

CONTROL OF HAZARDOUS MICROORGANISMS IN "KADDID" FROM CAMEL MEAT

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ABSTRACT

Trials of camel Kaddid (a dried salted meat) making were carried out in the laboratory by the traditional procedure. Batches of 5 kg each of camel fresh meat were purchased directly from the slaughterhouse. The meats were sliced, salted and exposed to the sun for drying. The batches were sampled at different times for the microbiological and physicochemical determinations. The former included the standard plate count, total and faecal Coliforms, Staphylococci, Salmonella and Clostridium, the later included the water activity and pH. Results indicated a considerable decrease in the water activity to reach 0.66 after 20 days whereas the pH showed a slight decrease. The microbiological characteristics showed a large phase during the first phase and then a rapid decrease to low levels. Numbers of coliforms and staphylococci were stabilised at less than 1cfu/g, respectively after 12 days and 18 days, and the standard plate count (SPC) were around 40 cfu/g after 20 days. The profiles stabilised at these values during storage.

Key words: Camel meat, drying, hygiene, Kaddid, salting

Removal of water i.e. drying, has been used to preserve foods since ancient times. Kaddid a dried salted meat manufacturing consists of salting, spicing and drying meat under ambient temperatures including a direct influence of sunrays (Bannani *et al*, 1995). This old procedure of meat preservation consists of a gradual dehydration of meat cut to a specific uniform shape that permits the equal and simultaneous drying of the whole batches of meat. Reducing the moisture content of the meat is achieved by water migration from meat to the surrounding air and the continuous migration of water from deeper meat layers to the peripheral zones. Bennani *et al* (1995), demonstrated that the preservation of Kaddid is mainly due to its low water activity. To our knowledge, no data have been published on the preservation of fresh camel meat except the use of some organic acid salt combined with *Bifidobacterium* (Al-Sheddy *et al*, 1999).

In the present study microbiological and physico-chemical characteristics of Kaddid from camel meat were carried out in the laboratory to study the microbiological and physico-chemical changes of the product during the ripening process.

Materials and Methods

Kaddid preparation in the laboratory

Batches of 5 kg each of fresh camel meat were purchased from the slaughterhouse of Rabat (Morocco). These were allowed to mature for 24 h at ambient temperature (around 20°C).

Trimming: This step consist of removing with a knife all visible contaminants and dirty spots. The covering fat from external and internal sides of the meat pieces and the visible connective tissue, such as tendons and superficial fasciae are carefully trimmed off.

Deboning-cutting: The aim of this step is to remove bones without less damage to the muscle. This step consists of cutting the muscles into thin strips. The length of the strips was with a rectangular cross-section of 1 x 1 cm.

Salting: The meat strips were salted at the concentration of 10% NaCl in a 10 L plastic container and allowed to take up salt for 12 h before hanging.

Strips hanging : The strips were hung for drying, using metal hooks, through an appropriate

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arrangement to facilitate air circulation along the whole length of the pieces for a uniform drying.

Physico-chemical determinations : pH of the samples was measured by a pH meter apparatus Crison MicroPH 2000. The activity of water was measured using Aqualab Cx2 instrument (Deacon Devices, Pullman, Washington, USA). Three slices (5 g each) were used to determine the activity of water.

Microbiological determinations : Sausage samples (10 to 15 g), were cut into small pieces and blended in 90 ml of saline water (8.5 g/l) to make the initial dilution (10^{-1}). Serial dilutions up to 10^{-6} were then prepared.

Standard plate count (SPC) : Appropriate dilutions (10^{-1} to 10^{-6}) of the samples were pour-plated on standard plate count agar (PCA) (Difco, USA). The plates were incubated at 30°C for 48 h.

Staphylococci : Staphylococci counts were determined by the pour-plate method. Appropriate dilutions up to 10^{-6} were plated on Mannitol Salt Agar (Difco, USA), the plates were incubated at 37 °C for 24 h. Yellow colonies were counted and checked for Gram and catalase reactions. The isolates were checked for their coagulase reaction on rabbit plasma.

Coliforms : Coliforms were enumerated on Deoxycholate agar (Difco, USA). The plates were incubated at 37°C for total coliforms and at 44°C for faecal coliforms for 24 hours. The appeared colonies on the medium were streaked on the same medium for more purification. Isolated colonies were cultured on trypticase soya agar slants and incubated for 24 hours for further identification. Cultures were stored at 4°C until identification.

