MICROBIOLOGICAL QUALITY OF FRIED CHICKEN BREAST STRIPS MANUFACTURED WITH MONOSODIUM GLUTAMATE SUBSTITUTES

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ABSTRACT: The impact of replacement monosodium glutamate (MSG) with a ratio of 1:1 mixture of salt and sugar on the microbiological quality of deep fat fried chicken breast strips throughout frozen storage (-18°C) for 90 days was assessed. Also, the antibacterial activity of monosodium glutamate and a mixture of salt (sodium chloride) and sugar can (sucrose) in a ratio of 1:1 was determined by disc diffusion assay, the minimum bactericidal concentration (MBC) and the minimum inhibitory concentration (MIC). Results showed that Gram-negative bacteria since the mean values of the inhibition zones were in the ranges of 11.8, 13.4, 15.4, and 16 mm when studying the effect of monosodium glutamate on Escherichia coli, Serratia marcescens, Bacillus cereus, and Staphylococcus aureus, respectively. Also, the mean values of the inhibition zones were in the ranges of 14.2, 15.6, 18.2, and 17 mm when studying the effect of the mix of sugar and salt in a ratio of 1:1 on Escherichia coli, Bacillus cereus, Serratia marcescens, and Staphylococcus aureus, respectively. The average MIC and MBC values of monosodium glutamate against Gram-negative bacteria were 37.5 and 75 µg/ml, respectively, while these values were 32.5 and 65 µg/ml against Gram-positive bacteria, respectively. Also, the average MIC and MBC values of the mix of sugar and salt in a ratio of 1:1 against Gram-negative bacteria were 27.2 and 55 µg/ml, respectively. While these values were 22.5 and 45 µg/ml, respectively against Gram-positive bacteria, respectively. Besides, the results demonstrated that the control of chicken strips possessed the uppermost values of total bacterial count and the least values of total coliform count compared to the remaining treatments. Salmonella and Escherichia coli were not found in both treatments up until the ending of the storage period. The development of chicken strips possessed the least counts of total Staphylococcus aureus, total psychrophilic bacteria, and yeast and mold counts compared to the remaining treatments.

Key words: Chicken breast strips, monosodium glutamate, antibacterial activity, microbiological quality.

INTRODUCTION

There is a tendency for many major manufacturers to move away from the use of artificial flavors in their products. One of these ingredients is monosodium glutamate, as it is controversial, and glutamate is one of the components that various companies have dedicated to removing from food products (Nguyen et al., 2020). Monosodium glutamate (MSG) is a flavor enhancer regularly combined with food products as chicken to improve palatability. Its notable impacts on sensory appeal have been demonstrated in several reports (Barylko-Pikielna et al., 2007; Miyaki et al., 2016). Elimination of this component is very possible to cause diminished consumer satisfactoriness.

The recent trend now is to use MSG alternatives to offset the flavor loss resulting from eliminating glutamate. The flavor improvement impact of MSG is primarily from glutamate which provides a savory taste sensation or umami. Additionally, there is numerous other umami eliciting ingredients as 5’-ribonucleotides and aspartate. Amongst nucleotides, guanylate (GMP) and inosinate (IMP) considerably contribute to taste enhancement and flavor (Wang et al., 2019). In theory, ingredients that are naturally fruitful in

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umami elements can be added instead of MSG in processed food products. The consumers desired natural extracts as tomato extract, mushroom extract, yeast extract, and as MSG alternatives in chicken processed products (Wang and Adhikari, 2018).

Similarly, sugars may provide umami taste characters in the glutamate glycoconjugates form (Hui et al., 2010). Moreover, potassium salts are responsible to improve umami taste intensity. Otherwise, through the boiling process, substantial amounts of potassium leach out from potatoes (Bethke and Jansky, 2008; Wijayasekara and Wansapala, 2017). Sodium chloride is a valuable component added to various food products which provides flavor improvement and food conservation (Chun et al., 2014).

