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Potential of Commercial Spice Mixes to Enhance the Quality and to Extend the Shelf Life of Raw Chicken Breasts

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ABSTRACT

Raw chicken harbors spoilage microorganisms such as the Mesophilic Aerobic Bacteria (MAB), Lactic Acid Bacteria (LAB), Spoilage Yeasts (SY) and *Pseudomonas*, which limit product shelf life. This study compared the potential of three spice mixes ("Tandoori", "Kalia" and "Massala") to extend the shelf life of raw chicken. Chicken breasts were marinated with each of the spice mixes (3% w/w), and subsequently refrigerated for up to 15 days. Marinated and non-marinated samples were withdrawn at three-day intervals and analyzed for enumeration of MAB, LAB, SY and *Pseudomonas*. After three days, chicken marinated with "Tandoori" and "Kalia" spices had a significantly ($P<0.05$) lower load of MAB (5.51-6.06 log cfu/g) compared with untreated control breasts (6.58 log cfu/g) although by Day 15, there were no significant differences ($P>0.05$) observed in the final MAB counts between treated samples (7.51-7.88 log cfu/g) and untreated controls (7.88 log cfu/g). There were also no significant ($P>0.05$) differences in the counts of *Pseudomonas* (2.65-3.64 log cfu/g), LAB (2.56-4.20 log cfu/g) and SY (2.60-4.15 log cfu/g) over the 15-day storage. Since the onset of microbial spoilage is marked by MAB reaching 7 log cfu/g, the microbiological shelf-life of marinated and non-marinated chicken breasts were estimated at 12 and 6 days respectively. However, based on the sensorial attributes, both marinated and non-marinated chicken received poor acceptability scores after six and three days respectively. Commercial spice mixes can thus extend the refrigerated shelf-life of raw chicken by three days to a maximum of six days.

Keywords: Breast, Chicken, Quality, Shelf-life, Spice Mixes

INTRODUCTION

The consumption of chicken has increased worldwide because it forms a major part of the human diet. Chicken has nutritional characteristics such as low lipid content and a high concentration of polyunsaturated fatty acids (Bourre, 2005). Chicken meat can however perish rapidly if it is not stored, processed, packaged or distributed correctly (EFSA, 2013). Raw meat can either be spoiled by microbial activity or by oxidative processes due to the high content of PUFA and high peroxidation index (Sebranek et al., 2005; Dal Bosco et al., 2016). Spoilage microorganisms associated with meat, especially lactic acid bacteria can cause undesirable changes in meats. Those changes make the chicken unattractive and unfit for human consumption (Gram et al., 2002; Doulgeraki et al., 2012).

Thus food industries worldwide have resorted to the addition of synthetic preservatives to meat products to prevent the uncontrolled growth of spoilage organisms and to increase their shelf life. In recent years, there has been a significant concern over the safety of these chemicals, thereby influencing consumers' preference for natural products such as spices and plant extracts over chemical preservatives (Govaris et al., 2010).

A spice can be referred to as a seed, fruit, root, bark, berry, bud or vegetable which is used in food to enhance its flavour (aroma and taste), colour or texture as well as to

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preserve the product from deterioration. The use of spices is very important in raw or fresh meat because the latter is mostly susceptible to spoilage (Thomas et al., 2012). Many studies have stated that spices have antioxidant properties by the virtue of their phenolic components (Konczak et al., 2010). Spices with known antimicrobial activity, are cloves, cinnamon and oregano as they contain eugenol, cinnamaldehyde and thymol respectively (Wang et al., 2011). Not only do spices delay the onset of microbial spoilage, but also enhance the safety of food by inhibiting the growth of foodborne pathogenic microorganisms (Devatkal and Naveena, 2010). For instance, Radha Krishnan et al. (2014) showed that spices, containing a high amount of phenolic compounds, decrease lipid oxidation and inhibit the growth of microorganisms, thus increasing the shelf life of poultry (Radha Krishnan et al., 2014). There are many studies, which have either investigated the antimicrobial activities of crude forms of spices (Smith-Palmer et al., 1998; Hara-Kudo et al., 2004; Skrinjar and Nemet, 2009; Aggarwal et al., 2015) or the essential oils of spices (Hammer et al., 1999; Dorman and Deans, 2000) against foodborne pathogens and spoilage microorganisms. As the Mauritian cuisine is diverse and is an amalgamation of European, Chinese and Indian cuisines, a wide variety of spices are used as ingredients and seasoning in Mauritian cooking. Moreover, spices are also used as decoctions in Mauritius (Mahomoodally et al., 2012). However, to our knowledge no studies have been attempted to test the potential of spice mixes, locally available in Mauritius to control the growth of spoilage bacteria in food systems. This study therefore aims at assessing the potential of commercially available spice mixes to enhance the quality and extend the shelf-life of raw chicken breast.

