

## Verifying the ability of mango peel nanoparticles to extend the shelf-life of chilled beef sausage.

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### Abstract

The current study aimed to evaluate mango peel nanoparticles (MPNPs) as a natural preservative containing antioxidants in prolongation of shelf life and keeping the chilled beef sausage quality. MPNPs were prepared using a high-energy planetary ball-mill. After that, prepared beef sausage was divided into 4 groups: group 1; untreated (control), group 2; beef sausage treated with 1% MPNPs, group 3; beef sausage treated with 2% MPNPs and group 4; beef sausage treated with 3% MPNPs. All samples were well mixed and packed in polyethylene packages and stored at  $4\pm 1^{\circ}\text{C}$  for 10 days. All samples were subjected for different examination including sensory evaluation, microbiological examination {aerobic plate count (APC) and total psychrotrophic count} and chemical analysis {pH, thiobarbituric acid (TBA) and Total volatile basic nitrogen (TVB-N)}. Statistical analysis of the obtained data showed that, the sensory evaluation illustrated that the beef sausage treated with 2% MPNPs were found to have satisfactory quality grade for all characteristics. Microbiological examination showed that MPNPs were able to minimize the APC and total psychrotrophic count of beef sausage compared to control one. Moreover, beef sausage mixed with MPNPs had the ability to improve pH, TBA and TVB-N values as matched with the control untreated one. The storage quality and safety parameters of beef sausage treated with MPNPs were within the permissible limit up to 6 days for MPNPs 1% and to 9 days for MPNPs 2% and 3% of refrigerated storage.

**Keywords:** Beef sausage, mango peel nanoparticles, microbiological examination, TBA.

### Introduction

Meat and meat products represent an essential source for protein, amino acids, fat and other nutrients (Biesalski, 2005). Beef sausage is one of the meat products with a short shelf-life as it deteriorates rapidly during storage by microbial growth and/or chemical changes (Turp, 2016), forming a potential health risk. Microbial contamination is a major public health threat and causes a great economic loss (Kingchiyaphum and Rachtanapun, 2012). In addition to microbial contamination, protein decomposition and lipid peroxidation are the main factors that reduce the meat quality and its shelf life (Vuorela *et al.*, 2005 and Lund *et al.*, 2011) with alteration of its color, odor, flavor and nutritional value (Fernández *et al.*,

1997). Oxidative processes result in lipid and protein degradation (Lui *et al.*, 2010). Lipid oxidation takes place with the production of free radicals, its by-product leads to undesirable alterations as oxidation of oxymyoglobin into metmyoglobin which results in dark brown color of meat (Lee *et al.*, 1998). Natural or synthetic antioxidants reduce these oxidative alterations (Ledesma *et al.*, 2015). Nowadays, consumers prefer natural antimicrobials than that chemically treated one (Jadhav *et al.*, 2013) as synthetic one has been found to cause health risk (Shahidi *et al.*, 1992). Natural antioxidants as orange peel (Vinay *et al.*, 2018), rosemary (Guo *et al.*, 2016), pomegranate peel (Morsy *et al.*, 2018) and mango peel powder (Kim *et al.*, 2010) have been used today. Man-

go peel constitutes 15%-20% which is discarded as waste from industrial processing (Thambi *et al.*, 2016). Reuses of biological wastes as mango peel are suggested to solve environmental wastes in addition to create income resources. Polyphenols, carotenoids, phytochemicals, enzymes, vitamins (C & E) and dietary fibers are bioactive ingredients in mango peel and all have functional and antioxidant properties (Kim *et al.*, 2010). In addition to its content of other compounds as protein, lipid, cellulose, hemicellulose, pectin and enzyme (Sogi *et al.*, 2013). Mango peel powder gives characteristic taste and flavor to the food. Moreover, it has antimicrobial activity toward different food borne pathogens which enable it to be used as therapeutic agent in addition to its nutritional value in food industry (Thambi *et al.*, 2016). Recently, new products have been created through nanotechnology to be used in food industries (Rodrigues *et al.*, 2017). These, increase product shelf life and improve its safety and quality (Morsy *et al.*, 2014). Therefore the object of the current study was to investigate the antioxidant as well as the antimicrobial efficiency of mango peel nanoparticle and its ability to prolong the shelf-life of

chilled beef sausage.