At present, there is limited research assessing the improvement impacts of MSG compared to the natural extracts in processed food products. Taking into account the ability of salty taste improvement, the MSG alternative can additionally be capable to improve the sensory appeal of meat products with decreased salt content. Earlier studies revealed that utilizing yeast extract effectively improved the fermented sausage taste (Campagnol et al., 2011). Likewise, the mushroom has been demonstrated to enhance the flavor of taco blends (Myrdal Miller et al., 2014). To substitute MSG, it is essential to demonstrate more scientific investigation to assess the performance of MSG and its substitutes in a salt-reduced food matrix. The objective of the present study was to assess the impact of a substitute of MSG with a mixture of salt and sugar at a ratio of 1:1 on the microbiological characteristic of deep fat fried chicken strips throughout frozen storage (-18°C).

**MATERIALS AND METHODS**

**Materials**

Fresh and chilled chicken breast slices without skin and bones were brought from Cairo Poultry Slaughterhouse Company, 10th of Ramadan City, Sharkia Governorate and transported under refrigerated conditions in an ice box containing a pack of ice (blue ice) to keep the temperature at 3±1°C and were kept in the refrigerator at the same temperature for 8 hours. The samples were moved under cool conditions to the Food Technology Department Laboratory belonging to Zagazig University, Egypt, and stored in the deep freezer at -18°C for three months up until processing.

Salt, phosphate, monosodium glutamate, a concentration greater than 90%, and spices were obtained from Cairo Poultry Slaughterhouses Company, Tenth of Ramadan City, Sharkia Governorate.

**Marinade Formula of Chicken Strips**

The marinade formula of chicken strips is shown in Table 1.

**Coating Formula of Chicken Strips**

The coating formula of chicken strips is shown in Table 2.

**Methods**

**Preparation of Chicken Strips**

After the preparation of chicken strips as depicted (Tables 1 and 2), samples were separated into two groups: the control group comprising MSG (C) and the remaining mixtures consisting of sugar and table salt by a ratio of 1:1 as MSG substitution (T).

**Preparation of Marinade Solution**

The amount of water under 5°C was put in a bag of polyethylene with high-density, then the quantity of food-grade sodium tripolyphosphate (STPP) was dissolved therein, followed up by resolving the MSG and salt in the case of MSG replacement or the control (a combination of sugar and salt in ratio of 1:1), in the treatment case and then the antioxidant and spices were added and mixed well to regulate the marinade solution. The specified quantity of raw chicken fillet strips was allowed to thaw in the refrigerator for 24 h and added to the previous brine solution. Ultimately, the bags were locked and flipped for 5 minutes and put in the refrigerator at a temperature of 3±1°C.

After 24 hours the bags were opened and the chicken strips were eliminated from the solution and placed on a stainless steel screen for five minutes to filter the extra brine solution, afterward the weight rise of chicken strips attained from the marinade solution was estimated as outlined in the formula of Sampaio et al. (2012).

marinade uptake % = marinated weight – raw weight/raw weight ×100.
### Table 1. Marinade formula of chicken strips

| Components                        | MSG substitution (g) | Control (g) |
|-----------------------------------|----------------------|-------------|
| Raw chicken strips                | 1800                 | 1800        |
| Sodium tripolyphosphate           | 11.25                | 11.25       |
| Potable water                     | 360                  | 360         |
| *monosodium glutamate substitution| 11.25                | ------------|
| Monosodium glutamate Purity ( > 90%) | ------------         | 11.25       |
| Spices                            | 22.95                | 22.95       |
| Salt                              | 15                   | 15          |
| TBHQ antioxidant                  | 2.25                 | 2.25        |

*Monosodium glutamate substitution: mixes comprise table salt (sodium chloride) and sugar (sucrose) by a ratio of 1:1.

Spices (garlic powder 9 g, onion powder 9 g, ginger powder 2.7 g, celery powder 2.25 g).