Salmonella : 25 g of the sample were added to 100 ml of sterile buffered peptone water (BPW) and incubated for 18 hours at 37°C. Two tubes of tetrathionate broth and 2 tubes of selenite cystein broth (Difco, USA) were inoculated with 1 ml from the BPW and incubated for 24 hours at 37°C. Positive tubes of both media were streaked on Hektoen agar (Difco, USA). The method described by Poelma *et al* (1984) was used for the identification of the suspected colonies blue green white with or without dark centre.

Spore forming bacteria : The initial dilution was heat activated at 80°C for 10 min and

immediately cooled in iced water. Anaerobic sulfite reducing Clostridium were grown on SPS medium (Difco, USA) in tubes which were then inoculated with 2, 1 and 0.5 ml of the heat activated dilution and incubated at 30°C for 24 hours. Dark colonies were counted.

Results

Physico-chemical characteristics

The water activity decreased markedly in the product during drying (Fig 1). The decrease pattern was fast during 10 days and the ultimate value reached was close to 0.75. The decrease continued more slowly during the following days to achieve a value of 0.66. This phenomenon is probably due to the free water driving into the product during the drying process. The water activity is the most important factor to monitor during the dehydrating process. Water activity must be reduced as quickly as possible to stop or to delay undesirable microorganisms in the product. The reached value in our case is 0.66. This may classify Kaddid as an intermediary moisture product.

The pH decreased curiously during the drying period (Fig 2). It is assumed that the pH decreases in post-rigor meats to reach an ultimate value around 5.4, and may increase during the maturation period to reach high values in spoiled meats. A pH reduction can delay the growth of undesirable microorganisms during the first stage of processing, while the moisture of the product is still high enough to support microbial growth. The slight decrease of the pH during the first stage of drying is not easy to explain.

Microbiological characteristics : Microorganisms associated with food hygiene were followed up during the dehydrating process by the determination of standard plate count (SPC), coliforms, staphylococci and enterococci. The microbial profiles are plotted in figure 3. This shows a marked decrease in the counts during the drying process to reach a minimum of less than 1cfu/g after 20 days. The reduction of coliforms numbers may ensure a good degree of preservation against undesirable and/or hazardous microorganisms. Similar decrease of coliforms was reported by Susan *et al* (2002) during drying of beef jerky in which *E. coli* became undetectable (< 10 cfu/g) after 30 days

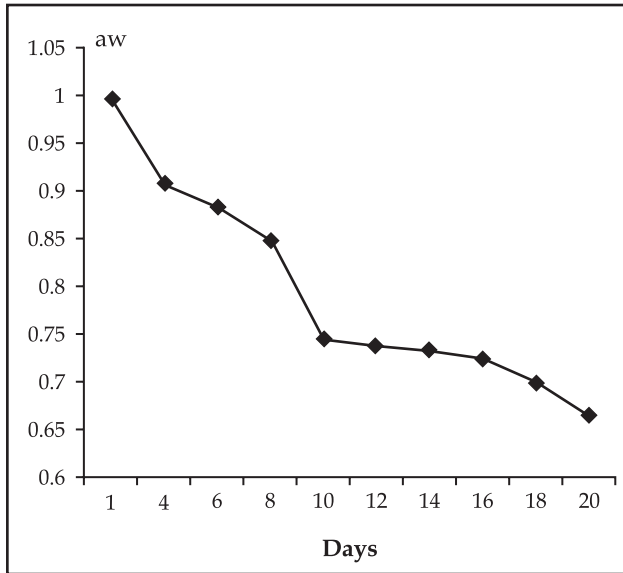


Fig 1. Water activity decrease during camel Kaddid making.