MATERIALS AND METHODS

Sample collection and treatment

A preliminary survey targeting a convenience sample of 50 consumers was conducted in order to identify different spice mixes typically used in households. A questionnaire was designed for that purpose and was administered face to face. The three most frequently used spice mixes identified were “Tandoori”, “Massala” and “Kalia” and chicken breast was identified as the most preferred part of poultry meat used by consumers.

Chicken breasts were bought from a chilled retailed outlet and transported to the laboratory in a cooler bag. The skin was removed and the chicken breasts were cut into cubes, weighing approximately 10g, with dimensions of 2 cm x 2 cm x 2 cm. Chicken samples were then either left untreated (U) or homogenously mixed with “Tandoori” (Tt), “Kalia” (Tk) or “Massala” (Tm) at a final concentration of 3% w/w as determined in the preliminary survey. Samples were subsequently placed in closed plastic containers and were kept at 4^{o}C for up to 15 days.

Microbiological analysis

Marinated and non-marinated samples were withdrawn for microbiological analysis every three days for a period of 15 days. Each sample was aseptically placed in a sterile stomacher bag to which 90 ml of sterile buffered peptone water was added. The mother sample and its dilutions were placed on Plate Count agar medium (OXOID), *Pseudomonas* agar (OXOID), *Lactobacillus* MRS agar (OXOID) and Potato Dextrose agar (PDA) for the enumeration of Mesophilic Aerobic Bacterial (MAB) counts, *Pseudomonas* counts, Lactic acid bacteria (LAB) counts and Spoilage Yeast (SY) counts respectively. The plates for the *Pseudomonas* counts and Lactic acid bacterial counts were incubated at 37^{o}C for 24±2 h and 37^{o}C for 48±2 h respectively. PDA plates were however incubated at room temperature (ca. 24°C) for five days. All the colonies were then enumerated with a colony counter.

Physicochemical analysis

For determination of pH, treated or untreated chicken samples (10 g) were minced and mixed with 90 ml of distilled water for 30 minutes using a magnetic stirrer. The pH of the mixture was then measured using a digital pH meter (Mettler Toledo). For the determination of the water activity of samples, a hand-held water activity meter (Novasina, Japan) was used. For the determination of instrumental surface colour (CIE L*a*b), chicken samples were minced and placed in a clean petri dish which was then inverted. Triplicate measurements of surface colour were then taken using a chromameter (Minolta CR-410, Konica Minolta, Japan). Drip loss of chicken samples was also determined by measuring cumulatively the volume of exudate lost after two hours, one day and 15 days through refrigerated storage. To determine drip loss, 10 g of marinated and non-marinated samples were initially weighed and then placed in a sealed ziplock bag and kept in the refrigerator at 4^{o}C. Their weight was then determined immediately (D_0), one day (D_1) and 15 days (D_{15}) through refrigerated storage. All the physicochemical analyses were carried out in three independent replicates and their measurements were taken in triplicates.

Sensory evaluation

Marinated and non-marinated chicken samples were prepared and stored at chilling temperature as described

previously. A sensory evaluation questionnaire was designed and sensory analysis was conducted with 10 untrained panellists at 3-day intervals for a period of 15 days. On each day of the analysis, samples were taken out and rated for different sensory parameters such as colour, aroma, texture and general appearance on a scale of 1-10, where 1 being the least accepted and 10 being most accepted.

Statistical design and analysis

The statistical analyses were conducted using the General Linear Model in MINITAB version 16.0 to determine the differences for the different treatments on the different days of storage. Significant differences were considered at the 95% confidence level ($P < 0.05$).

Ethical approval

The authors solemnly declare that publication ethics and good conduct were adhered to during preparation, reviewing, processing and proofreading of this article. No ethical clearance was needed to conduct the work.