### Table 2. Coating formula of chicken strips

| Components   | MSG substitution (g) | Control (g) |
|--------------|----------------------|-------------|
| Predust      |                      |             |
| Wheat flour  | 1000                 | 1000        |
| Corn starch  | 259.74               | 259.74      |
| Table salt   | 38.96                | 38.96       |
| Batter       |                      |             |
| Salt         | 7.90                 | 7.90        |
| Wheat flour  | 400                  | 400         |
| * Monosodium glutamate substitution | 17.28            | ------------|
| Monosodium glutamate Purity (>90%) | ------------     | 17.28       |
| Corn starch  | 49.38                | 49.38       |
| Spices**     | 6.89                 | 6.89        |

* Monosodium glutamate substitution mix comprises sugar and table salt by a ratio of 1:1.

** Batter spices comprise ginger powder 1.97 g, garlic powder 2.46 g, and black pepper powder 2.46 g.

Breading

| Components        | MSG substitution (g) | Control (g) |
|-------------------|----------------------|-------------|
| Wheat flour       | 1000                 | 1000        |
| Table salt        | 25.4                 | 25.4        |
| Corn starch       | 200                  | 200         |
| Sodium bicarbonate| 14                   | 14          |
Coating Stage

Predust stage

In the control, the components referred to in Table 2 were placed in a polyethylene bag containing 1000 gm wheat flour, 259.74 gm corn starch, and 38.96 g salt, and they were mixed well and the same ingredients were prepared in the same proportions in the case of the remaining treatments.

Batter stage

In the control, the components indicated in Table 2 were placed in a polyethylene bag containing 400 gm of wheat flour, 49.38 gm of corn starch, 7.90 g of salt, and 6.89 g of spices and 17.28 gm of monosodium glutamate. They were mixed well and the same ingredients were prepared in the same proportions in the case of treatment by replacing the amount of monosodium glutamate with a mix of sugar and salt, the ratio of 1:1. Then the previous ingredients were mixed with cooled water (3±1°C) in a ratio of 1 water to 1 of the previous dry ingredients in both the treatment and the control.

Breading stage

In the control, the components referred to in Table 2 were placed in a polyethylene bag containing 1000 g of wheat flour, 200 gm of corn starch, 25.4 g of salt and 14 g of sodium bicarbonate and they were mixed well and the same ingredients were prepared in the same proportions in the case of the remaining treatments. The chicken strips were separated from the marinade solution and placed on a stainless steel screen for 5 minutes to get rid of the excess of the marinade solution, and then they were placed in the bag containing the predust powder. The bag was closed and shaken for 10 seconds. The chicken breast pieces were separated from the bag and placed in a bag containing the batter solution then shacked for 5 seconds, then taken out from the batter bag and placed on a stainless steel screen for 5 seconds to get rid of the excess of the batter solution. Chicken strips were put in a bag containing the breading. The bag was closed and shaken for 10 seconds. The chicken breast pieces were separated from the bag and put in the frying oil mixture at 187°C in batches of 4 pieces each time. The pieces were removed from the oil when the temperature of the thick part of the pieces reached 74 ± 2°C using a thermocouple (Testo 0560-1110, Germany) and put on a stainless screen to drain the excess oil for the second treatment fresh oil (187°C) was used for frying. Samples were withdrawn from both treatments to conduct microbiological analyses at zero time. The fried chicken strips (control and treatment) were stored in the freezer at -18°C for 90 days. Samples were withdrawn after 30, 60, and 90 days for the microbiological analyses.

Deep-frying of marinade chicken strips

One and half litres of a mix of soybean and sunflower oil 1: 1 were put in an electric fryer and the temperature of the oil was elevated to 186-188°C, then the breaded chicken breast slices and marinated were put in the oil at a rate of four pieces every-time and the weight of each piece was around 40 g. While the temperature in the middle of the chicken breasts changed to 74-76°C, using a calibrated thermometer (Testo 0560-1110, Germany), they were removed from the excess oil. Before frying the treatment sample, fresh oil was used. Samples were preserved in a deep freezer at -18°C up until the test completion (Park and Kim, 2016).