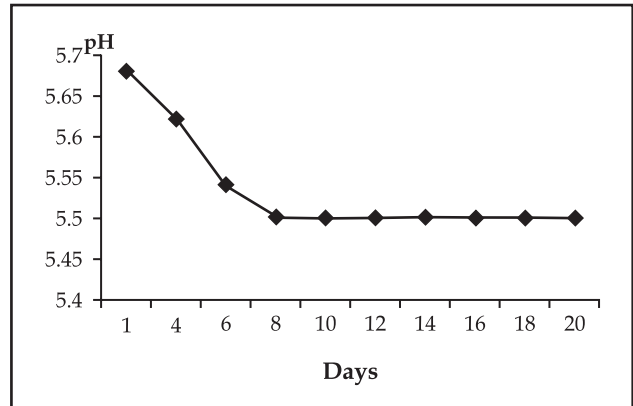


Fig 2. pH decrease pattern during camel Kaddid making.

of storage after treatment for 10 h at 62.5 or 68.3°C. However Keene *et al* (1997) suggested that homemade dried meat can be colonised by *E. coli* and become a source of human infections.

The SPC level in Kaddid, prepared by the traditional procedure from sheep meat, was determined by Bennani *et al* (1995) to be 7.5×10^5 cfu/g. In the present work, a constant profile during the first step of the process was observed, and the counts decreased to reach numbers

around 10^4 cfu/g after 20 days of ripening. The low level obtained at the end of the process may confirm that preservation was carried out properly. The level of SPC reached in the product made in our laboratory is lower than the level found in samples made by the particulars.

Staphylococci counts were decreased drastically by approximately 2 log units during Kaddid drying process. The initial staphylococci counts of the raw material was around 10^5 cfu/g, which decreased to less than one colony after 9 days. *Staphylococci* profiles were higher (6.2×10^5 cfu/g) in Kaddid samples reported by Bennani *et al* (1995).

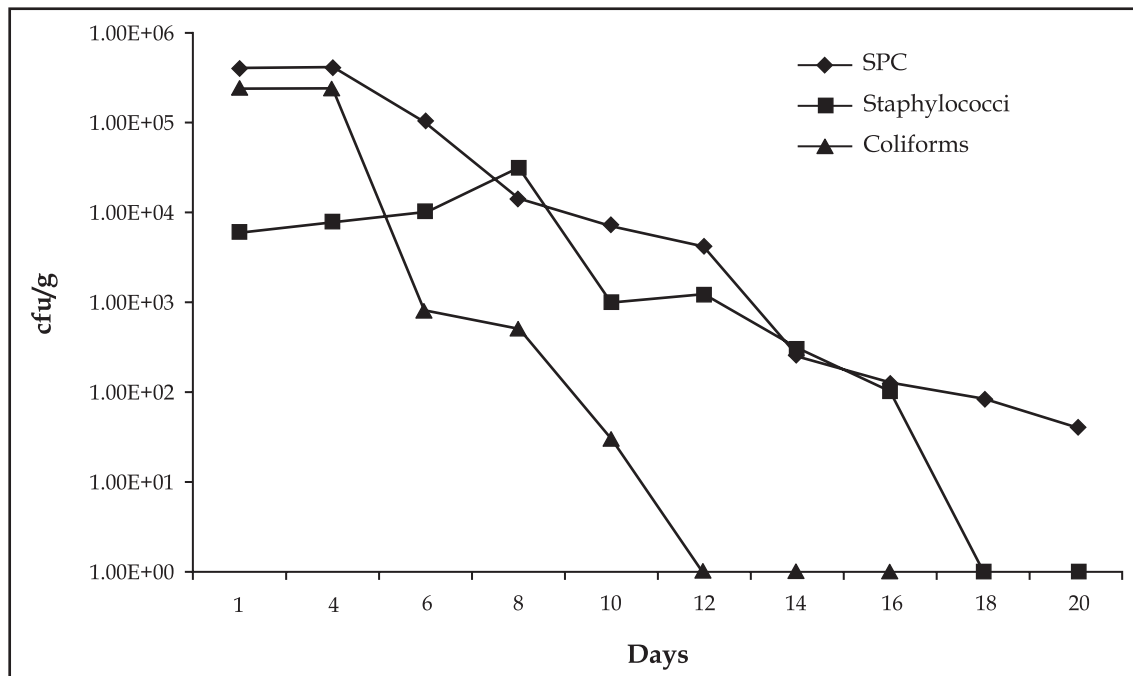


Fig 3. Microbial profiles (SPC, Coliforms and *Staphylococci*) during camel Kaddid making.

Salmonella and *Clostridium* were not detected in fresh camel meat and their absence in the final product would not, therefore, due to their inactivation by drying and salting, since they could not be detected in fresh meat prior to salting and drying. It is generally assumed that enterobacteriaceae are sensitive to salt and drying.