## Materials and Methods

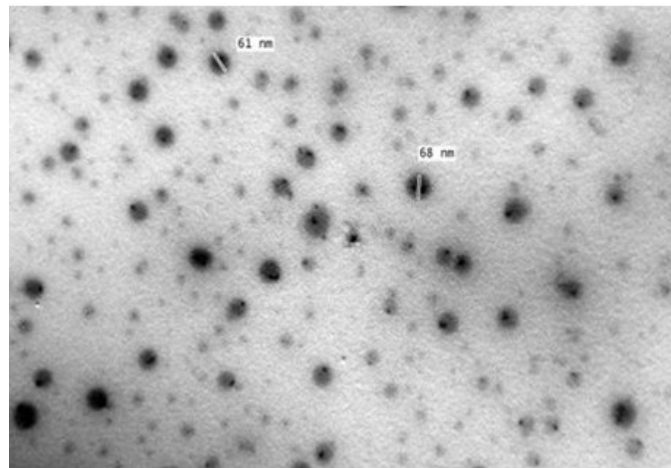
### 1- Mango peel nanoparticles (MPNPs):

#### 1.1-Preparation of mango peel nanoparticles (MPNPs)

Fresh mango fruits with no bruises were purchased from a local market in Egypt. The fruits were transported to the laboratory of National Research Centre were they washed then dried with clean sterile cloth before Peeling. The peels were removed using a sterile sharp knife. The peels were cut into 1-2 cm small pieces then were spread in oven and dried at 50 °C for 12 hours. After drying, the peels were ground with a grinder to micro-particles. These particles were reduced to nanoparticle size according to (Khataee *et al.*, 2017) by their crushing using a high-energy planetary ball-mill at a rotation speed of 320 rpm for 2 h to prepare MPNPs.

#### 1.2-Transmission electron microscopy (TEM):

The morphology and size of MPNPs were analyzed using TEM with negative staining of phosphor tungstic acid (PTA 1%) according to (Kaur *et al.*, 2016).



**Fig. (1):** TEM micrographs of mango peel nanoparticles.

Morphology of the MPNPs was observed using transmission electron microscopy and the size of the particles were qualitatively measured to correlate with the particle size analysis. Transmission electron micrographs of MPNPs are given in Fig. (1), images showed that MPNPs were spherical in shape and the parti-

cles size were in nanometric range.

#### 2-Preparation of beef sausage:

Beef meat samples including boneless neck, chuck and rounds along with associated fats were obtained from local markets at Cairo city, Egypt, and used for preparing beef sausage

samples. All sub cut fat and inter-muscular fat were also included as fat sources. Beef meat and fat tissue were transported to the laboratory using an ice box. Different ingredients used in preparing beef sausage samples e.g. table salt, starch and spices mixture such as black pepper, red pepper, nutmeg and ginger were obtained from local markets at Cairo, Egypt. Beef sausage samples were prepared according to the method described by (Saleh *et al.*, 2016). Meat and fat tissues were cut into pieces of about egg size and ground to particles of about a rice size, then the ingredients were blended to prepare sausage mixture emulsion, which was then stuffed by sausage filling machine previously washed by hot water and cased in mutton casings. The filling was consisted of lean meat 70%, fat 12%, sodium chloride 2.3%, water 9.3%, garlic 1%, onion 1.2% and spices mixture 4.2% and divided into four groups. Group 1: The same ingredients without addition of MPNPs (control group), group 2: one % of ingredients was replaced by MPNPs, group 3: two % of ingredients was replaced by MPNPs and group 4: three % of ingredients was replaced by MPNPs. All groups were packaged in polyethylene packages and stored at  $4^{\circ}\text{C} \pm 1$  for 10 days. Samples were taken at 0, 3, 6, 9- and 10-days interval and subjected to different analysis mentioned below.

### 3-Sensory evaluation:

Sensory evaluation was conducted by an experienced, trained nine-member sensory panel. The panel was composed of employees of the Animal Health Research Institute, Meat hygiene department. The sensory evaluation was determined after cooking of beef sausage using six scale evaluations (appearance, color, tenderness, juiciness, flavor and overall acceptability). sensory characteristics of sausages were evaluated on a 5-point score : a score of 5 was equivalent to highly acceptable, 4 very acceptable, 3 acceptable, 2 low acceptable and a score of 1 indicated unacceptable (Ali *et al.*, 2018).

