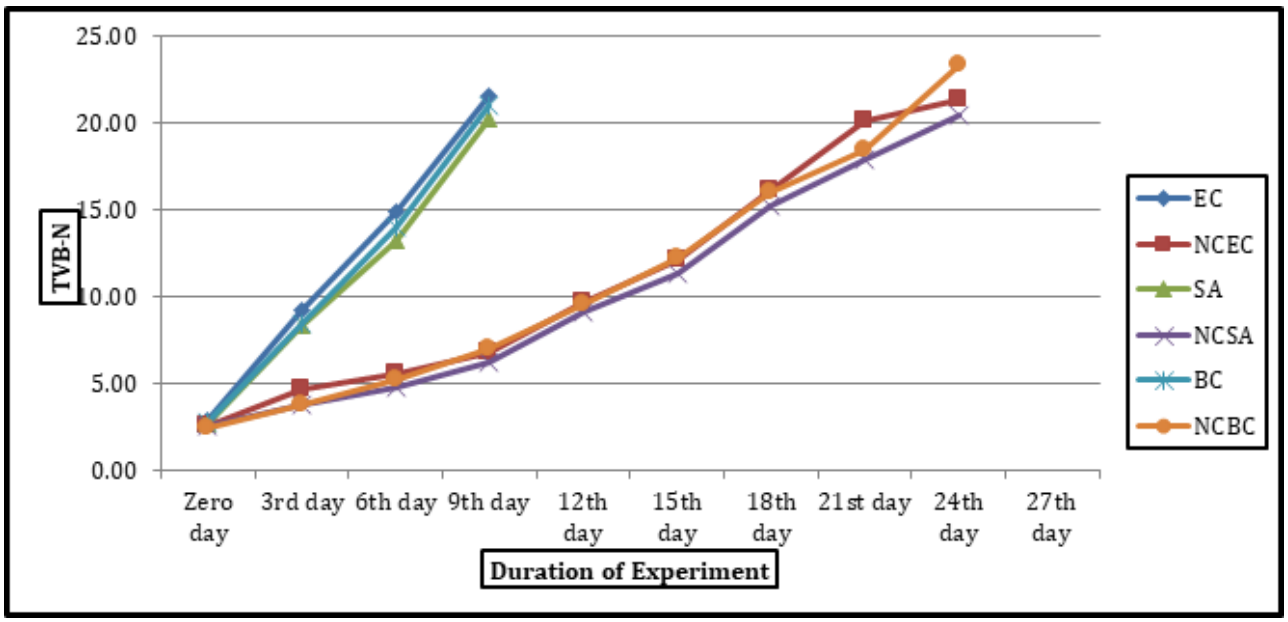

Figure 3

Changes in TVB-N values in chicken kofta inoculated with food poisoning bacteria and treated with nano-curcumin (10 ppm) during cold storage at 4ºC.