Discussion

The death of bacteria is enhanced by the ionic effect of sodium chloride, as well as the water binding characteristics in the product. Factors such as drying temperature, salt concentration, and pH are important when predicting the minimal activity of water for the growth of *E. coli* O 157 : H7 (Buchanan and Klawitter, 1992; Buchanan and Bagi, 1997). Rowbury *et al* (1996) reported that there are several ways in which Enterobacteria can be damaged in media containing a high sodium chloride concentration. Osmotic shock (Csonka, 1989) caused by a high concentration of Na⁺ in the cytoplasmic membrane (Karpel *et al*, 1991, Ohyma *et al*, 1992) may induce microbial inhibition and enhances lethal effects of Na⁺ and alkaline pH (Small *et al*, 1994) may also have an effect on the viability of microorganisms.

Doyle and Schoeni (1987) suggested the possibility of the existence of coliforms in processed beef products such as jerkies and other dried or semi dried meat products. An outbreak of *E. coli* infections has been associated with the consumption of venison jerky (Keene *et al*, 1997). Ryu and Beuchat (1999) reported that at the same activity of water (0.68), an increase in sodium chloride concentration resulted in significant reductions in the numbers of viable *E. coli* cells. The levels of coliforms reached in our case were less than 1 cfu/g. This low level indicated a successful preservation of camel meat by salting and drying.

Faith *et al* (1998a) reported that *E. coli* O 157 : H7 survived in greater numbers in salami (activity of water 0.90, 4.6- 4.8% Sodium chloride) stored at 4°C than at 21°C for 90 days and Clavero and Beuchat (1996) observed less than a 2 log₁₀ reduction in *E. coli* on salami stored at 5°C for 32°C. Faecal coliforms also survives in greater numbers in pepperoni (activity of water 0.88, 4.6-5.1 sodium chloride) at 4°C than at 21°C (Faith

et al, 1998b). Greater reductions in numbers of *E. coli* have been correlated with increased sodium chloride and sodium nitrite contents in pepperoni (Riordan *et al*, 1998) and curing agents in Chinese style sausage (Yu and Chou, 1997). Harrison *et al* (1998) reported that population of *E. coli* O157 : H7 exhibited a greater decline during drying when a nitrite and salt were added during jerky preparation.

Staphylococci are more salt tolerant than are enterococci or *E. coli*. They have a more rigid cell wall and higher internal turgor pressure. The mechanism of NaCl-induced osmotic tolerance among these bacteria were examined by determining the generation of osmoprotective activity of cellular extracts and intracellular concentrations of glycine betaine and potassium (K⁺) in response to graded amounts of NaCl. *Staphylococci* as well as *E. coli* require choline or glycine betaine to achieve maximal salt tolerance (Kunin and Rudy, 1991). Even if these microorganisms are salt tolerant they reduced to less than 1 cfu/g of the product. This level is safe since the product may have low activity of water and that the growth is delayed or stopped.

References

- Al-Sheddy I, Al-Dagal M, and Bazaraa WA (1999). Microbial and Sensory Quality of Fresh Camel Meat Treated with Organic Acid Salts and/or Bifidobacteria. *Journal of Food Science* (Vol. 64), No. 2.
- Bennani I, Zenati Y, Faid M, Ettayebi M (1995). Physico-chemical and microbiological characteristics of a dried salted meat product (Kaddid) in Morocco. *Z Lebensm Unters Forsch* 201:528-532.
- Buchanan RL and Bagi LK, Goins RV and Philips JG (1993). Response surface models for the growth kinetics of *Escherichia coli*. O 157: H7. *Food microbial*. 10:303-315.
- Buchanan RL and Bagi LK (1994). Expansion of response surface models for the growth of *Escherichia coli*. O 157: H7 to include sodium nitrite as a variable. *International Journal of Food Microbiology* 23:317-332.
- Buchanan RL and Bagi LK (1997). Affect of water activity and humectant identify on the growth kinetics of *Escherichia coli*. O 157: H7. *Food Microbiology*. 10:303-315.
- Buchanan RL and Klawitter LA (1992). The effect of incubation temperature, initial pH and sodium chloride on the growth kinetics of *Escherichia coli*. O 157: H7. *Food Microbiology* 9:185-196.
- Clavero MRS and Beuchat LR (1996). Survival of *Escherichia coli*. O 157: H7 in broth and processed salami as influenced by pH, water activity, and temperature and suitability of media for its recovery. *Applied and Environmental Microbiology* 62:2735-2740.