### 4-Microbiological examination

The microbiological quality of MPNPs treated and control sausage samples was assessed on the basis of aerobic plate count (APC) and total

psychrotrophic count according to the procedure of APHA (2001).

#### 4.1-Aerobic plate count (APC):

Aerobic bacterial count (APC) of the MPNPs treated and control sausage samples were determined on agar medium using Standard Plate Count Agar (Oxoid). Incubation was run at  $35^{\circ}\text{C} \pm 1^{\circ}\text{C}$  for 48h. Bacterial counts were calculated in  $\log_{10}$  cfu/g.

#### 4.2-Total Psychrotrophic count:

Psychrotrophic count was determined in a similar method to that for APC, except that plates were incubated at  $7 \pm 1^{\circ}\text{C}$  for 10 days. The colonies were counted and expressed as  $\log_{10}$  cfu/g.

### 5-Chemical analysis:

**5.1-Hydrogen ion concentration (pH):** It was carried out according to the technique recommended by AOAC (2000)

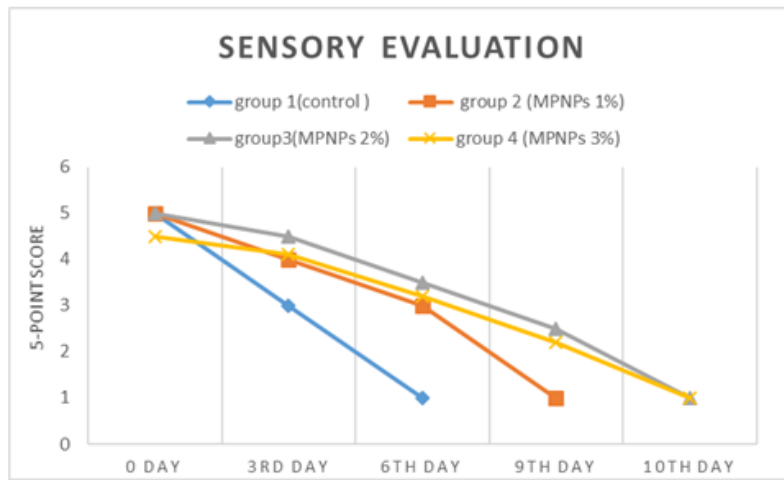
**5.2- Thiobarbituric acid (TBA):** It was carried out according to the technique recommended by AOAC (2000).

**5.3- Total volatile basic nitrogen (TVB-N):** It was done according to the technique recommended by AOAC (2000).

### 6- Statistical analysis

The data obtained were analyzed by one-way analysis of variance (ANOVA), using the Statistical Package of Social Science (SPSS). The values obtained were then expressed as mean  $\pm$  standard deviation (SD), with  $p < 0.05$  being therefore considered as statistically significant. Duncan's multiple range test was applied for the comparison of means and for the determination of the significant difference cause.

**Results and Discussion**



**Fig. (2):** The effects of various concentrations MPNPS on overall acceptability of the examined sausage samples stored at 4±1 °C.

(According to this figure, if the final score is 5 the sample has highly acceptable, 4 very acceptable, 3 acceptable, 2 low acceptable and a score of 1 indicated unacceptable.)

**Table (1).** Statistical analytical results of APC and total psychrotrophic count (log<sub>10</sub> CFU/g) in control and treated samples during cold storage at (4±1°C).

Group		control	MPNPs 1%	MPNPs 2%	MPNPs 3%
APC	0 day	4.03 <sup>a</sup> ±0.07	3.97 <sup>a</sup> ±0.06	3.94 <sup>a</sup> ±0.04	3.89 <sup>a</sup> ±0.03
	3 <sup>rd</sup> day	5.62 <sup>a</sup> ±0.08	4.70 <sup>b</sup> ±0.10	4.18 <sup>c</sup> ±0.13	4.11 <sup>c</sup> ±0.07
	6 <sup>th</sup> day	S	5.77 <sup>a</sup> ±0.06	5.08 <sup>b</sup> ±0.08	4.95 <sup>b</sup> ±0.04
	9 <sup>th</sup> day	S	S	5.52±0.08	5.38±0.08
	10 <sup>th</sup> day	S	S	S	S
Total psychrotrophic count	0 day	2.77 <sup>a</sup> ±0.06	2.69 <sup>a</sup> ±0.04	2.68 <sup>a</sup> ±0.08	2.72 <sup>a</sup> ±0.06
	3 <sup>rd</sup> day	4.10 <sup>a</sup> ±0.10	3.67 <sup>b</sup> ±0.10	3.18 <sup>c</sup> ±0.06	3.00 <sup>c</sup> ±0.08
	6 <sup>th</sup> day	S	3.90 <sup>a</sup> ±0.06	3.77 <sup>a</sup> ±0.08	3.45 <sup>b</sup> ±0.05
	9 <sup>th</sup> day	S	S	4.12±0.10	3.97±0.08
	10 <sup>th</sup> day	S	S	S	S

