Detection of Bacteria Associated with the Spoilage of Ready-To-Eat Instant Noodles

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ABSTRACT

Instant noodles have been part of the cooking culture of the Chinese for 4000 years, and have gained popularity among college students in most country. This is probably because of its convenience and affordability. In this study, bacteria that are present during spoilage of ready-to-eat instant noodles were isolated and tentatively identified with attempts to estimate spoilage time. A total of thirty packets of instant noodles, made up of three most popular brands, were purchased and code labeled CK, DT and TY to represent these brands. They were boiled for 3-5 minutes and packaged aseptically into semi-air tight plastic bowl plates. Samples were taken at the 12th, 24th and 36th hours post preparation for bacteriological examination. Sensory analysis was also done at different times by 5 semi-trained panelists considering odour, texture and colour. The results indicated that total aerobic counts ranged from 6.1-7.2 log cfug^{-1} at the 12th hour and by the 36th hour, counts had exceeded 8.0 log cfug^{-1}. Gram positive bacteria, mainly Bacillus and Staphylococcus species, were found to dominate all brands during spoilage and persisted till the 36th hour when the samples had been rejected from sensory point of view. Escherichia coli was initially isolated from samples CK and DT, but could not be detected at the 36th hour which might indicate its minor role in spoilage. Samples from the TY brand had the least bacterial counts and better sensory index even up till >20 hours post preparation, and could be said to have better shelf life. There seems to be a consistent relationship between bacterial counts and sensory evaluation.

Keywords: Spoilage, Instant noodles, Bacteria, Sensory Index, Shelf life.

INTRODUCTION

Many fast food products have flooded the market, with instant noodles emerging as one of the most popular items. This appears to be driven by their convenience, affordability and short preparation time. Instant noodle is a steamed and deep-oil fried noodle made from wheat flour (Matsuo and Tanaka, 2008; Bin, 2008). There is evidence that noodles have been part of the Chinese cuisine since 4000 years ago (Lu *et al*., 2005). Instant noodles are commercially available either in cups with the seasoning sprinkled over the noodles or in pouches with the seasoning provided in a satchet inside the pouch (Hou, 2001).

In Nigeria, consumption of instant noodles has increased from 1.1 billion packets in 2009 to 1.44 billion packets in 2013, making Nigeria the highest consumer in Africa followed by Egypt that consumed 130 million packets in 2013 (WINA, 2014). Instant noodles are stable and have shelf life of 4-6 months in tropical areas, and 6-12 months in Northern hemisphere. This is because of their low moisture content, fairly high sodium content (about 2100 mg/100g of noodles), and the resulting low water activity. They can be served after boiling in water for 3-5 minutes or soaking in hot water for 3-4 minutes (Hou, 2001). They are now being promoted as nutrient vehicles by fortifying either the wheat flour used in making the noodles or the seasoning powder (Winichagoon *et al.*, 2006). Consumption of noodles may lead to excessive intake of energy, fats and sodium, but not protein (Park *et al*., 2011). Spoilage of boiled noodles can be due to the presence of bacteria that contaminate it during preparation and handling. However, to the best of available information, reports on bacterial spoilage of ready-to-eat noodles is absent. It is pertinent to determine the bacteria associated with the spoilage of ready-to-eat noodles and spoilage time in order to engender hygiene during and after preparation, to better advise consumers and minimize wastage.

The objective of this study was to isolate and tentatively identify bacteria associated with the spoilage of ready-to-eat instant noodles, with attempts to also estimate the time for onset of spoilage using sensory evaluation.

MATERIALS AND METHODS:

Collection and Preparation of Samples: A total of thirty packets (120g each) of instant noodles were purchased from different shops in Enugu city of Nigeria. The samples were made up of ten samples from each of the three most popular brands of noodles and code labeled CK, DT, and TY. They were boiled with care to be as hygienic as possible, for 3-5 minutes as specified by the manufacturers and aseptically packed into transparent plastic bowl plates covered semi-air tight with lids. They were labeled appropriately and incubated at 25^{o}C. The nutritional contents per 100g, as stated on the packets by the manufacturers, were noted (data not shown).

Isolation, Identification and Bacterial counts: After 12 hours of preparation, 1g of sample from each brand was aseptically collected and serially diluted in peptone water, and plated out on Nutrient agar and McConkey agar media. This was repeated at the 24th and 36th hours. All cultured plates were incubated at 25^{o}C for 24 hours and discrete colonies were counted. The isolates were identified by morphological and some biochemical characteristics.

Sensory analysis: This was performed on the samples at the 10th, 20th, and 30th hour post preparation by a panel of 5 semi-trained individuals. A 3-point scoring system was employed to evaluate Odour, Texture and Colour. Where: 3= Very good, and 1= Unacceptable.