- Csonka LN (1989). Physiological and genetical responses of bacteria to osmotic stress. *Microbiology Reviews* (53):121-147.
- Doyle MP and Schoeni JL (1987). Isolation of *Escherichia coli*. O157: H7 from retail fresh meats and poultry. *Applied and Environmental Microbiology* (53):2394-2396.
- Faith NG, Parniere N, Larson T, Lorang TD, Kaspar CW and Luchansky JB (1998a). Viability of *Escherichia coli*. O 157: H7 in salami following conditioning of batter, fermentation and drying of sticks and storage of slices. *Journal of Food Protection* (61):377-382.
- Faith NG, Wierzbka RK, Ihnnot AM and Luchansky JB (1998b). Survival of *Escherichia coli*. O 157: H7 in full and reduced fat pepperoni after manufacture of sticks, storage of slices at 4°C or 21°C under air and vacuum, and baking slices on frozen pizza at 135, 191 and 246°C. *Journal of Food Protection* (61):383-389.
- Harrison, JA, Harrison MA and Rose RA (1998) Survival of *Escherichia coli* O157:H7 in ground beef jerky assessed on two plating media. *Journal of Food Protection* (61):11-13.
- Karpel R, Alon T, Glaser G, Schuldiner S and Padan E (1991). Expression of sodium proton antiporter (NhaA) in *Escherichia coli*. O 157: H7 is induced by Na⁺ and Li⁺ ions. *J. Biol. Chem.* (266):21753-21759.
- Keene WE, Sazie E, Kock J, Rice, DH, Hancock, DO, Balan VK, Zhao T, Doyle M P (1997). An outbreak of *E. coli* O157: H7 infections traced to jerky made from deer meat. *Journal of American Veterinary Medical Association* 162:77(15):1229-12231.
- Kunin CM, Rudy J (1991) Effect of NaCl-induced osmotic stress on intracellular concentrations of glycine betaine and potassium in *Escherichia coli*, *Enterococcus faecalis* and *Staphylococci*. *Journal of Laboratory and Clinical Medicine* 118(3):217-24.
- Ohshima T, Imaizumi R, Igarashi K and Kobayashi H (1992) *Escherichia coli* is able to grow with negligible sodium ion extrusion activity at alkaline pH. *Journal of Bacteriology* 174:7743-7749.
- Poelma PL, Andrews WH and Silliker JA (1984). Salmonella. In: Speck ML (ed) *Compendium of Methods for the Microbiological Examination of Foods*. APHA, Washington, D.C. pp 286-326.
- Riordan DCR, Duffy G, Sheridan JJ, Eblen BS, Whiting RC, Blair IS and McDowell DA (1998). Survival of *Escherichia coli*. O 157: H7 during the manufacture of pepperoni. *Journal of Food Protection* (61):146-151.
- Rowbury RJ, Goodson M., Lazim Z and Humphrey TJ (1996). Sensitisation to acid induced by sodium ions in *Escherichia coli*: dependence on (p) ppGpp and cAMP and suppression of the relA-associated defect by mutations in anvZ. *Micorbios* (85):161-177.
- Ryu J and Beuchat LR (1999). Changes in heat tolerance of *Escherichia coli*. O 157: H7 after exposure to acidic environments. *Food Microbiology* (16):309-316.
- Small P, Blankenhorn D, Welty D, Zinser E and Slonczewski JL (1994). Acid and base resistance in *Escherichia coli*. O 157: H7 and *Shigella flexneri*: role of rpoS and growth pH. *Journal of Bacteriology* (176):1729-1737.
- Sussan N, Albright Patrica A, Kedall John S Avens (2002). Effect of marinade and drying temperature on inactivation of *E. coli* O 157 : H7 on inoculated home dried beef jerky. *Journal of Food Safety* (Vol 22), N° 3.
- Yu C-F. and Chou C-C (1997). Fate of *Escherichia coli*. O 157: H7 in Chinese-style sausage during the drying step of the manufacturing process as affected by the drying condition and curing agent. *Journal of Science, Food and Agriculture* 74:551-556.