Values are expressed as mean ± SD with different alphabetical superscript along row are significantly different at (p < 0.05). S=spoiled.

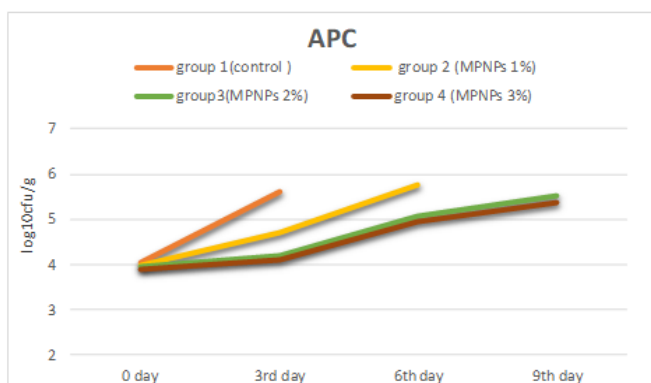


Fig. (3): Changes in aerobic plate count (APC) of treated and non-treated sausage stored at 4±1°C .

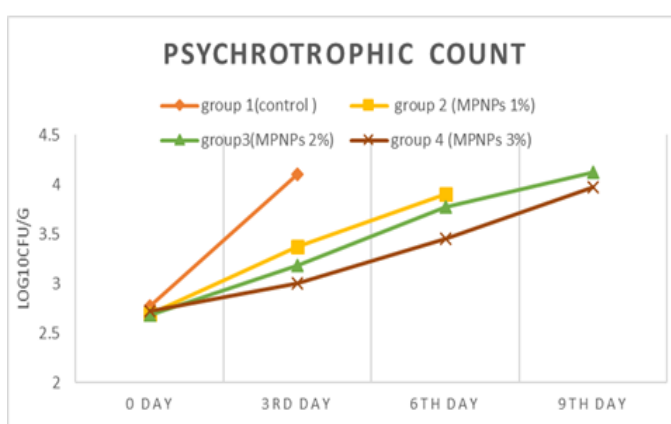


Fig. (4): Changes in Total Psychrophilic count of treated and non-treated sausage stored at 4±1°C.

Table (2). Statistical analytical results of pH value, TBA (mg MDA/kg) and TVB-N (mg/ 100 gm) in control and treated samples during cold storage at (4±1°C).