RESULTS AND DISCUSSION

Consumer use of spices

The most commonly used spice mixes among Mauritians were "Tandoori" (24.0%) followed by "Kalia" (21.9%) and "Massala" (17.7%), as these spices are ingredients used in many traditional Mauritian dishes. As a matter of fact "Tandoori chicken" is one of the favourite dishes of Mauritians and it is of Indian origin (Kioon, 2015). "Chicken Kalia" is considered as one of the most authentic Mauritian recipes and is also widely appreciated by Mauritians (Kioon, 2010). These spice mixes were most preferred by survey participants thanks to the virtue of their unique compositions and flavour. The survey also revealed that the spices were added to chicken at a ratio of approximately 30g to 1 kg of chicken or ca. 3% w/w of chicken.

Microbiological analysis

The MAB counts of chicken samples had increased from an initial population of 5.4 log cfu/g to a maximum of 7.9 log/cfu over the 15-day refrigerated storage (Table 1).

For samples that were marinated with "Tandoori", "Kalia" and "Massala" spice mixes, MAB counts were consistently lower than their untreated counterparts by a maximum of 1.0 log cfu/g. The exact mechanism of antibacterial action of spices and derivatives is not yet clear (Lanciotti et al., 2004), although it has been hypothesized that hydrophobic and hydrogen bonding of phenolic compounds to membrane proteins, partitioning of the lipid bilayer (Juven et al., 1994), perturbation of the permeability of bacterial cell membranes (Cox et al., 1998), membrane disruption (Caccioni et al., 2000), destruction of electron transport systems (Tassou et al., 2000) and cell wall perturbation (Odhav et al., 2002) could play a role. MAB counts of samples treated with "Tandoori", "Kalia" and "Massala" however reached levels of 7.9, 7.6 and 7.7 log cfu/g after 15 days respectively indicating that the spice mixes used in the study had not significantly suppress growth of mesophilic aerobes ($P > 0.05$). On the other hand, other authors have successfully demonstrated the effectiveness of spices and spice mixes to sustainably control spoilage and pathogen growth in food. Shelef (1983) indicated that high levels of spices inhibited growth of spoilage microorganisms in food such as chicken and fish. Grohs and Kunz (2000) tested the effectiveness of spice mixtures to inhibit the growth of various meat-spoilage microorganisms (*Bacillus subtilis*, *Enterococcus* spp., *Staphylococcus* spp., *E. coli* K12 and *Pseudomonas fluorescens*) and to stabilize the colour and smell of fresh-portioned pork meat.

Table 1. Mesophilic aerobic bacterial (MAB) counts (log_{10} cfu/g) of spice-marinated and non-marinated chicken samples over a 15-day storage period

Days	Treatments			
	U	Tt	Tk	Tm
0	5.4±0.2	5.4±0.6	6.0±0.0	5.9±0.1
3	6.6±1.2	5.5±0.5	6.1±0.1	6.7±0.5
6	7.2±0.8	6.3±0.9	7.0±0.2	7.0±0.2
9	7.5±0.8	6.4±0.8	7.1±0.1	7.1±0.2
12	7.7±1.0	7.3±1.5	7.2±0.1	7.2±0.5
15	7.9±1.2	7.9±1.2	7.6±0.4	7.7±0.6

U: Untreated, Tt: "Tandoori", Tk: "Kalia", Tm: "Massala"; Data represent mean values of three replicates ± standard deviation; Counts within the same row representing the same day of storage were not significantly different ($P > 0.05$).

Contrary to our findings, the authors showed that these spice mixtures were effective shelf-life extenders. The disparity in our results could be partly attributed to differences in the variety of spices used, the composition of spice mixes tested, the cultivar of spice vegetables and the marination procedure. Indeed, several scientific reports attributed the differences in the inhibitory effect of spices to variation in the resistance of different microorganisms to a given spice and of the same microorganism to different spices (Akgul and Kivanç, 1988). It is also worth mentioning that there is a considerable body of research on the antibacterial effectiveness of essential oils of a wide range of spices against different spoilage and pathogenic bacteria and their results consistently showed that

individual or combinational extracts exhibited strong antibacterial activity (Arora and Kaur, 1999; Elgayyar et al., 2001; Zhang et al., 2015). The high potency of the extracts noted in these studies could be attributed to the fact that they tested aqueous or alcohol-based extracts of spices and herbs. In our study on the other hand, we used commercial spice blends, which were crude mixtures of different dried spice vegetables.

