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Sensitivity of three pathogenic bacteria to Turkish cemen paste and its ingredients

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Abstract

Pastirma is a dry cured meat product which is pasted with cemen. This paste is prepared from ground fenugreek, garlic and red hot pepper (RHPP). In this study, the cemen mix/paste and all its ingredients were tested for their inhibitory effects on *Escherichia coli*, *Staphylococcus aureus* and *Yersinia enterocolitica*. All samples had a varying inhibitory effect against all the bacteria tested during 4 days of storage. Complete cemen paste showed the strongest inhibitory effect on the three pathogens compared to ingredients alone. Fenugreek and RHPP had a bacteriostatic effect while the cemen paste and garlic had a bactericidal effect. *S. aureus* was the most sensitive bacterium while *Y. enterocolitica* was the most resistant. The results of this study confirmed the protective effect of cemen paste and garlic in food preservation especially against *E. coli, S. aureus* and *Y. enterocolitica* proving safety for public health. Hence it might be concluded that cemen paste is the first hurdle to prevent bacterial contamination, and a low pH (~4.83) value would also add an additional barrier to secure safety of the product. Finally, it can be suggested that similar studies should be conducted on actual meat systems to confirm these findings.

Keywords: Fenugreek; Garlic; Red pepper; Pastirma; Cemen; Antibacterial effect

1. Introduction

Meat spoils readily unless kept at temperatures in the proximity of 0 °C or is processed immediately. Therefore, traditional methods of meat preservation, such as salting and drying, still continue to play an important role in many of the world's less developed countries (Norman & Corte, 1985; Yetim, 2000). Salted and dried meat products with specific textural and eating quality characteristics can be produced with an useable shelf-life of 3–5 months without noticeable deterioration, with unique taste and flavor which makes them a delicacy (Leistner, 1987).

Pastirma is one of the salted and partially fermented dry cured meat products which is pasted with cemen (outside covered with a paste which makes it different from its counterparts) and stored for several months without refrigeration, and it is highly regarded and very popular in most of the Middle East countries (Aksu & Kaya, 2001; Aksu, Kaya, & Ockerman, 2005; El-Khateib, Schmidt, & Leistner, 1987; Tekinsen & Dogruer, 2000). The end product is highly tasty and nutritious, and it contains an average of 45% moisture, 30% protein, 15% fat, 5% carbohydrate (from paste) and 5% ash under normal conditions and may be consumed either raw or cooked. The paste, called cemen used to cover pastirma is prepared from the following herbs; 50% fenugreek (ground), 35% garlic (fresh and ground) and 25% red pepper (ground) which are mixed with water to make a slurry like paste. It is then placed on the surface of the meat block, approximately 0.3–0.5 cm thick (Anon, 1991; Gokalp, Kaya, & Zorba, 1999). This paste covering the slabs of pastirma is both an important factor in the flavor and protection of the meat from drying and spoiling by contact with air during

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processing, which might cause the fat to oxidize and produce an undesirable flavor (Anon, 2005).

A number of traditional local dried meat processing techniques have been developed over hundreds of years, but there is no consistent technique which would ensure that the product will always be acceptable in quality and safety (Leistner, 1987; Leistner, 1989; Yetim, 2000). There have been some studies of the microbiological stability of pastirma because microbiological contamination may occur during the pastirma production, and the risk of spoilage or public health would increase. Cemen paste is the first hurdle to protect the product, but there is very limited research on the cemen paste and its ingredients for their antibacterial activity. For example, El-Khateib et al. (1987) stated that cemen paste had about 1.0×10^7 total bacteria and $<10^2$ *Enterobacteriacea* per gram of sample. Dogruer, Nizamlioglu, Gurbuz, and Kayaardi (1998) reported that the total microorganisms count was between 2.8×10^7 and 7.0×10^7/g on the first day and decreased to 2.2×10^6–3.4×10^6/g on the 60th days of storage.