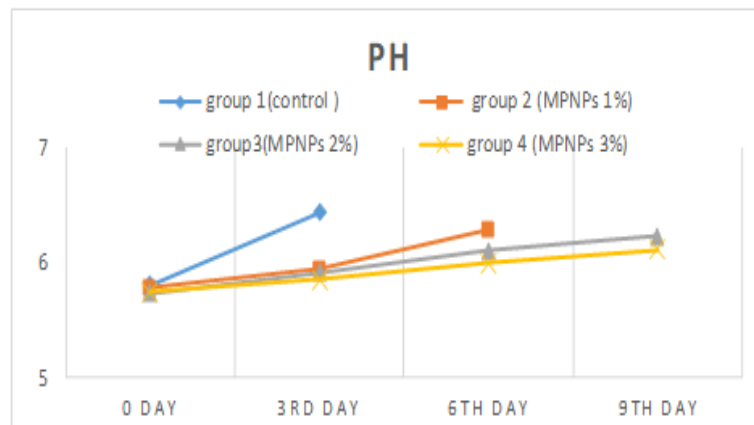
Group		Control	MPNPs 1%	MPNPs 2%	MPNPs 3%
pH	0 day	5.81 <sup>a</sup> ± 0.04	5.78 <sup>a</sup> ± 0.03	5.75 <sup>a</sup> ± 0.02	5.74 <sup>a</sup> ± 0.04
	3 <sup>rd</sup> day	6.44 <sup>a</sup> ± 0.06 c	5.95 <sup>b</sup> ± 0.04	5.92 <sup>b</sup> ± 0.04	5.86 <sup>b</sup> ± 0.04 a
	6 <sup>th</sup> day	S	6.29 <sup>a</sup> ± 0.06	6.11 <sup>b</sup> ± 0.09	6.00 <sup>b</sup> ± 0.05
	9 <sup>th</sup> day	S	S	6.23± 0.08	6.12± 0.9
	10 <sup>th</sup> day	S	S	S	S
TBA	0 day	0.32 <sup>a</sup> ± 0.01	0.32 <sup>a</sup> ± 0.02	0.31 <sup>a</sup> ± 0.02	0.30 <sup>a</sup> ± 0.01
	3 <sup>rd</sup> day	0.86 <sup>a</sup> ± 0.07	0.55 <sup>b</sup> ± 0.04	0.56 <sup>b</sup> ± 0.08	0.51 <sup>b</sup> ± 0.04
	6 <sup>th</sup> day	S	0.77 <sup>a</sup> ± 0.08	0.76 <sup>a</sup> ± 0.04	0.70 <sup>a</sup> ± 0.04
	9 <sup>th</sup> day	S	S	0.89± 0.09	0.87± 0.11
	10 <sup>th</sup> day	S	S	S	S
TVB-N	0 day	10.69 <sup>a</sup> ± 1.10	10.53 <sup>a</sup> ± 1.02	10.31 <sup>a</sup> ± 1.01	10.31 <sup>a</sup> ± 0.80
	3 <sup>rd</sup> day	19.11 <sup>a</sup> ± 0.35	14.35 <sup>b</sup> ± 1.10	13.15 <sup>c</sup> ± 0.60 a	13.19 <sup>c</sup> ± 1.09
	6 <sup>th</sup> day	S	17.60 <sup>a</sup> ± 1.04	16.69 <sup>b</sup> ± 1.07	15.86 <sup>c</sup> ± 1.00
	9 <sup>th</sup> day	S	S	19.14± 0.09	18.37± 0.09
	10 <sup>th</sup> day	S	S	S	S

Values are expressed as mean ± SD with different alphabetical superscript along row are significantly different at (p < 0.05)

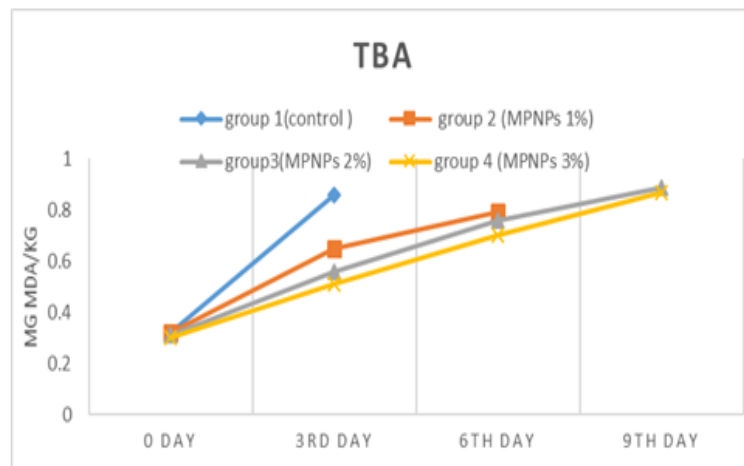
TBA: 0.9 mg malondialdehyde (MDA)/kg beef sausage (ES. 1972/2005)

TVB-N: 20mg/100gm beef sausage (ES. 1972/2005).

