THE ROLE OF NISIN, POTASSIUM SORBATE AND SODIUM LACTATE AS ADDITIVE IN IMPROVING THE CHEMICAL AND QUALITATIVE CHARACTERISTICS OF CHILLED GROUND BEEF.

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ABSTRACT

This study was conducted to evaluate the effects of nisin, potassium sorbate and sodium lactate and their interactions as preservatives in fat oxidation, some quality and microbial characteristics of chilled ground beef meat stored at 4C° for 0,4,8 and 12 days. The study included nine different treatments: T1 (control treatment without addition), T2 (nisin at a concentration of 150 mg/kg), T3 (potassium sorbate 0.075 g/kg), T4 (sodium lactate 3%), T5 (nisin 150 mg/kg + potassium sorbate 0.075 g/kg), T6 (nisin 150 mg/kg + sodium lactate 3%), T7 (Sodium Lactate 3% + Potassium Sorbate 0.075 g/kg), T8 (Nisin 150 mg/kg + Sodium Lactate 3% + Potassium Sorbate 0.075 g/kg) and T9 (Addition of Butylated hydroxyanisole(BHA 0.01%)). All treatments showed significant (p<0.01) decreases in dry matter percentage of (fat and ash) and increase in protein and moisture and water holding capacity (WHC) percentage as compared with the control treatment at any refrigerated storage time. Thiobarbituric acid (TBA) were lower (P<0.01) in nisin, potassium sorbate and sodium lactate treatments when compared with the control treatment at any refrigerated storage time. Nisin, potassium sorbate and sodium lactate treated samples reduced (P<0.01) the total plate count of bacteria during refrigerated storage times. It can be concluded that nisin, potassium sorbate and sodium lactate had positive significant influence on quality characteristics and microbial safety of ground beef meat when stored under refrigeration at 4C° up to 12 days.

Key words: nisin, beef, thiobarbituric acid, water holding capacity, TBC.
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دور النيسين وسوربات البوتاسيوم ولاكتات الصوديوم كأضافات في تحسين الصفات الكيميائية والنوعية للحم البقر المفروم المبرد

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المستخلص

أجريت هذه الدراسة لتقييم تأثير النيسين وسوربات البوتاسيوم ولاكتات الصوديوم كمواد حافظة في أكسدة الدهون، وبعض الخصائص النوعية والمايكروبية للحم البقر المفروم المبرد المخزن عند 4 م° للفترات 0، 4، 8 و 12 يوم. اشتملت الدراسة على تسع معاملات مختلفة كالآتي: T1 (معاملة السيطرة بدون اضافة)، T2 (نيسين 150 ملي غرام/غرام)، T3 (سوربات البوتاسيوم 0.075 غرام/كيلو غرام) T4 (لاكتات الصوديوم 3%)، T5 (نيسين 150 ملي غرام/غرام + سوربات البوتاسيوم 0.075 غرام/كيلو غرام)، T6 (نيسين 150 ملي غرام/غرام + لاكتات الصوديوم 3%)، T7 (لاكتات الصوديوم 3% + سوربات البوتاسيوم 0.075 غرام/كيلو غرام)، T8 (نيسين 150 ملي غرم/غرام + لاكتات الصوديوم 3% + سوربات البوتاسيوم 0.075 غرام/كيلو غرام)، T9 (اضافة BHA) أظهرت جميع المعاملات انخفاضاً معنوياً (p <0.01) في نسبة المادة الجافة من (الدهن والرماد)وزيادة في نسبة البروتين الرطوبة و قابلية اللحم على الاحتفاظ بالماء (WHC)، انخفضت قيم حامض الثايوباربتيورك والعدد الكلي للبكتريا في معاملات الاضافة مقارنة مع معاملة السيطرة خلال فترات الحفظ، يمكن الاستنتاج أن اضافة النيسين وسوربات البوتاسيوم ولاكتات الصوديوم كان لها تأثير إيجابي على الخصائص النوعية والمايكروبية للحم البقر المفروم المبرد على درجة 4 م° حتى 12 يوما من الخزن.