Shelf-life can be defined as the period in which a product remains safe and suitable for consumption. This means that it has not deteriorated in quality or spoiled in any way that the consumer would find it unacceptable (EFIC, 2013). The limit of acceptability or the onset of spoilage of poultry products is usually marked by mesophilic aerobes attaining populations of 7.0 log cfu/g in fresh poultry (Cox et al., 1998). The microbiological shelf-life of non-marinated chicken was thus estimated to be < 6 days while the shelf-life of chicken marinated with "Tandoori" was estimated to be < 12 days. "Kalia" and "Massala" were not as effective shelf-life extenders as "Tandoori" as the chicken products had a shelf-life of < 6 and < 9 days respectively. Similarly Khanjari et al. (2013) and Kuswandi et al. (2014) also observed that the microbiological shelf-life of untreated chicken was ca. 6-7 days at refrigeration temperature. *Pseudomonas* spp., Lactic acid bacteria and Spoilage yeasts counts on non-marinated chicken increased from an initial density of 2.9, 2.6 and 2.6 log cfu/g to a maximum of 3.2 log cfu/g although the difference between the final and initial density was not significantly different (P > 0.05) (Table 2).

Table 2. The initial and final population density of *Pseudomonas* spp., Lactic acid bacteria (LAB) and Spoilage Yeasts (SY) (log cfu/g) of spice-marinated and non-marinated chicken

Parameter	Days	U	Tt	Tk	Tm
Pseudo.	0	2.9±0.0	2.6±0.8	2.7±0.1	2.8±0.2
	15	3.2±0.1	3.0±0.1	3.0±0.0	2.9±0.0
LAB	0	2.6±0.1	2.6±0.0	2.6±0.7	2.7±0.6
	15	3.2±0.1	3.1±0.4	4.2±0.6	4.2±0.6
SY	0	2.6±0.0	2.4±0.9	3.0±0.5	2.9±0.2
	15	3.2±0.5	3.8±1.1	4.0±0.7	4.2±0.6

U: Untreated, Tt: "Tandoori", Tk: "Kalia", Tm: "Massala"; Data represent mean values of three replicates ± standard deviation; Counts within the same row representing the same day of storage were not significantly different (P>0.05).

For chicken marinated with one of the different spice mixes, *Pseudomonas* spp. had increased to a maximum of 3.0 log cfu/g although these final counts were not significantly different from the final densities of their untreated counterparts (P > 0.05). The *Pseudomonas species* isolated from poultry could likely be *P. fluorescens*, *P. putida* or *P. fragi* (Russell, 2009). *Pseudomonas* spp. has generally been considered to be the predominant Specific Spoilage Organism (SSO) in poultry (Barnes and Impey, 1968; Cerveny et al., 2009). SSO is defined as the part of the total microbiota responsible for spoilage of a given product within the spoilage domain, which is the range of product characteristics and storage conditions within which a given SSO causes product rejection (Dalgaard, 1995). In fact, Davies and Board (1998) indicated that *Pseudomonas* spp. made up approximately 85% of the entire bacterial population on poultry refrigerated for about two weeks and fluorescent and non-pigmented strains of *Pseudomonas* spp were mostly found in spoiled chicken. Jay et al. (2007) and Rukchon et al. (2014) also mentioned that the primary spoilage organism in chicken kept at low temperature reportedly belongs to the genus *Pseudomonas*. However, in the current study, *Pseudomonas* spp. did not appear to be the predominant spoilage bacteria as it had increased only by 0.35 log cfu/g compared to other bacterial species. It is possible that other microbial species that were not enumerated in this study could have been responsible for product spoilage since there are over 25 bacterial genera that make up the microbiota of poultry (Lahellec et al., 1975). Lahellec et al. (1975) indicated that in a study of 5920 isolates from chicken carcasses, pseudomonads were found to constitute only 30.5% of the microbial biota while the rest consisted of *Acinetobacter*, *Flavobacterium* and *Corynebacterium* in relative abundances of 22.7%, 13.9% and 12.7% respectively and yeasts and *Enterobacteriaceae* in relatively lower in numbers.

