The prevalence of bacterial contamination of table eggs from retail markets by *Salmonella* spp., *Listeria monocytogenes*, *Campylobacter jejuni* and *Escherichia coli* in Shahrekord, Iran

Abstract

Introduction and objective: Contaminated egg and its products are increasing the risks of illness in humans. The significance of these diseases in humans can vary from mild symptoms to life threatening conditions. This study was conducted to determine the contamination of egg to food borne pathogens; *Salmonella* spp., *Listeria monocytogenes*, *Campylobacter jejuni* and *Escherichia coli* in Shahrekord.

Materials and methods: One hundred normal eggs randomly purchased in the spring and summer of 2008 from small and big supermarkets of Shahrekord, and delivered to the food microbiology lab to be tested. The contents of eggs cultured for those bacteria on selective agar and standard microbiological tests performed to identify the isolated organism.

Results: The result showed that there was no contamination by *Salmonella* spp., *Listeria monocytogenes* and *Campylobacter jejuni* in all 100 eggs. However, 19 samples were contaminated by *Escherichia coli*, four samples by *Proteus* spp. and one sample by *Klebsiella* spp. Average colony count of coli form bacteria was 20cfu/g and *Escherichia coli* was 12/6cfu/g.

Conclusion: We concluded that *Salmonella* spp., *Listeria monocytogenes* and *Campylobacter jejuni* contamination of eggs does not make up a serious health hazard in this area. *Escherichia coli* are known to contaminate the surface of egg while mechanical process can spread the bacteria through eggs.

Significance and impact of the study: It is important to remember that control is required at all levels in the food chain and by separating cooked and raw.

Keywords: Egg; *Salmonella* spp.; *Listeria monocytogenes*; *Campylobacter jejuni*; *Escherichia coli*
Introduction
Microbial contamination of egg has important outcome to the poultry industry and illness from contaminated egg is a serious public health problem around the world. The significance of these diseases in humans can vary from mild symptoms to life threatening situation [1]. The egg and its products are an important component source of necessary nutrients and a major food within the human diet. In spite of the antibacterial factors, it can be infected with different bacteria such as Salmonella spp., Listeria monocytogenes, Campylobacter jejuni and Escherichia coli. Campylobacter is the most common identified cause of food borne disease.

It has been found mostly in poultry, egg, red meat, unpasteurized milk and untreated water. The egg can act as a vector in the transmission of food poisoning organism. Although it doesn't grow in food, it spreads easily, so only a few bacteria in a piece of undercooked chicken could cause illness [2,3]. Salmonella spp. is the second-most-common cause of food poisoning after Campylobacter spp. It has been found in unpasteurised milk, eggs and raw egg products, meat and poultry. Salmonella spp., can be inside of the completely normal-appearing eggs, and if the eggs are eaten raw or undercooked, the bacterium can cause illness [4].

Listeria monocytogenes can cause illness in pregnant women, babies and people with reduced immunity and febrile gastroenteritis in healthy people. The estimated annual incidence of listeriosis is quite low and poultry farms are not frequently examined for L. monocytogenes, but relatively high prevalence of contaminated raw chicken products has been reported. Thus, contaminated farms can be the source for contamination of the slaughter and processing environment [5]. E. coli is one of the most common bacteria which cause diarrhea especially in children. The elderly, infants, and those with impaired immune systems may have a more severe illness. In these patients, the infection may spread from the intestines to the blood stream, and then to other body sites and can cause death unless the person is treated promptly with antibiotics.

Many investigations around the world reported the outbreak, contamination of egg by the Salmonellas spp., C. jejuni, L. monocytogenes and E. coli [2,6,7]. A couple of reports have confirmed and introduced the egg as dangerous food stuff. So, for the first time we investigated the contamination of the retail eggs by these bacteria in Shahrekord, Iran.

Material and methods
One hundred normal eggs were randomly purchased in the spring and summer of 2008 from small and big supermarkets of Shahrekord, and sent to the food microbiology lab for bacteriological tests. To produce statistically reliable results, the minimum number of eggs was selected based on the number of samples from which the standard error starts converging to an asymptotic value. In the culture of the egg contents, surface of each of the eggs was first disinfected with 70% ethanol. The eggs were broken and the content thoroughly mixed for approximately 1min using a blender.

