MICROBIOLOGY AND FOOD HYGIENE IN PUBLIC FOOD SERVICES

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ABSTRACT

A microbiological survey of ready-to-eat food samples using validated methods was performed on 150 samples from 30 public food premises. The determinants investigated were aerobic and anaerobic colony counts, total *Enterobacteriaceae, Escherichia coli, Staphylococcus aureus, Bacillus spp. and Clostridium spp., Salmonella* spp. and *Campylobacter* spp. Results indicated that using current guidelines for ready-to-eat foods, only 13% of samples were satisfactory, 15.6% were acceptable and 71.4% were of unsatisfactory quality. Unsatisfactory results were due to the presence of high aerobic colony counts, *E. coli*, total Enterobacteriaceae and *S. aureus*. There were significant correlations between bacteriological quality and temperature of storage, food hygiene training and waste product management policies. This study on the microbiology of ready-to-eat foods suggests that there is need to improve on hygienic practises in public food service outlets in order to obtain relatively safe products for consumption.

KEYWORDS: Food hygiene, HACCP, Microbiological quality, Public food, viable counts

INTRODUCTION

In Nigeria, there are a large number of public food services distributed along the country, where a considerable number of people eat daily. Serious consequences relating to national productivity and development can arise from lack of hygiene in such food outlets (Gutierrez *et al*, 1999).

The microbiological safety of food is achieved by as far as possible ensuring the absence of pathogenic microorganisms and by all means preventing their multiplication (Beckers, 1988). Food products that have been subjected to an adequate heat-treatment during processing are free of vegetative pathogens and depending on the treatments, of spore-formers and are generally regarded as safe. Thorough epidemiological investigations of several food-borne disease outbreaks have demonstrated that the presence of vegetative pathogens such as *Salmonella* spp. or *Listeria monocytogenes* in the consumed products was frequently due to post-process recontamination (Reij *et al*, 2004). The majority of studies on pathogens in foods are devoted to investigations on their presence in raw materials or on their growth and behaviour in the finished products. Reference to recontamination is, however, only made in relatively few publications and very little is published on the sources and routes of these pathogens into products after the final lethal processing step.

The hitherto used traditional control of food hygiene focused on assessment whether the controlled sanitary and technological practice is consistent with requirements of regulations sometimes comprises also details of minor importance. To put it briefly, in the course of the production process are many check-up points, but only some or possibly only one is a critical control point. Moreover, by periodic supervision the microbiologist and the hygienist are able to record the hygienic and technological state typical only for the time of control. In effect, microbiological examination of final products does not provide information on the conditions of contamination nor ensure protection against it. For these and other reasons the conclusion is reached that the hitherto used traditional approach of the hygiene supervision is not quite effective and must be replaced by a more active approach focused on the control of factors threatening the wholesomeness already during the production process (Matyas, 1992).

The new approach to supervision of food hygiene is the HACCP (Hazard Analysis Critical Control Point) concept. HACCP is a science-based system designed to prevent, reduce or eliminate hazards in food products through appropriate controls during production and processing. HACCP is a systematic approach to the identification, evaluation, and control of food safety hazards be they biological, chemical or physical. The system can be applied to all pathogenic agents transmitted by foods to man from bacteria and their toxins to viruses, parasites, moulds and mycotoxins. It serves as a reliable means to assure food safety and protect consumer's health rather than sporadic microbiological monitoring of end products.

The present study was therefore aimed at establishing the microbiological quality of ready-to-eat food and determination of the relationship between the microbiology of the product and production processes with a view to supervising food hygiene in public food outlets using the HACCP system.

MATERIALS AND METHODS

Collection of samples

A total of 150 ready-to-eat food samples were collected from 30 public food premises in South-western Nigeria. Samples were transferred into sterile, screw-capped glass containers. The containers containing the samples were brought to the laboratory immediately for microbiological analyses in ice coolers.

All the stages in the food processing line were closely followed and monitored at the processing sites. Critical Control Points (CCP) were identified and the risks associated with these points were confirmed by microbiological analyses of samples taken during processing. The observations made were used to determine the critical limits for the CCPs identified during the processing public food.