الكلمات المفتاحية: نيسين، لحم مفروم، حامض الثايوباربتيورك، قابلية مسك الماء، العدد الكلي للبكتريا.
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INTRODUCTION

Nowadays, public health concerns related to microbial food safety have emerged, as many epidemiological reports have indicated that meat and meat products are the main agents responsible for diseases caused by foodborne pathogens, and perishable foods such as meat provide excellent conditions for the growth of dangerous microorganisms (18). It is known that the growth of microorganisms is greatly affected by environmental factors, food preservatives can be considered as a kind of environmental factors that affect the growth of microorganisms (38). Therefore, attention to storage conditions hinders microbial growth and significantly reduces meat spoilage; moreover, adding some salts to meat is a catalyst in delaying undesirable biological, chemical and physical changes and improving sensory evaluation such as color, flavor, tenderness and juiciness (23). One of the methods used to preserve meat and control meat-borne pathogens is nisin, which is one of the bacteriocins produced by lactic acid bacteria. It has been shown that nisin is active in prevent or controlling spoilage and disease-causing bacteria in many meat products (19, 26, 31) have been shown to be effective in inhibiting Gram-positive bacteria and their spores (1). Therefor the objective of this work was to study the role of nisin, potassium sorbate and sodium lactate as additive in improving the chemical and qualitative characteristics of chilled ground beef meat.

MATERIALS AND METHODS

Samples: The objective of this experiment was carried out in the Meat Science and Technology Laboratory of of animal production department and in the animal nutrition laboratory for postgraduate students at College of Agricultural Engineering Sciences, University of Baghdad. We took 18kg of meat from the leg of the calf at age of 1.5 years immediately after the slaughter process from the local market and it was minced with an electric mincing machine, and it was left at the refrigerator for 12 hours to remove the rigor mortis state. Then the meat was divided into 9 parts of 2 kg for each. After that, each part was treated with the additives (Nisin, Potassium Sorbate, Sodium Lactate, BHA).

Treatments

The experiment included nine treatments as follows: T1(control treatment without addition), T2(nisin 150mg/ kg), T3(potassium sorbate 0.075 g/ kg), T4(sodium lactate 3%), T5(nisin 150mg/ kg + potassium sorbate 0.075g/ kg), T6(nisin 150mg/ kg + sodium lactate 3%), T7(sodium lactate 3% + potassium sorbate 0.075g/ kg), T8(nisin 150mg/ kg + sodium lactate 3% + potassium sorbate 0.075g/ kg) and T9(addition of Butylated hydroxyanisole(BHA 0.01%)), each treatment was homogenized separately by hand using medical gloves to obtain a separated homogeneous sample. The samples were placed in polyethylene bags and stored in the refrigerator for different periods of days (0, 4, 8, 12 days) at 4C˚ to conduct the effect of the mentioned additives on the quality and microbial characteristics of the chilled ground beef meat.

Laboratory analyses

The chemical composition (moisture, protein, fat and ash) was determined according to the method of (12). Water holding capacity (WHC) according to (8), TBA were tested separately in duplicate according to the method of (34) and Total Bacterial Count (TBC) was counted according to (28)

Statistical analysis

Data was statistically analyzed using Completely Randomized Design Model (CRD) as a factorial experiment (4×9). Duncan's multiple rang test was used to determine the significant differences among treatments means and periods using SAS(SAS, 2012) (20, 30).

RESULTS AND DISCUSSION

Results in Table (1) shows the interaction between the different treatments and periods of cold storage for the chilled ground beef meat of moisture. All treatments showed a significant ($p<0.01$) increases in moisture percentage with the increase of storage time (0, 4, 8 and 12 days), nisin and salts treatments showed a significant ($p<0.01$) increases in moisture percentage as compared with control treatment. T3 showed the highest percentage of moisture (75.13%) at zero time than other treatments when compared with the control treatment (64.21%) in the last period of cold storage. The increases in the moisture

percentage of the different addition treatments for the chilled ground beef meat may be attributed to the synergistic effect of salts and nisin on increasing the moisture percentage, in addition to the increase in the solubility of meat proteins that bind with water when the ionic strength is increased (6). The periods of cold storage were indicated a significant difference (P<0.01) between the periods of cold storage, where the moisture reached its highest level in the first period (0 days) down to its lowest percentage in the last period of cold storage (12 days). While a decrease in the percentage of moisture is noted with the increase of periods of cold storage and an increase in the dry matter represented by fat, protein and ash. This is consistent with (6) who found the same effect of sodium lactate, potassium sorbate and cetyipyridinium chloride on the physicochemical, sensory and microbial characteristics of chilled minced beef.