Each sample was serially diluted with sterile peptone water and presumptive, confirmatory and complementary tests were done for counting and identifying coli form and, E. coli. All tests were duplicated and the means recorded. We used selective agar to culture Salmonella spp. (Selenite cystein, Xylose Lysine Deoxycholate, Salmonella Shigella agar SS), C. jejuni (Skirrow agar, microaerophilic condition) and L. monocytogenes (Frazer broth, BA-PALCAM; Blood agar with lithium
chloride, polymyxin B sulfate, acriflavine-HCl, and ceftazidime). After 24-48h incubation, standard microbiological techniques including cellular morphology and staining and biochemical (TSI, SIM, IMViC, nalidixic acid resistance, cephalothin sensitivity) tests performed to identify the isolated organism; E. coli, Proteus spp. and Klebsiella spp. [6,8]. All of the culture media were provided by Merck, Germany.

**Results and discussion**

The result showed that there was no contamination by Salmonella spp., L. monocytogenes, and C. jejuni in the whole content of all 100 eggs purchased in the spring and summer of 2008 from small and big supermarkets of Shahrekord, Iran. In the present study 19 samples (19%) were contaminated by E. coli, four (4%) samples by Proteus and one sample (1%) by Klebsiella spp. The overall prevalence rate of bacterial contamination of egg was 24%. Average colony count of coli form bacteria was 20cfu/g and E. coli was 12/6cfu/g.

Regarding the increasing consumption of egg and its products, it is necessary to investigate egg contamination. Several factors implicated in egg contamination. The egg shell contamination resulted from deposition of faecal material on the shell, ovariun or oviduct and gut flora, debris material, egg crates, packing and storage, cloths and hands of poultry workers, dust, the environment, weather conditions, transporting and marketing [9]. Among the common contaminant organisms pathogenic to human beings are Salmonella spp., L. monocytogenes, C. jejuni and E. coli [7,10]. The isolated bacteria could cause severe health problems like, diarrhea, nausea and abdominal pain since they are pathogenic. The results of this study showed that there was no contamination by Salmonella spp., L. monocytogenes, and C. jejuni in the whole content of all 100 eggs.

Several reports showed that egg was not internally infected with C. jejuni. Shane *et al.* [11] reported that hens with faecal shedding C. jejuni in farms did not produce infected eggs. Jones investigated the shell and egg content and showed that one shell sample (0.5% of total samples) was Campylobacter positive. Two shell samples (1.1% of total samples) were Salmonella positive. Twenty-one percent of samples were positive for Listeria (33 shells and 5 contents) and no Salmonella was found. Other report showed that the Salmonella contamination in Spain was 1%, Poland 5%, England 0-7%, and India 1.8% [12].

Fortunately in our study Salmonella was not isolated and this suggested that all the study eggs were Salmonella free. This may be attributed to the fact that poultry farmers practice strict medication and care. Nineteen (19%) samples were contaminated by E. coli, four samples to Proteus and one sample to Klebsiella. Average colony count of coli form bacteria was 20cfu/g and E. coli was 12cfu/g. The mean total of coli form count and mean log were higher than the acceptable limits of 10.00 as set by the International Commission on the Microbiological Specification for Food (ICMSF), showing a hazardous implication on the health of egg consumers [13,14].

Jones *et al.* [15] reported the average Enterobacteriaceae less than 0.1 log cfu/ml for the egg contents, with 36.7% of the samples being positive. Cortes *et al.* [7] showed that 45% of eggs were contaminated with E. coli. E. coli are known to contaminate the surface of egg while mechanical process can spread the bacteria through eggs and meat. Contamination with the pathogen while in the field, occur through improperly decomposed manure, contaminated water
and poor hygienic practices of the farm workers [15,16].

Conclusion
Contamination by Salmonella spp. is a major concern in the poultry industry. Fortunately, in our study, the absence of Salmonella spp., L. monocytogenes, and C. jejuni showed the increased notice to hygiene, and hazard analysis critical control point (HACCP) management system have reduced the potential for contamination of these bacteria. However, egg was a source of E. coli, Proteus and Klebsiella contamination. It is important to remember that control is required at all levels in the food chain and by separating cooked and raw. We can help prevent bacterial contamination of table egg from causing egg borne diseases.

Conflict of interest statement: All authors declare that they have no conflict of interest.

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