The counts of LAB in marinated samples ranged from 3.1 - 4.2 log cfu/g, compared with 3.2 log cfu/g in the non-marinated chicken although these final counts were not significantly different from the final densities of their untreated counterparts (P > 0.05). The higher population of LAB noted in samples marinated with "Kalia" and "Massala" could be attributed to the lower pH of these spice mixes. Both of these spices have chili powder in varying proportions (Cuizinemaurice, 2014), and chili powder is known to have a low pH of 4.4 (Peter and Babu, 2012). Adding these spices to chicken is likely to lower the pH and favour the growth of acidophilic and acid-tolerant microorganisms such as lactic acid bacteria (Hutkins, 1993; 2009). This could partly explain the higher population density attained by LAB in marinated chicken over none-marinated chicken. Moreover, lactic acid bacteria are spoilage microorganisms that can occur in spices such as onion and garlic powder, and these ingredients are present in varying proportions in "Kalia" spice mix (Cuizinemaurice, 2014). Davies and Board (1998) reported that even moderate levels of lactic acid bacteria in poultry can in fact result in the release of off-flavours and deterioration of the colour of chicken (Franz

et al., 2010). Hence selecting spices having low counts of lactic acid bacteria, particularly the hetero-fermentative variety, is important for manufacturing of products with an extended shelf-life (Sperber, 2007).

Although spoilage of poultry meat has been largely attributed to bacteria (Corry, 2007), yeasts can also be present in the microbiota. Russell (2009) mentioned that fungi are usually of less importance in poultry spoilage except when antibiotics are employed to suppress bacterial growth. In fact, yeasts have been reported to attain population density as high as 10^6 cfu/g on fresh chicken carcasses during storage (Hinton et al., 2002). In our study, we observed that spoilage yeasts proliferated to a greater extent in spice-marinated chicken (3.8-4.2 log cfu/g) than in the none-marinated samples (3.2 log cfu/g) although these differences were not significant ($P > 0.05$). The higher counts of yeast organisms on marinated chicken noted in our study could be because of the presence of indigenous microorganisms already present in these commercial spice blends. Indeed, spices and their plants can be contaminated with microorganisms during cultivation, processing and packaging (Ito et al., 2008). In addition, we also observed that the addition of spices depressed the water activity of chicken meat by a maximum of 0.23 (Table 4) thus potentially encouraging the growth of spoilage yeasts compared to bacteria (Beuchat, 1983). The yeast isolates could likely belong to the genus *Candida*, *Rhodotorula*, *Debaromyces* or *Yarrowia*, as these are predominant yeasts in poultry (Jay et al., 2007). Viljoen et al. (1998) also indicated that *Candida* and *Debaromyces* were the two most dominant genera of yeasts on both fresh and spoiled carcasses although *Rhodotorula* was not found on any spoiled carcasses. Ismail et al. (2000) further mentioned that the two most abundant species of *Candida* and *Debaromyces* were *C. zeylanoides* and *D. hansenii* on fresh and spoiled poultry.

Physicochemical characteristics of chicken

pH of chicken samples

The pH of chicken marinated with "Tandoori", "Kalia" and "Massala" had decreased significantly ($P < 0.05$) from 10.00 to 6.51, 7.18 and 7.24 respectively and did not change significantly ($P > 0.05$) over the 15-day storage period. On the other hand, the pH of untreated samples had decreased significantly ($P < 0.05$) over the 15-day period from 10.00 to 7.86 (Table 3).

The changes in the pH of chicken during refrigerated storage is comparable to the findings of Zhang et al. (2015) who had observed that spice extract treatments comprising of clove, rosemary and clove + rosemary spice extracts reduced the pH to final values of 5.62, 5.58 and 5.48 respectively which were lower than the pH of the control (6.66). Istrati et al. (2015) also observed that the pH values of beef treated with six different marinades comprising of wine, spices such as black pepper and garlic, and herbs such as thyme and marjoram, decreased from an initial value of 5.70 to a minimum value of 4.90. On the other hand, the same author showed that the pH of untreated beef increased to a final value of 6.16. Zhang et al. (2015) mentioned that the pH increase of control untreated samples could have been caused by the utilization of amino acids by bacteria, which are released during protein degradation following depletion of stored glucose. Indeed, accumulation of ammonia and products of amino acid decomposition are thought to result in an increase in pH (Gill, 1983).

Water activity of chicken samples

The mean a_w of untreated chicken meat was 0.93 while the mean a_w of chicken marinated with the different spices ranged from 0.83 to 0.91 on the day of addition of the spice mix (Table 4). However, over the 15-day storage period, the water activity of chicken treated with "Tandoori", "Kalia" and "Massala" decreased from 0.83 to 0.65, 0.89 to 0.64 and 0.91 to 0.65 respectively. No significant difference was observed among the water activity of samples treated with the difference spice mixes ($P>0.05$) (Table 4).