Table 1. Effect of nisin, potassium sorbate and sodium lactate on moisture of chilled ground beef meat

Treatments No.	Moisture (%) Storage periods (days)				Range
	0	4	8	12	
T1	71.27JKL ±0.07	70.63O ±0.04	67.70T ±0.06	64.21Z ±0.03	68.45h ±0.84
T2	73.62CD ±0.03	71.65HI ±0.02	68.38R ±0.06	65.93V ±0.03	69.89c ±0.89
T3	75.13A ±0.04	73.81C ±0.03	71.15KL ±0.06	69.10Q ±0.04	72.29a ±0.70
T4	72.25F ±0.06	70.91MN ±0.10	68.05S ±0.06	64.73X ±0.06	68.98f ±0.87
T5	74.80B ±0.05	71.77GH ±0.33	70.00P ±0.06	67.11U ±0.04	70.92b ±0.84
T6	73.75C ±0.04	71.93G ±0.07	63.12A ±0.06	66.90U ±0.05	68.92f ±1.26
T7	72.97E ±0.06	71.10LM ±0.03	68.35R ±0.05	65.62W ±0.04	69.51e ±0.83
T8	73.52D ±0.03	71.35JK ±0.04	68.40R ±0.05	65.78VW ±0.03	69.76d ±0.88
T9	71.48IJ ±0.07	70.75NO ±0.05	67.98S ±0.05	64.50Y ±0.04	68.67g ±0.82
Range	73.19a ±0.24	71.54b ±0.18	68.12c ±0.40	65.98d ±0.28	

The averages that bear different letters, differ significantly (P<0.01) among them (lowercase letters are main effect of transactions and storage times and large letters are the effect of the intraction between treatments and storage times).

Table (2) shows the effect of the interaction between the different treatments and periods of cold storage on the protein percentage of the chilled ground beef, as it was noticed a significant increase (P<0.01) in the percentages of protein between different treatments with the progression of the cold storage periods; T3 (potassium sorbate 0.075 g/kg) recorded a highest protein percentage (19.40%) among the different treatments compared to the control treatment in the first period of the cold storage (0 days) up to 23.32% in the last period of the cold storage (12 days), while T1 (control without addition) had the lowest protein percentage (16.36%) in the 0-day storage period and for all subsequent periods. Also noted from Table (2) a significant differences (P<0.01) between the value of the different treatments, where T3 (potassium sorbate 0.075 g/kg) outperformed the rest of the treatments, followed by T2, which did not differ significantly with T5 and T6 then T8, T7, T4 and T9 then T1. Some studies indicated a high percentage of protein in meat by the effect of adding natural antioxidants when preserving by refrigerating and freezing (2) and this may be due to an increase in the solubility of meat proteins which works to increase the bonding with water when ionic strength are increased and the effectiveness of the enzymes (Cathepsins) and (Calpains), which contribute to increasing the solubility and separation of muscle fiber proteins (3). The results of the statistical

analysis indicated that the percentage of protein increased with the progression of the periods of cold storage, as it was at its lowest percentage in the first storage period (0 days) and its highest percentage in the 12 day cooling period, where protein, fat and ash increase at the expense of moisture percentage with the progression of the cold storage period, which led to an increase in the percentage of protein in all treatments and for all storage periods (0, 4, 8 and 12 days) as compared with the control treatment.

Table 2. Effect of nisin, potassium sorbate and sodium lactate on protein of chilled ground beef meat