One of the main ingredient of the cemen is fenugreek (*Trigonella foenum-graecum*) which is a leguminous plant grown in northern Africa, the Mediterranean, European, western Asia and northern India. Fenugreek seed have been used medicinally and as a spice (food) for many years (Billaud & Adrian, 2001). Many research reports have indicated that fenugreek seed reduced the total serum cholesterol levels and lowered blood glucose and peroxidation lipids in blood plasma (Anuradha & Ravikumar, 1998). There have also been many reports about antibacterial effects of garlic (Fernandez-Lopez, Zhi, Aleson-C, Perez-Alvarez, & Kuri, 2005; Harris, Cottrell, Plummer, & Lloyd, 2001; Sallam, Ishloroshi, & Samejima, 2004). Dogruer et al. (1998) reported that cemen paste had a positive effect on the microbiological quality of pastirma, and they concluded that this result was from the antimicrobial effects of garlic in the cemen. There is no information on the survival or inhibition of some well-known pathogens in cemen paste. Therefore, the objective of this study was to evaluate the effects of cemen paste and its basic ingredients for their potential to inhibit some common pathogenic bacteria (*Escherichia coli*, *Staphylococcus aureus* and *Yersinia enterocolitica*) encountered in foods.

2. Materials and methods

2.1. Plant materials

Basic cemen ingredients; fenugreek (*Trigonella foenum-graecum* L.), garlic (*Allium sativum* L.) and red hot pepper powder (*Capsicum annuum*) which were purchased from retail markets in Kayseri, Turkey.

2.2. Preparation of test bacteria

Three bacterial species including *Escherichia coli* ATCC 25922, *Staphylococcus aureus* ATCC 2392 and *Yersinia enterocolitica* ATCC 1501 (American Type Culture Collection, Rockville, MD) were used to determine the antibacterial activity in each sample. Bacterial species were obtained from stock cultures and were grown in nutrient broth. *E. coli* and *S. aureus* were incubated at 37 °C for 18 h, *Y. enterocolitica* was grown in nutrient broth and incubated at 22 °C for 18 h. Broth cultures were prepared overnight in nutrient broth resulting in a final cell concentrations of approximately 10^6–10^7 cfu/mL.

2.3. Preparation of cemen paste

Experimental cemen was made of fenugreek, garlic, red hot pepper powder (RHPP) and water. The 29.1% of fenugreek, 20.4% of garlic, 8.7% of red hot pepper powder and 41.8 % of fresh potable water and were homogenously mixed and the mixture was aseptically kneaded. They were then packaged and stored at 4 °C during which time analyses were conducted.

2.4. Chemical analyses of cemen paste

Ten gram of cemen was mixed with 10 mL of distilled water, and the pH was measured using a pH meter (Hanna Inc., Italy). Total moisture (%) contents were determined by heating at 105 °C to a constant weight.

2.5. Determination of inhibitory effects

The inhibitory effects of cemen paste and the basic ingredients against bacterial species were measured using serial dilution method. The fenugreek, garlic and red hot pepper powder (RHPP) were prepared at the 29.1%, 20.4% and 8.7% concentrations in nutrient broth, respectively. Then, these samples were sterilized in an autoclave. Additionally, to appropriate with the practical applications in pastirma processing, the 58.2% (CP1) and 29.1% (CP2) concentrations of cemen paste were mixed in nutrient broth to provide basic nutrients for the bacteria and then the broths were autoclaved. All the samples were separately inoculated with %1 of fresh bacterial culture (*E. coli*, *S. aureus* and *Y. enterocolitica*). Flasks were inoculated with *E. coli* and *S. aureus* and were incubated at 37 °C for 4 days; *Y. enterocolitica* was incubated at 22 °C for 4 days. Every day, the number of colonies in the tubes was counted using serial dilution method in nutrient agar. The growth inhibition level (%) caused by each the ingredient of the cemen and the cemen paste on test bacteria was determined according to the following equation:

$$\text{Growth inhibition level}(\%) = \frac{\text{control population} - \text{treated population}}{\text{control population}} \times 100$$

The control flask was prepared as described above except that the flask contained no added cemen ingredients (Sagdic, 2003). Bacterial counts are reported as log cfu/g or cfu/ml.