Table 3. pH of spice-marinated and non-marinated chicken

Days	U	Tt	Tk	Tm
0	10.00 ± 0.41^{Aa}	6.51 ± 0.21^{Ac}	7.18 ± 0.05^{Ab}	7.24 ± 0.13^{Ab}
15	7.86 ± 0.18^{Ba}	7.60 ± 0.33^{Aa}	7.09 ± 0.14^{Ab}	6.88 ± 0.17^{Ab}

U: Untreated, Tt: "Tandoori", Tk: "Kalia", Tm: "Massala"; Data represent mean values of three replicates ± standard deviation; Means within a column with different uppercase superscripts differ significantly ($P < 0.05$); Means within each row with different lowercase superscripts differ significantly ($P < 0.05$).

Table 4. Water activity of spice-marinated and non-marinated chicken

Days	U	Tt	Tk	Tm
0	0.843 ± 0.023^{Aa}	0.687 ± 0.043^{Ab}	0.800 ± 0.017^{Aa}	0.540 ± 0.013^{Ac}
15	0.773 ± 0.020^{Ba}	0.613 ± 0.036^{Ab}	0.570 ± 0.027^{Bb}	0.387 ± 0.047^{Ac}

U: Untreated, Tt: "Tandoori", Tk: "Kalia", Tm: "Massala"; Data represent mean values of three replicates ± standard deviation; Means within a column with different uppercase superscripts differ significantly ($P < 0.05$); Means within each row with different lowercase superscripts differ significantly ($P < 0.05$).

The decrease in the water activity of treated samples over the storage duration could be due to the steady loss of free water by osmosis due to the osmotic pressure exerted by the spice marinades (Gurtler et al., 2014). Moreover, since the spice mixes used in the study have an inherently low water activity of ca. 0.3 (Peter and Babu, 2012), their use in the marinating of chicken could have depressed the water activity of the chicken meat by simple osmosis, or dehydration. When spices are applied to high moisture food products such as raw chicken, the dry spices attempt to reach equilibrium with the food product with which it is in contact by drawing available water from within the flesh to the outside while the spices try to permeate into the food interior (Parish, 2017). The result is a reduction in the water activity (a_w) of the chicken products. Since the minimum threshold to support the growth of spoilage fungi (0.80) is lower than the threshold for bacteria (0.93), the marinated chicken products could more likely support the growth of spoilage yeasts over spoilage bacteria as already indicated above. After 15 days of refrigerated storage, the a_w of untreated chicken had decreased from 0.93 to 0.86 and this is likely due to considerable drip loss reaching as high as 20.1% (Table 6). From our observations, we could infer that drip loss is inversely related with water activity; the higher the drip loss, the lower the water activity of the chicken product. On the other hand, Oliveira et al. (2015) noted that different thawing treatments of chicken breasts resulted in different degrees of drip loss with no effect on the a_w.

Surface colour characteristics of chicken samples

The lightness (L*) values of untreated samples had decreased from 54.3 to 51.3 over the 15-day refrigerated period (Table 5). Galobart and Moran (2004) similarly observed that L* values for refrigerated poultry fillets decreased following 48-h of storage and attributed it to the considerable drip loss. Indeed, we also observed a maximum drip loss of 13.9 and 20.1 % in untreated samples after 1 and 15 days of refrigerated storage (Table 6). Galobart and Moran (2004) also mentioned that further decreases in L* values during prolonged storage could relate to meat drying and shrinkage. In fact, we also observed that longer storage of up to 15-days resulted in concomitantly lower L* values (51.3), higher drip loss (20.9%) and lower water activity (0.77) compared to the initial L* values (54.3), drip loss (0.6%) and lower water activity (0.84) of untreated samples. Indeed, the extensive drip loss after 15 days was observed in the form of white exudation from the chicken meat.

The L* values for marinated chicken were lower than their untreated counterparts and ranged from 40.9 to 46.9 on the day of application. Indeed, untreated chicken initially appeared pale pink and translucent while the marinated chicken exhibited the colour of the added spices i.e. appeared reddish, brownish or yellowish with the addition of the "Tandoori", "Kalia" and "Massala" spice mixes respectively.