Microbiological analysis

Approximately 10g of each food sample was homogenized in 90ml sterile 0.1% peptone water with a blender (National, Model MX395N) for 30 seconds (normal speed). The mixture was serially diluted ten-fold in sterile peptone water by the method of Meynell and Meynell (1970) and from the ten-fold dilutions; colony-forming units (cfu) were determined using the pour plate method. Pour plate counts were carried out using the following media, temperatures and incubation periods: Nutrient agar (Oxoid, U.K.) 37°C, 48 h; Sabouraud Dextrose Agar (SDA) (LAB M, idg Plc, U.K) 30°C, 72 h; MacConkey Medium (Oxoid, U.K) 37°C, 48 h; Plate Count Agar (Oxoid, U.K.) 37°C, 72 h. One set of Nutrient agar and PCA plates were incubated anaerobically in anaerobic jars using Oxoid gas generating kit (Harrigan and McCance, 1976).

After incubation, colonies were randomly picked from the plates used for total viable counts. The isolates were purified by sub-culturing and grouped according to their cultural features. They were them tested for Gram reaction (Claus, 1992) and classified by their micro-morphology. Representative isolates were identified after subjecting them to biochemical tests such as pattern of assimilation of various sugars, growth at different temperatures, production of specific enzymes and utilization of complex substrates. The discriminatory scheme of Sneath *et al* (1986) was used to identify bacterial isolates while representative yeast isolates were identified to the level of species according to the procedures of Kreger-Van Rij (1984). Mould isolates were identified to the level of genus by their micro-morphological features as well as the color and nature of their sporulating structures and conidia (Barnett and Hunter, 1972). They were then sub-cultured and identified to the level of species according to Onions *et al* (1981).

RESULTS

A total of 12 different genera of microorganisms were isolated from the food samples as shown in Table 1. These included *Bacillus* spp, *Escherichia coli, Clostridium* spp, *Salmonella* spp, *Staphylococcus aureus, Campylobacter* spp, *Streptococcus* spp, *Proteus* spp *Saccharomyces* spp, *Penicillium* spp, *Aspergillus flavus, Aspergillus niger* and *Rhizopus* spp. *Bacillus* spp, *Clostridium* spp. The moulds were found to be associated with the foods from the beginning in the raw materials up to the left-overs. Microbiological assessment showed that almost all processing steps, except handling of left-overs were critical control points (Table 2). Hazards identified were mainly microbiological. All cooked food samples had high numbers of microflora in them with cfu/gram ranging from 8.6×10^3 to 1.8×10^5. Results of total viable counts indicated that according to the current guidelines for microbiological quality of ready-to-eat foods, only 13% of samples were satisfactory (d" 10^4 cfu/gram), 15.6% were acceptable (10^4 to d" 10^5) and 71.4% were of unsatisfactory quality (e"10^5).

Table 1: Distribution of micro-flora isolated from food samples collected at public food service outlets in south-western Nigeria

Isolate	Raw food	Stored raw food	Cooked food	Stored food	Ready-to-eat food	Left-overs
*Bacillus*spp.	+	+	+	+	+	+
Escherichia coli	+	+	-	+	+	+
Clostridium spp.	+	+	+	+	+	+
*Salmonella*spp.	+	+	-	-	+	+
Staphylococcus aureus	+	+	-	+	+	+
Campylobacter spp	-	-	-	+	+	+
Streptococcus spp	-	-	-	+	+	-
Proteus spp	+	+	-	-	+	+
Saccharomyces spp	+	+	-	-	-	+
Pencillium spp	+	+	+	+	+	+
Aspergillus flavus	+	+	+	+	+	+
Aspergillus niger+	+	+	+	+		+
Rhizopus spp	+	+	+	+	+	+

DISCUSSION

A wide variety of microorganisms, some of which constitute serious health risks (*e.g. Staphylococcus aureus, Escherichia coli, Bacillus* spp., *Penicillium spp,* and *Aspergillus flavus*) were isolated from the raw food samples which are usually purchased from the open market under conditions that allow the organisms in/on them to thrive. . The exteriors of harvested grains, legumes and other food substrates retain some of the natural microflora they had while growing on the field in addition to contamination from soil, insects, and other sources. Processing removes some of the microorganisms but then, there is a possibility of re-contamination during food preparation and handling. Microorganisms were also detected in samples of ready-to-eat cooked food samples.