Antibacterial Activity of Mono-Sodium Glutamate and Mixture of Salt and Sugar Against Pathogenic Bacteria

Determination of Disc diffusion assay

Fresh medium (nutrient broth, NB) was employed in all tests to recover the bacteria by sub-culturing. A slight portion of the inoculum of each bacterium was combined in 5 ml of nutrient soup and preserved during the night at 37°C. The pathogenic bacteria namely, *Escherichia coli*, *Bacillus cereus*, *Serratia marcescens*, and *Staphylococcus aureus* were obtained from the Agricultural Microbiology Department belonging to the Faculty of Agriculture, Zagazig University, Egypt. The bacteria were cultured on nutrient agar (NA) plates and preserved in the nutrient agar slants at 4°C, during the night. Cultures in the nutrient soup were utilized for the laboratory study. The antibacterial activity of C and T was determined by employing the method of disc diffusion as
outlined by National Committee for Clinical Laboratory Standards (NCCLS) (2003). The recording of zone of inhibition was determined following the incubation at 30°C or 37°C for a day. Bactericidal impacts of C and T were studied by the method that was modified and shown by Langfield et al., (2004). The diluted bacterial culture (0.1 ml) was expanded on a sterile NA plate. The discs were dried out of 6 mm diameter of Whatman filter paper No: 1 earlier drenched in (C and T) suspension (10, 20, 30, 40, and 50µg) were put on the seeded plates against Gram-positive bacteria and Gram-negative.

**Determination of MIC and MBC**

Luria broth was utilized in a micro dilution procedure for estimating MBC and MIC. An inoculant of 10 ml (2.5x10^8 CFU/ml) from every particular bacterial culture was incorporated into 1 mL of (NB). Every tube containing a test strain was accompanied by one of each concentration of (C and T). The determination of MIC was achieved by estimating the turbidity of the bacterial progression after incubation for a day. The impeded concentration was 99 percent of bacterial growth is deemed the MIC (Dash et al., 2012). In accordance with the basic method, the MBC values of the particles were estimated by subculturing the MIC dilutions onto the sterile Muller Hinton agar plates incubated at 30°C or 37°C for 24h. The lowest concentration of (C and T) was recorded. The value of MBC is equal to the concentration where 100% of the bacterial progression was halted and associated with the positive control which implies no treatment. The culture of bacteria with no silver nitrate was applied as a control. The whole solution volume utilized in each flask was 50 ml.

**Microbiological examination**

The aerobic plate count (APC) and psychrophilic bacteria count were applied as outlined in APHA (1992). Potato dextrose agar was employed for mold and yeast recording. The plates were incubated at 25°C for five days. The violet red bile agar was utilized for the account of coliforms. The plates were incubated at 37°C for 24 h, following APHA (1992).

*Staphylococcus aureus* was applied according to ISO, 4833-1 (2013). *Salmonella* spp was applied according to ISO, 6579 (2004).

**Statistical Analysis**

The analysis of variance (ANOVA) was applied by employing SAS software (SAS institute, 1998). The averages were significantly separated by the least significant differences (LSD) at p < 0.05. All tests were performed in three triplicates.

**RESULTS AND DISCUSSION**

**Antibacterial Activity of Mono-Sodium Glutamate and Mixture of Salt and Sugar**

**Disc diffusion assay**

The treatment group emerges as a replacement of antibacterial agents and can overcome the bacterial resistance to the antibiotics. Consequently, it is essential to broaden the application of T as an antibacterial agent. (C and T) were assessed against two ram–positive bacteria (*Bacillus cereus, Staphylococcus aureus*) and two Gram–negative bacteria (*Serratia marcescens, Escherichia coli*) and utilizing disc diffusion procedure to estimate the activity of antibacterial of four different concentrations of (C and T) as shown in Table 3. An average of inhibition zones of three replications was recorded. From this table, it can be noticed that the superior concentration of C and T the wider the inhibition zone. This was true with all pathogenic bacteria species tested. In addition, Gram positive bacteria tested in this investigation were more susceptible to C and T than that of Gram- negative bacteria since the mean values of the inhibition zones were in the ranges of 11.8, 15.4 16, and 13.4 mm when studying the effect of C on *Escherichia coli, Bacillus cereus, Staphylococcus aureus*, and *Serratia marcescens* respectively. Also, the mean values of the inhibition zones were in the ranges of 14.2, 17, 15.6 and 18.2 mm when studying the effect of T on *Escherichia coli, Bacillus cereus, Staphylococcus aureus*, and *Serratia marcescens* in the same order. These results are in line with those of Bhuvaneswari et al. (2015) for monosodium glutamate and Shee et al. (2010) for salt and sugar.
Determination of MBC and MIC