Treatments No.	Protein (%) Storage periods (days)				Range
	0	4	8	12	
T1	16.36S ±0.06	17.75P ±0.05	19.25L ±0.06	20.60HI ±0.05	18.49g ±0.48
T2	18.39MN ±0.08	19.61JK ±0.07	21.53F ±0.16	22.64BC ±0.03	20.54b ±0.49
T3	19.40KL ±0.1	20.38I ±0.06	22.00E ±0.10	23.32A ±0.09	21.27a ±0.45
T4	17.40Q ±0.05	18.51M ±0.08	20.41I ±0.09	21.50F ±0.06	19.45e ±0.48
T5	18.62M ±0.08	19.80J ±0.05	20.50I ±0.11	22.80B ±0.05	20.43b ±0.46
T6	18.55M ±0.04	19.43KL ±0.10	21.40F ±0.07	22.37CD ±0.06	20.43b ±0.45
T7	17.90OP ±0.06	18.45M ±0.08	20.58HI ±0.09	21.82E ±0.06	19.68d ±0.47
T8	18.15NO ±0.08	19.15L ±0.09	21.10G ±0.05	22.32D ±0.09	20.18c ±0.49
T9	16.70R ±0.37	17.93OP ±0.07	19.44KL ±0.06	20.87GH ±0.11	18.73f ±0.48
Range	17.94d ±0.18	19.00c ±0.16	20.69b ±0.17	22.02a ±0.16	

The averages that bear different letters, differ significantly (P<0.01) among them (lowercase letters are main effect of transactions and storage times and large letters are the effect of the intraction between treatments and storage times).

The results of the interaction between the treatments and the periods of cold storage of fat percentage are shown in table (3). All treatments showed a significant ($p<0.01$) decrease in fat percentage with the increasing of storage time (0, 4, 8 and 12 days), nisin and salts treatments showed a significant ($p<0.01$) decrease in fat percentage when compared with control treatment. Lowest percentage of fat (3.86%) was associated with T3 in the first storage period (zero days of chilling) as compared with the control treatment which showed the highest percentage of fat percentage (12.61%) in the last storage period (12 days of chilling). This may be attributed to the decomposition caused by lipase and phospholipase, which leads to a decrease in the percentage of fat in the meat, especially that these added salts (potassium sorbate and sodium lactate) increase the effectiveness of these enzymes (6). For the periods of cold storage, the results of the statistical analysis indicated a significant difference ($P<0.01$) between periods. The percentage of fat was at its lowest level in the first storage period (0 days) and then began to rise with the passage of storage periods until it reached its highest percentage (9.72%) in period of 12 days of refrigeration, and this is normal, since the increase of the storage period works to reduce the moisture and raise the percentage of dry matter that is represented by both protein, fat and ash, as the percentage of fat increases with the progression of the refrigeration storage period until it reaches its highest level in the 12-day storage period (8).

Table 3. Effect of nisin, potassium sorbate and sodium lactate on fat of chilled ground beef meat

Treatments No.	Fat (%) Storage periods (days)				Range
	0	4	8	12	
T1	10.33E ±0.05	9.55G ±0.04	10.66D ±0.04	12.61A ±0.05	10.78a ±0.34
T2	6.06S ±0.09	6.92Q ±0.05	7.24P ±0.04	9.24H ±0.04	7.36f ±0.35
T3	3.86X ±0.11	4.16W ±0.04	5.08U ±0.04	5.73T ±0.05	4.70i ±0.22
T4	8.42L ±0.06	8.68J ±0.04	9.34H ±0.06	11.22C ±0.04	9.41c ±0.33
T5	4.63V ±0.05	7.41O ±0.05	7.60N ±0.04	8.15M ±0.04	6.94h ±0.41
T6	5.72T ±0.05	6.88Q ±0.03	7.37OP ±0.03	8.63JK ±0.04	7.15g ±0.31
T7	7.26P ±0.04	8.50KL ±0.04	8.99I ±0.03	10.24E ±0.03	8.74d ±0.32
T8	6.30R ±0.04	7.58N ±0.04	8.23M ±0.05	9.57G ±0.05	7.92e ±0.35
T9	9.82F ±0.04	9.23H ±0.05	10.31E ±0.04	12.17B ±0.04	10.37b ±0.33
Range	6.93d ±0.41	7.65c ±0.30	8.31b ±0.32	9.72a ±0.39	

The averages that bear different letters, differ significantly (P<0.01) among them (lowercase letters are main effect of transactions and storage times and large letters are the effect of the intraction between treatments and storage times)

The results of the interaction between treatments and the periods of cold storage of ash percentage are shown in table (4). All treatments showed a significant (p<0.01) decrease in ash percentage with the increasing of storage time (0, 4, 8 and 12 days), nisin and salts treatments showed a significant (p<0.01) decrease in ash percentage when compared with control treatment. Lowest percentage of ash (1.06%) was recorded in T3 in the first storage period (zero days of chilling) when compared with the control treatment which showed the highest percentage of ash percentage (1.73%) in the last storage period (12 days of chilling) For the storage periods, the results of the statistical analysis indicated significant differences (P<0.01) in the percentage of ash between the rates of the storage periods, where the highest percentage was in the 12-day storage period, then the 8-day period, followed by the 4-day period, and it was the lowest in the period 0 days, and this is due to the decrease in moisture with the increase of the cold storage periods, which leads to an increase in the dry matter concentration (7).