Table 5. Comparison of the initial and final surface colour characteristics of spice-marinated and non-marinated chicken during a 15-day storage period

Treatment	Days of storage	Colour parameters		
		L^*	a^*	b^*
U	0	54.3±2.13	9.3±1.01	6.1±0.68
	15	51.3±3.68	9.6±1.62	7.8±0.89
Tt	0	44.1±1.80	13.6±2.19	4.3±0.98
	15	41.6±1.70	7.8±0.75	9.9±0.88
Tk	0	43.7±1.43	5.1±0.53	19.1±1.29
	15	42.9±1.74	6.6±1.09	18.1±0.88
Tm	0	46.9±1.39	6.9±0.17	24.7±1.20
	15	45.6±1.91	7.8±1.07	24.6±2.91

U: Untreated, Tt: "Tandoori", Tk: "Kalia", Tm: "Massala"; L*: Lightness, a*: Redness, b*: Yellowness; Data represent mean values ± standard deviation

As expected, the redness (a*) value for samples marinated with Tandoori (13.6) was higher than either untreated (9.3), Kalia (5.1) or Massala (6.9) marinated samples since tandoori spices are red in colour due to the presence of sweet paprika (Cuizinemaurice, 2014) and occasionally due to the presence of synthetic dye E124 also known as cochineal red (EFSA, 2015). Sweet paprika has a characteristic red colour due to the presence of red-pigmented carotenoids such as capsanthin, capsorubin, zeaxanthin and cryptoxanthin (Zachariah and Gobinath, 2008). However, the redness (a*) of chicken marinated with Tandoori had decreased after 15 days reaching a final mean value of 7.8. The reason for this downshift may be due to the loss of oxy-myoglobin in the meat as well as compositional changes undergone in the Tandoori spice mix (Khan et al., 2015).

The initial b* values were highest in chicken treated with Massala (24.7) and Kalia (19.1), compared to either untreated (6.1) or Tandoori (4.3) chicken, due to the different shades of yellowness of the two spice mixes. Indeed, Massala and Kalia spice mixes both comprise of different proportions of turmeric, which is also known as the "yellow root". After 15 days, chicken marinated with Massala or Kalia still had a persistent yellow colour with b* values of 24.6 and 18.1 respectively. Untreated and tandoori-marinated chicken had slightly higher b* values of 7.80 and 9.94 respectively probably due to acquisition of a slightly brownish colour. Colour changes undergone during storage of poultry arise when by-products generated during lipid oxidation interact with the myoglobin pigment (Khan et al., 2015).

Drip loss

Water within meat exists inbound, immobilized or free forms. Bound water molecules associate with electrically charged reactive groups of muscle proteins (Montgometry, 2007), while immobilized water molecules are attracted to the bound molecules in layers that become successively weaker as the distance from the reactive group on the protein becomes greater (Mills et al., 1989). Free water refers to water molecules that are only held by weak forces (Montgometry, 2007). Drip loss can be determined by quantifying the amount of free water lost in raw chicken meat, cooked whole meat or cooked comminuted meat products (Hertog-Meischke et al., 1997; Lawrence et al., 2003; Otto et al., 2006) and usually gives an indication of the juiciness (Montgometry, 2007). In this study, drip loss of 7.8-16.7% was observed after one day of refrigerated storage for marinated and non-marinated samples (Table 6). Indeed, drip loss can be expressed as milligrams per gram (mg/g) of sample or as a percentage (Montgometry, 2007). However, after 15 days of refrigerated storage, the cumulative % drip loss attained 17.6-30.2% probably due to extensive exudation by osmosis.

Table 6. Cumulative drip loss (% w/w) of marinated and non-marinated chicken

Days of storage	U	Tt	Tk	Tm
0	0.0	0.0	0.0	0.0
1	13.9	10.1	16.7	7.8
15	20.1	19.9	30.2	17.6

U: Untreated, Tt: "Tandoori", Tk: "Kalia", Tm: "Massala"

Extensive drip loss could also probably explain the lowering of the water activity of treated samples from 0.91 to 0.65 and untreated samples from 0.93 to 0.86. Another reason for the increase in the drip water loss is that immediately after refrigeration, the surface of the chicken becomes colder than inside the cell and hence the rate of moisture loss increases, leading to a surge in the loss of water (Garcia et al., 2010). Qiao et al. (2002) studied the effect of marinating on drip loss and colour of broiler breast fillet. A marinade made of water (92.5%), salt (5%), and phosphates (2.5%) was applied for 24 h. Results showed considerable variation in drip loss, pH and meat colour. Drip loss of raw chicken fillets was positively correlated with lightness of raw fillets while water-holding capacity was negatively correlated with lightness (Qiao et al., 2002). Montgometry (2007) mentioned that the pH can greatly affect moisture binding in meat. However, in our study, we did not observe any clear-cut association between pH of samples and extent of drip loss.