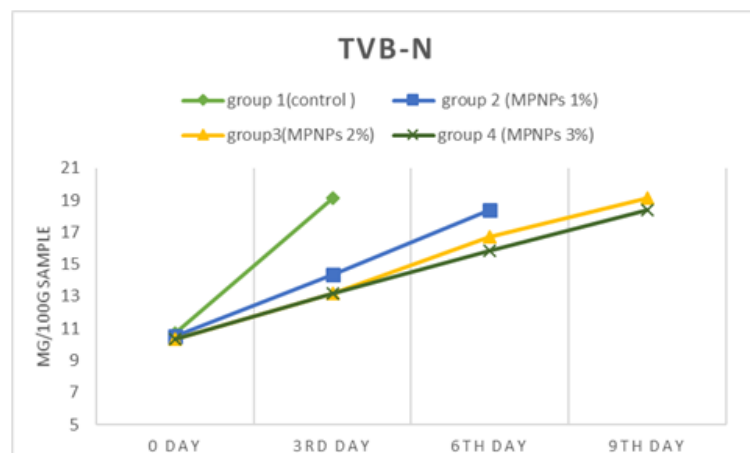
S =spoiled



**Fig. (5):** Changes in PH of treated and non-treated sausage stored at 4±1°C.



**Fig. (6):** Changes in TBA of treated and non-treated sausage stored at 4±1°C.



**Fig. (7):** Changes in TVB-N of treated and non-treated sausage stored at 4±1°C:

Consumers and meat processors prefer the use of natural additives that exhibit antioxidant, antimicrobial and health benefits to overcome the problems that may arise from the use of synthetic ingredients. Therefore, the current study was designed to evaluate the antioxidant and antibacterial effect of MPNPs as natural ingredient for improving the quality of fresh sausage during refrigerated storage at  $4\pm 1^{\circ}\text{C}$ .

The sensory scores of both the control and treated sausage samples with MPNPs 1%, 2% and 3% were decreased with storage period. The observed data regarding the shelf life of the 1<sup>st</sup> control group of sausage as determined by panelists indicated that the sausage was found spoiled on the 6<sup>th</sup> day while the obtained data indicated sound samples until the 3<sup>rd</sup> day of storage period, while the 2<sup>nd</sup>, 3<sup>rd</sup> and the 4<sup>th</sup> treated groups were spoiled on the 9<sup>th</sup>, 10<sup>th</sup> and 10<sup>th</sup> day of storage, respectively. So, the analytical data was recorded till the 6<sup>th</sup>, 9<sup>th</sup> and 9<sup>th</sup> day of storage only. The panelists rejected the sausage samples of both untreated and treated groups at the time of its spoilage throughout the storage period as it considered unacceptable for the consumers even though the microbial load did not exceed of  $6 \log_{10}\text{cfu/g}$  (**ES: 1972/2005**). At zero day the sensory scores of the 1<sup>st</sup>, 2<sup>nd</sup> and 3<sup>rd</sup> groups had no significantly between each other. Meanwhile, group 4<sup>th</sup> of sausages showed lower score than the other groups at ( $p < 0.05$ ) as the addition of 3% MPNPs to sausage give yellow color to sausage with no effect on the other scales of evaluation. Moreover, the 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> groups scored higher than the 1<sup>st</sup> group at ( $p < 0.05$ ) during the storage period (Fig 2). From the obtained data, it can be concluded that all treated (1%, 2% and 3% MPNPs) groups maintain better sensory quality for the sausage than the untreated control one and 3<sup>rd</sup> group was the best.

In general, increase concentration of mango peels nanoparticles led to increase the acceptability and extend shelf life for prepared beef sausage (**Abdel-Moemin, 2015**). The mango Peel maintained the redness, color, the pleasant odor and retarded the off-odor of sausage (**Le, 2012**). These findings indicated that mango peel can be utilized as a natural antioxidant and

antimicrobial due to its high content of phenolic compounds.