Table 4. Effect of nisin, potassium sorbate and sodium lactate on ash of chilled ground beef meat

Treatments No.	Ash Storage periods (days)				Range
	0	4	8	12	
T1	1.14IJK ±0.05	1.45CDEF ±0.06	1.55ABCD ±0.02	1.73A ±0.05	1.46a ±0.07
T2	1.10JK ±0.04	1.22HIJK ±0.06	1.35EFGH ±0.07	1.56ABCD ±0.08	1.30d ±0.06
T3	1.06K ±0.05	1.15IJK ±0.06	1.25GHIJK ±0.05	1.40DEFGH ±0.05	1.21e ±0.04
T4	1.11JK ±0.05	1.40DEFGH ±0.09	1.55ABCD ±0.10	1.70AB ±0.03	1.44ab ±0.07
T5	1.10JK ±0.04	1.31FGHI ±0.06	1.40DEFGH ±0.05	1.52BCDE ±0.05	1.33cd ±0.05
T6	1.11JK ±0.05	1.25GHIJK ±0.06	1.45CDEF ±0.03	1.60ABC ±0.04	1.35bcd ±0.06
T7	1.12JK ±0.04	1.35EFGH ±0.06	1.43CDEFG ±0.02	1.57ABCD ±0.05	1.36bcd ±0.05
T8	1.15IJK ±0.06	1.26FGHIJ ±0.10	1.42CDEFG ±0.03	1.65AB ±0.05	1.37bcd ±0.06
T9	1.10JK ±0.07	1.39DEFGH ±0.06	1.45CDEF ±0.04	1.71A ±0.05	1.41abc ±0.07
Range	1.11d ±0.01	1.30c ±0.02	1.42b ±0.02	1.60a ±0.02	

The averages that bear different letters, differ significantly (P<0.01) among them (lowercase letters are main effect of transactions and storage times and large letters are the effect of the intraction between treatments and storage times).

Table (5) shows the effect of the interaction between different treatments and cold storage periods on the water holding capacity (%) of chilled ground beef, where a significant increase (P<0.01) was noted in the meat water holding capacity in T4 (sodium lactate 3%). which amounted to 69.58%, which recorded the highest significant difference as compared to the control treatment, which amounted to 57.45% in the first period (0 days), while the control treatment recorded the lowest significant difference of 30.76% in the period 12 days, and there were significant differences between the treatments and different storage periods. It is noted from the same table that there are significant differences (P<0.01) between the rates of the addition treatments, as it was noted a significant increase in T8 (nisin 150 mg/kg + sodium lactate 3% + potassium sorbate 0.075 g/kg), which amounted to 50.73% as compared to with the control treatment (43.06%), followed by the T7, then T5, T4, T3, T6, T2, and finally T9 in comparison with the control treatment. Determining the level of the meat ability to hold water gives a perception of the degree of protein denaturation in the meat and thus determining the quality of meat (32), and these results agreed with those of (21) and (35, 37). The results of the statistical analysis indicated that there were significant (P<0.01) differences between the periods, as the ability of meat water holding capacity was at its highest level in the first period (0 days), reaching (63.26%) and then began to decrease with the passage of the cold storage period. Until it reached its lowest percentage in the 12-day period of cryopreservation, which amounted to (34.89%), the water holding capacity is affected by the contraction that occurs in muscle tissue during the progression of the cryopreservation periods, where the water holding capacity is decreases and the moisture lost is large, and this leads to a rise in the drip loss rate (6), as well as the reason may be due to the protein losing its ability to store water (protein buffering capacity) as the increase of the distance from isoelectric point (27) and the increase in moisture loss during cryopreservation (22).