The minimum bactericidal concentration (MBC) and the minimum inhibitory concentration (MIC) of C and T were determined because both were simple to evaluate despite diverse concentration units such as µg/ml, mg/l or ppm and provide precise information respecting the microorganism susceptibility.

MICs of C and T were estimated by standard microdilution procedure against two Gram positive (G⁺) and two Gram-negative (G⁻) bacterial strains.

Data presented in Table 4, show that the average MBC and MIC values of (C) against Gram negative bacteria were 37.5 and 75µg/ml, respectively. While these values were 32.5 and 65 µg/ml, in the same order against Gram positive bacteria. Also, average MIC and MBC values of T against Gram negative bacteria were 27.2 and 55 µg/ml, in the same order. While these values were 22.5 and 45 µg/ml, respectively against Gram-positive bacteria. These results are in line with those of Bhuvaneswari et al (2015) for monosodium glutamate and Shee et al (2010) for salt and sugar.

Microbiological Properties of Raw Chicken Strips

Total aerobic bacterial, Escherichia coli, coliform, Staph. Aureus, Salmonella, psychrophilic bacteria and mold and yeast counts of raw chicken strips were shown in Table 5. Results showed that Total aerobic bacterial, Escherichia coli, Staph. Aureus, coliform, Salmonella, psychrophilic bacteria, and mold and yeast counts were 2.6×10², ND, 6.7×10¹, 7.2×10¹, ND, 3.3×10⁶ and 6.4×10¹ cfu/gm, in the same order. These findings are in consonance with the results of Eglezo et al. (2008), Al-Nehlawi et al. (2013) and Rouger et al. (2017).

Microbiological Tests of Deep Fat-Fried Chicken Strips Throughout Frozen Storage (-18°C)

The microbiological tests were assessed to estimate the microbiological shelf life validity and quality during frozen storage. Microbial progress in meat and meat products be able to cause by structural component degradation, slime formation, off odors, reduction in water holding ability, and appearance and texture adjustments which decrease their nutritional rate, shelf life and quality (Doulgeraki et al., 2012).

Table 6 indicates that there were substantial variations in APC between the control of chicken strips and other chicken strips samples. These results implied that APC declined gradually during the storage period up until the ending of the storage period. Moreover, the finding demonstrated that control of chicken strips possessed the uppermost APC compared to the remaining treatments. This could be owing to the antimicrobial activity of sugar or salt.
Table 4. The minimum bactericidal concentration (MBC) and minimum inhibitory concentration (MIC) values of (C and T) against pathogenic bacteria species.

| Determination | C (µg/ml) | T (µg/ml) |
|---------------|-----------|-----------|
| Pathogenic bacteria | MBC | MIC | MBC | MIC |
| Gram-negative pathogenic bacteria | | | | |
| *E. coli* | 80 | 40 | 60 | 30 |
| *Serratia marcescens* | 70 | 35 | 50 | 25 |
| Gram-positive pathogenic bacteria | | | | |
| *Bacillus cereus* | 70 | 35 | 50 | 25 |
| *Staphylococcus aureus* | 60 | 30 | 40 | 20 |

Table 5. Microbiological analysis of raw chicken strips.

| Count (cfu/gm) | Raw chicken strips |
|----------------|--------------------|
| Total aerobic bacterial | 2.6×10^5 |
| *Escherichia coli* | ND |
| Total coliform | 7.2×10^1 |
| *Staph. aureus* | 6.7×10^1 |
| Salmonella | ND |
| Psychrophilic bacteria | 3.3×10^6 |
| Mold and yeast | 6.4×10^1 |