The present data were compared with the **ES. 1972/2005** for frozen sausage because there was not a standard for chilled sausage. The recorded data in table (1) and Fig. (3) showed that mean APC of control sausage sample was  $4.03\pm 0.07 \log_{10} \text{cfu/g}$  at 0 day then it increased to  $5.62\pm 0.08$  at the 3<sup>th</sup> day of storage, whereas the analysis was stopped at the 6<sup>th</sup> day of chilled storage as the samples were spoiled. Meanwhile, the MPNPs 1% treated samples were  $3.97\pm 0.06$ ,  $4.70\pm 0.10$  and  $5.77\pm 0.06 \log_{10} \text{cfu/g}$  at 0, 3<sup>rd</sup> and 6<sup>th</sup> day, respectively while found spoiled at the 9<sup>th</sup> day. MPNPs 2% treated samples were recorded  $3.94\pm 0.04$ ,  $4.18\pm 0.13$ ,  $5.08\pm 0.08$  and  $5.52\pm 0.08 \log_{10}\text{cfu/g}$  at 0, 3<sup>rd</sup>, 6<sup>th</sup> and 9<sup>th</sup> day respectively, and MPNPs 3% treated samples were  $3.89\pm 0.03$ ,  $4.11\pm 0.07$ ,  $4.95\pm 0.04$  and  $5.38\pm 0.08 \log_{10} \text{cfu/g}$  at 0, 3<sup>rd</sup>, 6<sup>th</sup>, 9<sup>th</sup> day respectively. On the other hand, the total psychrotrophic count in the 1<sup>st</sup> control group, 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> group samples at 0 day were  $2.77\pm 0.06$ ,  $2.69\pm 0.04$ ,  $2.68\pm 0.08$ , and  $2.72\pm 0.06 \log_{10}\text{cfu/g}$ , respectively. While the four groups recorded  $4.10\pm 0.10$ ,  $3.90\pm 0.06$ ,  $4.12\pm 0.10$  and  $3.97\pm 0.08$  on the 3<sup>rd</sup>, 6<sup>th</sup>, 9<sup>th</sup> and 9<sup>th</sup> day of cold storage for the 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> groups, respectively. (Table 1 and Fig. 4).

The statistical analysis of the above mentioned data revealed a significant difference between the microbial load of control group and the MPNPs treated one during storage period. The more pronounced effect was MPNPs 2% followed by 3%. The result came in accordance with that of (**Abdeldaim and Ali, 2012**) who revealed that a significant decrease of Aerobic plate count of beef burgers treated with irradiated mango peel powder. Also, **Bhat et al. (2017)** recorded that, mango peel powder decreased the aerobic plate count and yeast & mould count of chicken culets during storage. **Thambi et al. (2016)** stated the antimicrobial effect of mango peel powder against various pathogen and they suggested its usage in food industry. The decrease in the microbial load can be attributed to the constituent of mango peel powder as it contains phenolic compounds which have antimicrobial activity (**Bhat et al.,**

**2017).** Phenolic compounds change the pH and electrical potential releasing the protons outside, this will lead to the coagulation of the bacterial cytoplasmic content then loss of normal cell metabolism and cell death (**Raybaudi-Massilia *et al.*, 2009**).

The present data in table (2) and fig. (5) showed that pH value of beef sausage samples during storage for 10 days. pH value of control sample increased throughout the experimental period from day 0 ( $5.81 \pm 0.04$ ) to ( $6.44 \pm 0.06$ ) at the 3<sup>rd</sup> day of storage, while the 1<sup>st</sup> control group samples were found spoiled at the 6<sup>th</sup> day of storage. Meanwhile the MPNPs 1%, 2% and 3% treated samples pH values were  $5.78 \pm 0.03$ ,  $5.75 \pm 0.02$  and  $5.74 \pm 0.04$  at 0 day meanwhile they were  $6.29 \pm 0.06$ ,  $6.23 \pm 0.08$  and  $6.12 \pm 0.09$  at the 6<sup>th</sup>, 9<sup>th</sup> and 9<sup>th</sup> day of storage while these groups were found spoiled at 9<sup>th</sup>, 10<sup>th</sup>, and 10<sup>th</sup> day of storage, respectively. Activation effect of the microbial load, protein hydrolysis and the appearance of alkyl groups cause an increase in the pH values (**Yassin, 2003**). Also, the storage at refrigeration temperature (4°C) causes many chemical reactions including enzymatic reactions and formation of volatile nitrogenous basic compound (**Ibrahim and Desouky, 2009**) which increases pH value. The decrease in pH value of treated groups may be attributed to the antimicrobial effect of MPNPs.

Concerning to the TBA results in table (2) and Fig. (6), its mean value for control sample was  $0.32 \pm 0.01$  (mg MDA/kg) at 0 day and increased to  $0.86 \pm 0.07$  (mg MDA/kg) at the 3<sup>rd</sup> day of storage. While TBA values of treated samples were recorded  $0.65 \pm 0.04$ ,  $0.56 \pm 0.08$  and  $0.51 \pm 0.04$  (mg MDA/kg) in the 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> group samples at the 3<sup>rd</sup> day, respectively. And  $0.79 \pm 0.08$ ,  $0.89 \pm 0.09$  and  $0.87 \pm 0.11$  (mg MDA/kg) in the 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> group samples at 6<sup>th</sup>, 9<sup>th</sup> and 9<sup>th</sup> day of storage period respectively, which considered below the permissible limit. TBA value is used as an indicator of lipid oxidation in meat products (**Raharjo and Sofos, 1993**). The data revealed a significant difference between control and treated samples. During storage TBA values of all samples were gradually increased