Table 5. Effect of nisin, potassium sorbate and sodium lactate on water holding capacity of chilled ground beef meat

Treatments	WHC (%) Storage periods (days)				Range
	0	4	8	12	
T1	57.45H ±0.04	46.18Q ±0.04	37.86Y ±0.06	30.76F ±0.05	43.06i ±2.99
T2	62.75F ±0.05	50.69O ±0.05	38.49W ±0.04	34.45D ±0.04	46.59g ±3.34
T3	64.10D ±0.05	52.73L ±0.04	40.66U ±0.04	35.80AB ±0.04	48.32e ±3.31
T4	69.58A ±0.06	51.20N ±0.05	38.92V ±0.04	34.90C ±0.04	48.65d ±4.06
T5	64.33C ±0.06	53.64K ±0.04	41.84T ±0.04	35.97A ±0.03	48.94c ±3.29
T6	63.90E ±0.05	51.40M ±0.05	39.11V ±0.05	35.64B ±0.04	47.51f ±3.35
T7	64.50C ±0.29	53.85J ±0.04	42.61S ±0.08	36.92Z ±0.04	49.47b ±3.19
T8	64.72B ±0.06	55.76I ±0.06	44.25R ±0.04	38.22X ±0.04	50.73a ±3.08
T9	58.06G ±0.07	46.52P ±0.05	38.11X ±0.06	31.39E ±0.04	43.52h ±3.00
Range	63.26a ±0.67	51.33b ±0.59	40.20c ±0.41	34.89d ±0.44	

The averages that bear different letters, differ significantly (P<0.01) among them (lowercase letters are main effect of transactions and storage times and large letters are the effect of the intraction between treatments and storage times).

Table (6) shows the effect of the interaction between treatments and the cold storage periods on the value of thiobarbituric acid (TBA) for chilled ground beef, It was noticed a significant (P<0.01) increase in the value of TBA (2.25 mg MDA/kg) for T1 (control

treatment without addition) during the storage period of 12 days of chilling as compared with T8 (nisin 150 mg/kg + sodium lactate 3% + potassium sorbate 0.075 g/kg), which recorded the lowest significant (P<0.01) difference (0.026 mg MDA/kg) in the period of storage of 0 days, and there were significant (P<0.01) differences between treatments and for different storage periods. It is noted from the same table that there are significant (P<0.01) differences between the rates of the addition treatments, as it was noted a significant superiority in the value of T1 (1.68 mg MDA/kg), followed by the effect of the T2, which amounted to (0.98 mg MDA/kg), which did not differ significantly with T9 which amounted to (0.95 mg MDA/kg), followed by T4, T6, T5, T3, and T7, which were similar in effect to treatment T8, and these results agreed with the results of researcher (21) and (29), where his TBA values decreased after fish was treated with sodium lactate salts after comparing the results of the treatments with the control treatment and for all periods of the experiment. Also, these results were in agreement with the results of (4) after the grounded beef was treated with nisin, sodium lactate and sodium triphosphate, where the TBA values decreased for all experiment treatments and all cryopreservation periods in comparison with the control treatment, the reason for this is may be the addition of natural antioxidants reduces the proportion of malondialdehyde (MDA) in fats to beef stored in cold storage for 11 days of storage (15). As for the periods of storage, the results of the statistical analysis indicated a significant (P<0.01) difference between the different periods, as the value of TBA was at its lowest level in the 0-day cold storage period, which amounted to (0.12 mg MDA/kg), then it began to rise with the increase of the storage periods till it reached its highest value during the 12-day refrigeration storage period (1.76 mg MDA/kg), This is due to the increase in the concentration of malondialdehyde, which is a secondary product of lipid oxidation and the breakdown of peroxides, and an increase in its production occurs with the progression of storage periods (11), and it is noted that all addition treatments did not exceed the upper limit allowed in TBA values of 2 mg (malondialdehyde / kg meat) (14,16,17).