ND: Not detected

Table 6. Impact of substituting MSG with a mixture of salt and sugar in a ratio of 1:1 on the microbiological quality of deep fat fried chicken strips throughout frozen storage (-18°C).

| Viable count (cfu/g) | Storage period (day) |
|----------------------|----------------------|
|                      | Zero | 30  | 60  | 90  |
| Total Bacterial count (T B C) | | | | |
| C                     | 2.95×10^1 | <10 | <10 | <10 |
| T                     | 4.59×10^1 | <10 | <10 | <10 |
| Total coliform count (T C C) | | | | |
| C                     | 0.51×10^1 | <10 | <10 | <10 |
| T                     | 0.80×10^1 | 0.59×10^1 | 0.42×10^1 | <10 |
| Salmonella | | | | |
| C                     | Not-D | Not-D | Not-D | Not-D |
| T                     | Not-D | Not-D | Not-D | Not-D |
| *E. coli* | | | | |
| C                     | Not-D | Not-D | Not-D | Not-D |
| T                     | Not-D | Not-D | Not-D | Not-D |
| *Staph aureus* | | | | |
| C                     | 0.57×10^1 | <10 | <10 | <10 |
| T                     | 0.72×10^1 | 0.50×10^1 | 0.30×10^1 | <10 |
| Psychrophilic bacteria | | | | |
| C                     | 3.74×10^2 | 1.34×10^4 | 1.14×10^4 | 0.51×10^4 |
| T                     | 3.69×10^3 | 2.52×10^4 | 2.46×10^4 | 2.40×10^4 |
| Yeast and mold | | | | |
| C                     | 2.51×10^1 | >10 | >10 | >10 |
| T                     | 6.13×10^1 | >10 | >10 | >10 |
Similar findings were demonstrated by Aksu and Alp (2012), Malay et al. (2013) Hwang et al. (2011), Prejsnar et al. (2018) and Bouacida et al. (2020).

Table 6 presents the variations in coliform counts. The findings revealed that the total coliform count declined steadily during the storage period up until the ending of the storage period. Besides, the results indicated that the control of chicken strips possessed the least counts of the total coliform count compared to the remaining treatments. Similar findings were disclosed by Hwang et al. (2011), Prejsnar et al. (2018) and Bouacida et al. (2020).

The displayed results in Table 6 revealed that Escherichia coli count was not detected in both treatments up until the ending of the storage period. Similar findings were demonstrated by Hwang et al. (2011), Prejsnar et al. (2018) and Bouacida et al. (2020).

The results described in Table 6 revealed that Salmonella was not detected in both treatments up until the ending of the storage period. Similar findings were demonstrated by Hwang et al. (2011), Prejsnar et al. (2018) and Bouacida et al. (2020).

Table 6 indicates the variations in Staph coagulase counts. The findings revealed that Staph aureus count declined steadily during the storage period up until the ending of the storage period. Additionally, the results displayed that the control of chicken strips possessed the least counts of total Staph coagulase compared to the remaining treatments. Similar findings were mentioned by Hwang et al. (2011), Prejsnar et al. (2018) and Bouacida et al. (2020).

Table 6 shows the variations in psychrophilic bacteria counts. The findings showed that the total count of psychrophilic bacteria elevated steadily during the storage period up until the ending of the storage period. Besides, the results revealed that the control of chicken strips possessed the least values of total psychrophilic bacteria count compared to the remaining treatments. Similar findings were disclosed by Hwang et al. (2011), Prejsnar et al. (2018) and Bouacida et al. (2020).

The variations in mold and yeast counts of deep fat fried chicken strips throughout the frozen storage are displayed in Table 6. The results revealed that total mold and yeast count declined steadily as the storage period increased up until the ending of the storage period. In addition, the results indicated that the control of chicken strips possessed the least counts of total mold and yeast compared to the remaining treatments. Similar findings were pointed out by Hwang et al. (2011), Prejsnar et al. (2018) and Bouacida et al. (2020).

Conclusion

Monosodium glutamate can be replaced with a mix of 1:1 salt and sugar in the manufacture of chicken strips which gained high microbiological quality.

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