Table 2: Evaluation of Critical Control Points (CCPs) in public food service outlets

Step	Hazard	CCP	Critical Limit
Purchase of raw materials	Microbiological	Yes	Wholesome materials,Careful handling
Storage of raw materials	Microbiological,	Yes	Storage under aseptic conditions, No new Chemical organisms introduced
Processing of raw materials	Microbiological water	Yes	Use of clean equipments and potable
Food preparation	Microbiological	Yes	Clean utensils, safe water with no faecal indicator organism
Food storage	Microbial re-contamination	Yes	Storage materials must not introduce new organisms, low microbial load
Food Service	Microbial re-contamination	Yes Yes	Free from pathogens, low microbial load, no new organisms introduced
Discard of leftovers	Microbiological, Physical and chemical	No	None but proper waste disposal recommended
Washing up	Microbiological, Chemical	No	None but use of non-toxic soaps, cleansing and draining recommended

It was very significant to note that spore-forming bacteria (*Bacillus* and *Clostridium* spp.) and moulds were found associated with the foods from the beginning with the raw materials up to the left-overs. *Bacillus* and *Clostridium* are spore-forming bacteria that are commonly found in soil, water (through soil-water contamination) and on vegetables. The presence of these bacteria and moulds in food samples in this work may

be unavoidable because the spores of some strains of these organisms are resistant to temperatures as high as 100°C for more than 1 hour. Furthermore, the oxygen level during cooking may be sufficiently low to permit the growth of the clostridia.

The total viable counts of microorganisms in the food samples were relatively high when assessed using the guidelines for microbiological quality of ready-to-eat foods (Anon, 2001). High bacterial and fungal counts may be attributed to the fact that the food handlers held the foods at temperatures lower than 46°C for more than 4 hours. Previous studies had revealed that viable counts of foods prepared in advance and kept at ambient temperatures (20°C to 46°C) for a long period of time (4 hours or more) reach critical levels (Abdulla and Nick, 1995; Fracina and Alexander, 1999).

Staphylococcus epidermidis and *Staphylococcus aureus* are common environmental bacteria and could have been introduced after cooking through cross-contamination. *Staphylococcus aureus* is known to produce an enterotoxin of importance in food-borne illness. The utensils used by the food handlers to serve food could also constitute a health risk. The food workers store their serving utensils on the open table, where they can be readily contaminated with food-poisoning organisms from raw food and from environment (Adrian, 1992).

Escherichia coli, Proteus spp, Klebsiella spp and *Shigella spp* are members of the bacterial family Enterobacteriaceae that normally live in the intestinal tracts of human and animals. *Clostridium perfringes* and some *Streptococcus spp* can also be found in human and animal intestines. The presence of these organisms in the food samples indicated direct or indirect feacal contamination. The presence of *Escherichia coli*, the indicator organism for feacal contamination in food samples, suggests a general lack of cleanliness in handling and improper storage conditions of foods. The water used to wash the serving utensils and storage containers might also have been contaminated with faeces from animal and human intestines.

Since adequate time-temperature exposure during cooking is adequate to inactivate or kill all vegetative microbes and most of their spores and also to destroy some toxins such as botulinal toxin (Abdulla and Nick, 1995; Anon, 2001) the detection of viable microorganisms in cooked food samples could be as a result of inadequate cooking and/or re-contamination. The key components of the HACCP system include identifying potential problems that could cause food to be unsafe to eat, establishing and monitoring targeted control points in order to minimize such problems and then documenting the results obtained. The problems identified were mainly microbiological and critical limits included purchase and use of wholesome raw materials, storage of raw materials and foods under aseptic conditions. Careful handling so as to avoid the introduction of new organisms and use of potable water in food processing are also very important.

The National Advisory Committee on Microbiological Criteria for Foods has endorsed HACCP as an effective and rational means of assuring food safety from harvest to consumption. Traditionally, industry and regulators have depended on spot-checks of manufacturing conditions and random sampling of final products to ensure safe food. This approach, however, tends to be reactive, rather than preventive, and can be less efficient than the HACCP system. A number of U.S. food companies are already using individually tailored HACCP systems in their manufacturing processes, systems are also in place in Canada and in other countries. It will be to the overall advantage of our citizens to adopt this new and efficient programme to ensure food safety in Nigeria.

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