Table 6. Effect of nisin, potassium sorbate and sodium lactate on Thiobarbituric Acid (TBA) Value of chilled ground beef meat

Treatments No.	TBA (mg MDA/kg) Storage periods (days)				Range
	0	4	8	12	
T1	0.75LM ±0.03	1.67EF ±0.04	2.05B ±0.05	2.25A ±0.02	1.68a ±0.17
T2	0.08Q ±0.005	0.85KL ±0.03	1.15HI ±0.07	1.86C ±0.03	0.98b ±0.19
T3	0.056Q ±0.002	0.60NO ±0.04	0.95JK ±0.04	1.65F ±0.06	0.81f ±0.17
T4	0.070Q ±0.004	0.70MN ±0.04	1.05IJ ±0.05	1.79CDE ±0.04	0.90cd ±0.18
T5	0.045Q ±0.005	0.61NO ±0.03	0.96JK ±0.05	1.70DEF ±0.03	0.82ef ±0.18
T6	0.055Q ±0.004	0.60NO ±0.04	1.05IJ ±0.07	1.82CD ±0.04	0.88de ±0.20
T7	0.034Q ±0.003	0.55O ±0.02	0.90K ±0.05	1.52G ±0.04	0.75g ±0.16
T8	0.026Q ±0.003	0.42P ±0.04	0.87KL ±0.06	1.45G ±0.02	0.69g ±0.16
T9	0.042Q ±0.003	0.75LM ±0.06	1.23H ±0.02	1.80CDE ±0.09	0.95bc ±0.19
Range	0.12d ±0.04	0.75c ±0.07	1.13b ±0.07	1.76a ±0.04	

The averages that bear different letters, differ significantly (P<0.01) among them (lowercase letters are main effect of transactions and storage times and large letters are the effect of the intraction between treatments and storage times).

Figure (1) shows the effect of the interaction between treatments and storage periods on the total number of bacteria in chilled ground beef. It is noted that the logarithm of the total number of bacteria decreased significantly (P<0.01) in T6 (nisin 150 mg/kg + 3% sodium lactate), which amounted to 0.75 colony-forming units/gm of meat in the 0-day of cold storage, while the logarithm of the total number of bacteria increased in the T1 (control) in the 12-day of cold storage, as it recorded 9.11 colony-forming units/gm of meat, and significant (P<0.01) differences between treatments and for different storage periods. Table (7) shows a significant effect (P<0.01) of the treatment on the total number of bacteria, as the number of bacteria decreased in T6, T2, T4, T5, T7, T8, T3 and T9, respectively, as compared with control treatment. In a study conducted by (33), it was concluded that potassium sorbate inhibits Gram-negative bacteria more effectively than Gram-positive bacteria, due to the fact that nisin has a narrow spectrum of effect on bacteria in meat, as it only affects Gram-positive bacteria. As well as the study conducted by (24) to compare the effect of nisin, potassium sorbate and sodium lactate on the growth of *Staphylococcus aureus* and the production of Staphylococcus enterotoxins (SEA), which concluded that sodium lactate was the best among the treatments. These results were in agreement with what was stated (5). This may be due to the fact that T6 (nisin 150 mg/kg + 3% sodium lactate) treatment is the best in inhibiting the total number of bacteria, psychrophilic bacteria and coliform bacteria. As for the periods of cold storage, table (7) shows a significant increase (P<0.01) in the logarithm of the total number of bacteria in the period 12, 8 and 4 days as compared to the period 0 days of cold storage and this results came in agreement with those of (10, 13, 25, 34, 36). It is noted from the results that the total number of bacteria for all addition treatments was within permissible limits for the total count of bacteria in meat by the Iraqi Central Organization for Standardization and Quality Control (7 logarithmic cycles) (16).