Sensory evaluation

The four sensory attributes, which generally influence consumers' decision to purchase fresh chicken meat, are colour, juiciness, flavour and texture/tenderness. Both marinated and control samples had a pleasant texture and appearance on the initial day of storage with ≥ 90% of the panelists indicating moderately high to very high acceptance of the products. After three days of storage, treated and untreated samples were equally well received by the panelists with more than 90% showing moderately high to very high acceptance. This observation is very much congruent with findings of Radha Krishnan et al. (2014) who also showed that the sensorial quality of chicken breasts left untreated or treated with spice extracts fared well in all three attributes up to three days of storage. However, the sensory quality of all samples began to deteriorate after six days of refrigerated storage with more than 70% of the panelists indicating low to no acceptance of the products tested. In fact, after six days of storage, none of the samples were accepted due to their odour, texture and appearance as all of them released very pungent putrid odours and had a sticky appearance. This finding is in agreement with that of Radha Krishnan et al. (2014) who also demonstrated a lower acceptability score for control samples after 6 days. However, contrary to our findings, Radha Krishnan et al. (2014) indicated that the sensory attributes of samples treated with spices only deteriorated significantly after 12 days of storage as opposed to 6 days noted in our study. After 15 days, we observed that all samples appeared sticky and slimy. In fact, the slime layer was the result of individual white colonies forming on the spoiled breast fillet that eventually coalesce to form a biofilm (Russell, 2009). Kong et al. (2007) observed that pork marinated with spices started to produce off-odours characteristic of putrefaction after seven days of storage and more pronounced discoloration was observed on the 14th day for both control and treated pork. Kong et al. (2007) attributed the off-odors to lipid oxidation and ammonia production from breakdown of proteins. Russell (2009) on the other hand mentioned that off-odors do not result from breakdown of the proteins in skin and muscle, rather are released from the direct microbial utilization of low molecular weight nitrogenous compounds such as amino acids, which are present in skin and muscle. Among off-odor producers in general, there is a selection of bacterial species that forms part of the microbiota of fresh poultry (Thomas et al., 1987; Erkmen and Bozoglu, 2016). These include psychrotrophic bacteria such as *Pseudomonas* spp., *Acinetobacter, Moraxella* and *Shewanella putrefaciens* (Thomas et al., 1987). *S. putrefaciens* isolates tend to produce sulphide-like odours as this organism is known to produce hydrogen sulphide,

methyl mercaptan and dimethyl sulphide (Thomas et al., 1987). As Ayres et al. (1950) indicated, the release of off-odors generally precedes the development of sliminess, with the former being first detected when the population reaches about 7.2-8.0 log cfu/g or log cfu/cm^2. Indeed, we also observed significant slime formation after 15 days coinciding with aerobic plate counts reaching 7.9 log cfu/g. Slime formation is an evidence of superficial spoilage that tends to occur because the inner portions of poultry tissue are generally sterile or contain relatively few organisms. The spoilage biota therefore, is restricted to the surfaces (Tellez et al., 2013) and grows in an environment of high humidity such as in the refrigerator.

CONCLUSION

The spice mixes "Tandoori", "Kalia", and "Massala" are frequently used in Mauritius for marination of chicken. The spice mixes variably inhibited the growth of mesophilic aerobic bacteria achieving a maximum reduction of 1.0 log cfu/g, relative to untreated controls. Since a population density of TVC exceeding 7 log cfu/g indicates the onset of spoilage, Tandoori and Massala were found to extend the microbiological shelf-life of the product by 6 and 3 days respectively. However addition of the commercial spice mixes did not improve the sensory attributes of marinated chicken over their untreated counterparts and the sensory shelf-life of both treated and untreated chicken breasts were < 6 days due to significantly reduced acceptability scores.

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Competing interests

The authors declare that they have no competing interests.

Author`s contributions

SS made substantial contributions to conception of the study, acquisition, analysis and interpretation of data and writing-up of the first draft of the manuscript. AR contributed in the design of experiments and reviewing of the manuscript. HN was involved in critically reviewing, revising and formatting the manuscript. HN and AR have given final approval of the version to be published.

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