Sensory Index (SI) = (2•C + 2•O + 1•T)/5.

The samples were considered "spoilt" when SI reached 1.8 ie end of shelf life is ≤ 1.8 (Method from Bruckner, 2010).

RESULTS AND DISCUSSION

Bacterial counts of the samples cultured on Nutrient agar and McConkey agar were recorded per time (Tables 1 and 2). At the 12th hour, Samples from DT had the highest bacterial load on Nutrient agar (7.2 log cfug^{-1}) and McConkey agar (6.8 log cfug^{-1}). By th 24th hour, bacterial counts could not be determined on Nutrient agar for CK samples because of the high bacterial load followed by DT samples that had 8.4 log cfug^{-1} as shown in Table 1. Enterobacterial count for DT samples decreased by almost a log cycle between the 12th and 24th hour (Table 2). The high bacterial load recorded for samples CK and DT could be due to their relatively high percentage of total carbohydrate (31% and 26.78% respectively) compared to 20% for TY as presented by the manufacturers (data not shown). Generally, the bacterial

counts are in consonance with the observations of Waduwawara and Manage (2009) that worked with spoilt cooked rice.

Table 1: Bacterial counts per time obtained from the three brands of noodle samples cultured on Nutrient agar

		(Log cfug^{-1})	
Brand	**12th hour**	**24th hour**	**36th hour**
CK	6.4	ND	ND
DT	7.2	8.4	ND
TY	6.1	7.3	ND

ND: Not determined (Colonies not discrete)

Table 2: Enterobacterial counts per time obtained from the three brands of noodle samples cultured on MacConkey agar

		(Log cfug^{-1})	
Brand	**12th hour**	**24th hour**	**36th hour**
CK	5.2	Nil	Nil
DT	6.8	5.9	Nil
TY	Nil	Nil	Nil

Moreover, Gram positive bacteria were found to dominate all brands of noodles during spoilage as shown in Tables 3-5. This is supported by previous studies that associated spoilage of cereals with Gram positive bacteria (Brian *et al*., 1981; Ueda *et al*., 1980). *Staphylococcus* and *Bacillus* species were isolated from all the samples examined, and were found to persist till the 36th hour. These two genera have been implicated previously as among the bacteria that contaminate cereal products (Norman *et al.,* 2009). Gram negative bacteria were not isolated from TY samples. Waduwawara and Manage (2009) reported finding Gram positive bacteria increasing with time while Gram negative bacteria decreased in samples of cooked rice. Since college students consume the largest amount of instant noodles among other convenience

foods (Moon *et al*., 1999), the presence of *Staphylococcus* and *Bacillus* species in ready-to-eat noodles after 12 hours of preparation could trigger disease outbreak among students (Granum, 2001).

Table 3: Bacterial isolates obtained from CK brand of instant noodle at different periods post preparation.

Brand	12th hour	24th hour	36th hour
***Bacillus* sp.**	+	+	+
***Staphylococcus* sp.**	+	+	+
***Pseudomonas* sp.**	+	+	+
Escherichia coli	+	--	--

Key: "+" means "Present"; "--" means "Absent"

Table 4: Bacterial isolates obtained from DT brand of instant noodle at different periods post preparation.

Brand	12th hour	24th hour	36th hour
***Bacillus* sp.**	+	+	+
***Staphylococcus* sp.**	+	+	+
***Pseudomonas* sp.**	+	+	+
Escherichia coli	+	+	--
***Streptococcus* sp.**	+	+	--

Key: "+" means "Present"; "--" means "Absent"

Table 5: Bacterial isolates obtained from TY brand of instant noodle at different periods post preparation.

Brand	12th hour	24th hour	36th hour
***Bacillus* sp.**	+	+	+
***Staphylococcus* sp.**	+	+	+
***Lactobacillus* sp.**	+	+	+

Key: "+" means "Present"

Table 6 presents the sensory indices for all samples during the period of examination. Samples CK and DT were found to be unacceptable by the 20th hour unlike TY.

Table 6: Sensory indices of samples at different periods

Brand	10th hour	20th hour	30th hour
CK	2.0	1.5	1.0
DT	2.0	1.8	1.8
TY	2.6	2.0	1.6

Alklint *et al*., 2004 found fundamental difference between shelf life based on microbial counts and sensory evaluation. This could not be supported by the present study, as there was consistent relationship between bacterial counts and sensory analysis.

In conclusion, Gram positive bacteria were found to be more associated with the spoilage of ready-to-eat noodles than Gram negative bacteria. On the average, spoilage seems to commence before the 20th hour after preparation. However, TY samples were found to be most stable.

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