along the time of storage, MPNPs lower the TBA values than control one. Increasing trend of TBA value during chilling is an indicator for the continuous lipid oxidation and oxidative by-product formation (**Abdou *et al.*, 2018**). According to **ES.1972/2005** the maximal permissible limit is 0.9 mg MDA/kg for TBA. The result came in accordance with that of **Baht *et al.* (2017)** who showed that mango peel powder lower thiobarbituric acid reacting substances of stored chicken cutlets up to 10 days. Also, **Abdel-Moemin (2015)** recorded that Mango Kernel lowers the thiobarbituric acid reacting substances and increases the shelf life of beef sausages. Mango peel has antioxidant properties by its phenolic compounds (**Abdel-daim and Ali, 2012 and Palmeira *et al.*, 2012**) and decreases oxidative rancidity and oxidative by-product during refrigerator (**Baht *et al.*, 2017**). (**Abdalla *et al.*, 2007**) also suggested that mango waste extracts could be used as a source of natural antioxidants. Mango peel is a rich source of phenolic compounds, which exhibit antioxidant activity.

The obtained data in Table (2) and Fig. (7) revealed that the mean value of TVB-N of the control group samples recorded  $10.69 \pm 1.10$  at the 0 day and  $19.11 \pm 0.35$  at the 3<sup>rd</sup> day of storage, period, while the examination was not continued on the 6<sup>th</sup> day of storage because the control group samples were found in a state of complete spoilage. Furthermore, TVB-N values were recorded  $10.53 \pm 1.02$ ,  $10.31 \pm 1.01$  and  $10.31 \pm 0.8$  at the 0 day for the 2<sup>nd</sup>, 3<sup>rd</sup> and the 4<sup>th</sup> treated groups respectively, while  $17.60 \pm 1.04$ ,  $19.14 \pm 0.09$  and  $18.37 \pm 0.9$  at the 6<sup>th</sup>, 9<sup>th</sup> and the 9<sup>th</sup> day of the storage for the 2<sup>nd</sup>, 3<sup>rd</sup> and the 4<sup>th</sup> treated groups respectively, such levels considered below the permissible level of TVB-N (20 mg /100 g) of sausage according to **ES. 1972/2005**.

A significant difference was statistically recorded between the control sample and treated one. The results showed that the TVB-N values were increased through the storage time in beef sausage with the lowest increment in the treated groups with the MPNPs 1% followed by 2% then 3%. TVB-N value is an important indicator for measuring the extent of protein degradation into amino acids then the meat and



meat products putrefaction (**Han, et al., 2001**). Also, TVB-N increment is an indicator of microbial activity and proteolytic enzymes (**Yassin, 2003**). (**El-Nashi et al., 2015**) achieved that, the elevation of TVB-N value during cold storage of meat is due to nitrogenous substances breakdown by microbial activity. In the present study MPNPs were able to reduce the TVB-N increment during storage of beef sausage, antimicrobial activity and anti-fungal of mango peel powder were achieved by (**Thambi et al., 2016**). A Similar finding was recorded by (**Baht et al., 2017**) who declared that mango peel powder 3% can be incorporated in chicken cutlets for prolonging its shelf life in refrigerator.

### Conclusion

The present study achieved that the beef sausage stored at chilling temperature ( $4\pm 1^{\circ}\text{C}$ ) was within the permissible limit at 3<sup>rd</sup> day as the microbial load, TBA and TVB-N, while the 1<sup>st</sup> control group samples were found spoiled at 6<sup>th</sup> day. Meanwhile MPNPs 1% was able to maintain these parameters for 6 days, while MPNPs 2% and 3% for 9 days. Treatment of sausage by MPNPs 2% was the best way to keep the quality and safety parameters of sausage as MPNPs significantly reduced the APC and psychrotrophic as well as hindering the deterioration of sausage stored at chilling temperature ( $4\pm 1^{\circ}\text{C}$ ) and extending its shelf-life aiding in the prevention of economic loss and providing the customer with a meat product containing natural agent with antioxidant and antimicrobial activities against food-borne pathogens and spoilage organisms.

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