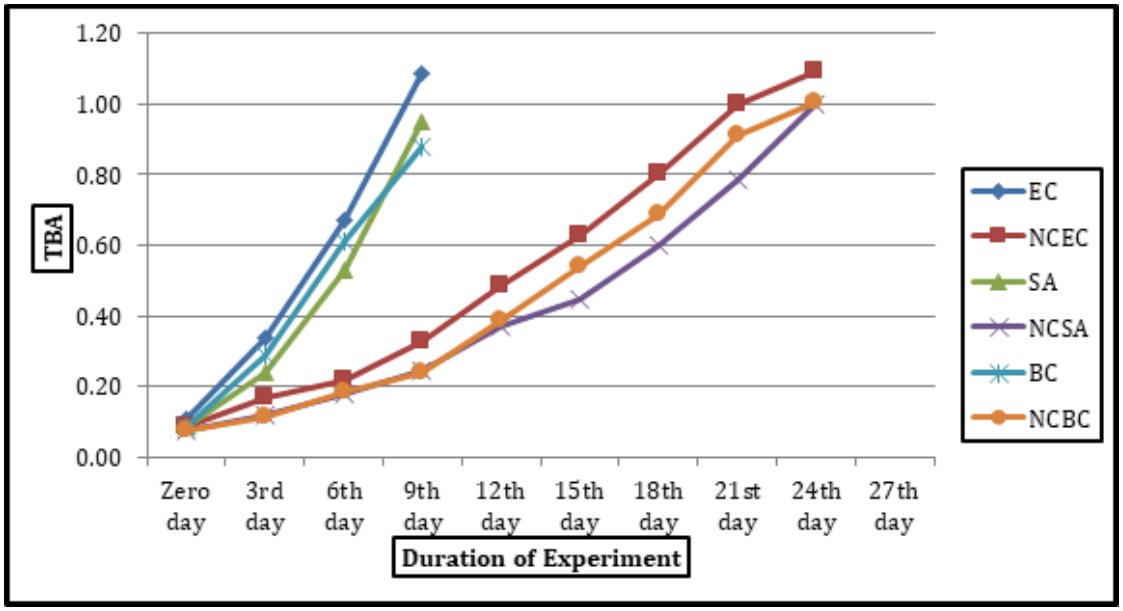

Figure 4

Changes in TBA values in chicken kofta inoculated with food poisoning bacteria and treated with nano-curcumin (10 ppm) during cold storage at 4ºC.

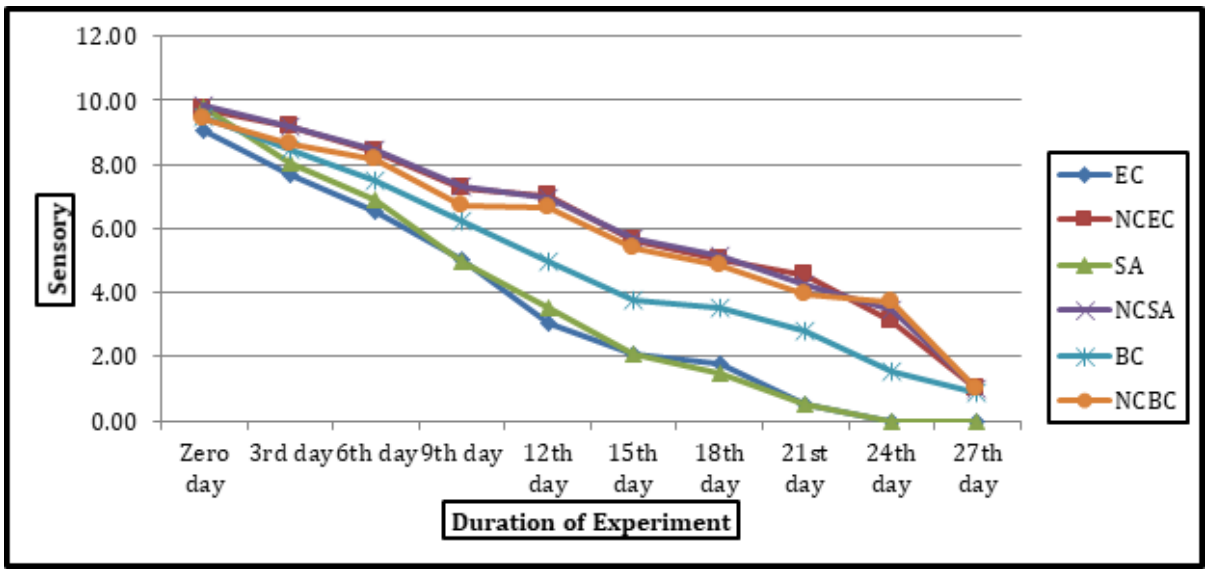

Figure 5

Sensory responses of chicken kofta inoculated with food poisoning bacteria and treated with nano-curcumin (10 ppm) during cold storage at 4ºC.