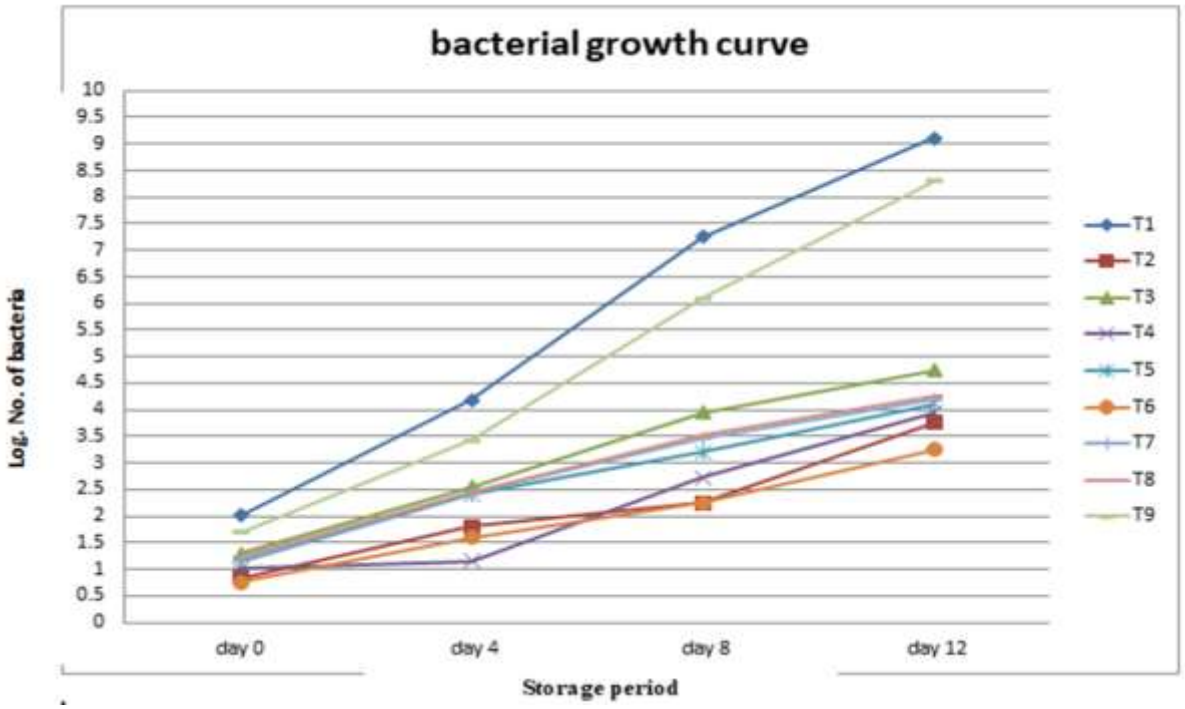

Figure 1. effect of the interaction between treatments and storage periods on the logarithm of the total bacterial count (colony-forming unit/ g of meat) of chilled ground beef meat stored in 4˚C

Table 7. Effect of nisin, potassium sorbate and sodium lactate on Total Bacterial Count (TBC) of chilled ground beef meat

Treatments No.	TBC (colony-forming units/gm of meat) Storage periods (days)				Range
	0	4	8	12	
T1	2.01O ±0.02	4.18F ±0.04	7.25C ±0.14	9.11A ±0.05	5.63a ±0.82
T2	0.83ST ±0.01	1.80P ±0.10	2.52KL ±0.01	3.75H ±0.13	2.22g ±0.32
T3	1.30Q ±0.05	2.55KL ±0.02	3.95GH ±0.02	4.75E ±0.13	3.13c ±0.39
T4	1.01RS ±0.04	2.15NO ±0.02	2.73K ±0.02	3.95GH ±0.02	2.46f ±0.31
T5	1.13QR ±0.06	2.40LM ±0.05	3.20J ±0.10	4.10FG ±0.05	2.70e ±0.33
T6	0.75T ±0.02	1.60P ±0.06	2.25MN ±0.13	3.52I ±0.01	2.03h ±0.30
T7	1.18QR ±0.04	2.45LM ±0.02	3.45I ±0.05	4.20F ±0.10	2.82d ±0.34
T8	1.23QR ±0.03	2.46LM ±0.03	3.52I ±0.01	4.25F ±0.13	2.86d ±0.34
T9	1.70P ±0.09	3.44I ±0.03	6.10D ±0.05	8.30B ±0.16	4.88b ±0.76
Range	1.23d ±0.07	2.55c ±0.14	3.88b ±0.31	5.10a ±0.38	

The averages that bear different letters, differ significantly (P<0.01) among them (lowercase letters are main effect of transactions and storage times and large letters are the effect of the intraction between treatments and storage times).

Conclusion

We conclude that the addition of salts (potassium sorbate and sodium lactate) with nisin could improve the chemical and physical characteristics of the chilled ground beef meat, and extend the microbial stability of the ground beef stored under refrigeration at 4˚C for about 12 days.

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