

N. J. Microbiology, 2(2), 1982, pp 168.- 172
© The Nigerian Society for Microbiology 1982.

BACTERIA CAUSING BEEF SPOILAGE IN A MEAT SHOP IN IBADAN

By:

D. O. ALONGE

Department of Veterinary Public Health and
Preventive Medicine
University of Ibadan, Ibadan.

ABSTRACT

A study of the bacteria found growing on and spoiling beef in a meat shop in Ibadan is made. Those in the genera *Micrococcus*, *Lactobacillus*, *Flavobacterium* and *Pseudomonas* were found to be the main spoilage organisms. *Pseudomonas* spp were isolated under both warm and cold storage conditions and hence they constitute the major bacteria that will spoil stored meat.

INTRODUCTION

Today, meat quality means more than just an attractive appearance. Quality includes characteristics such as tenderness, flavour, freshness and wholesomeness. These are affected by the quantity and quality of the microbial growth causing chemical changes resulting in the meat being unacceptable to the consumer due to slime, souring or putrid odours.

The identity of the bacteria present on fresh meat is necessary to anticipate the extent of spoilage under subsequent storage and handling procedures. While storage of the meat in open chillers and fridges will encourage the growth of *Pseudomonas* spp mainly, packing in polythene sheets create anaerobic atmosphere for *Clostridium* species. Vacuum packaging of meat will prevent spoilage by *Pseudomonas* while facultative anaerobes like *Lactobacillus* and *Microbacterium* are not prevented from growing (CSIRO, 1977).

Bacterial spoilage is manifested in a number of ways, the most common being, off odours, slime, rancidity of fats, bone-taint discolorations and phosphorescence. Spoilage usually begins to become apparent when the surface count exceeds 10^7 organisms/cm² or per gram. (Nottin-gham, 1971).

Species of *Pseudomonas*, *Achromobacter*, *Streptococcus*, *Bacillus*, *micrococcus* and *Lactobacillus* have been shown to grow on beef and produce slime indicating spoilage (Ayres, 1960; Gardner, 1969; and Gill, et al, 1979). These are differentiated from the intrinsic bacteria of meat by Gill, 1979. However many workers believed that *Pseudomonas* spp. constitute the most important spoilage bacteria even in meat stored in chillers (Brown, et al, 1958; Smith, et al, 1975 and Grau, 1974).

This work aims to classify by genera the bacterial flora of fresh meat (beef) based on cultural studies and some biochemical activities of the organism. This is with a view to anticipate the type of spoilage and possible public health implications. No evidence of any work done in Nigeria in this field is available.

MATERIALS AND METHOD

Meat samples were taken from beef carcasses kept overnight in chillers at the Ministry of Agriculture Meat Shop, Bodija, Ibadan.

I modified the template surface sampling method developed by Yokoya and Zulzke (1975)

by not limiting myself to a swab of an area of 10cm^2 using metal templates but obtaining samples of the surface top layer of 10cm^2 and 0.2cm thick. The samples are then weighed and homogenised in 50ml of 0.1 per cent peptone water using a Colworth's stomacher. The suspension is allowed to settle and the clear upper layer is decanted and used for plating. Duplicate plates were made on blood agar for initial and subsequent isolations. Duplicate plates were incubated aerobically for 24 hours at 35°C and 4°C respectively. Identifications of the isolates were made with the aid of Bergey's Manual of Determinative Bacteriology, 8th edition (1974).

Characterisation was based on colonial cultural characteristics, individual bacterial characteristics motility and some biochemical reactions (see Table 1).

TABLE 1
Cultural, Morphological, Biochemical Characters Used to Differentiate the
Surface Bacteria of Beef Meat

	Micrococcus	Lactobacillus	Bacillus	Staph.	Flavo- bacterium	Pseudomonas	Aeromonas	Acinetobacter	Achromobacter/ Alcaligenes
Gram	+ve	+ve	+ve	+ve	-ve	-ve	-ve	-ve	-ve
Shape	cocci	rod	rod (spore)	cocci	rod to coccobacilli	rod	rod	mainly coccobacilli	rod to coccobacilli
Motility	-ve	-ve	+ve	-ve	-ve	+ve	+ve	-ve	+ve
O-F	O or no reaction	F	both	F	mainly O	O or no reaction	F	O	O or no reaction
Catalase	+ve	-ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Oxidase	+ve (variable)	-ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Nitrate Reduction	-ve (variable)	-ve	variable	+ve	mainly	+ve	+ve	variable	-ve
Indole	-ve	-ve	ND*	-ve	+ve	-ve	+ve	-ve	-ve
Methyl Red	-ve	-ve	ND	-ve	+ve	-ve	+ve	-ve	-ve
V-P	-ve	-ve	ND	-ve	+ve	-ve	+ve	+ve	-ve
Others (Colonial character- istics)	Cream- white, smooth, dis- crete, large cocci in 2's and 4's	Yellowish, mucoid, spreading, loses +ve gram easily. In 1's 2's or 3's.	Rough, mucoid, irregular, yellowish, wide zone of haemo- lysis.	Cream- white, smooth, entire, cocci in bunches	Yellowish smooth, discrete, haemo- lytic, small rods.	Greyish, rough, mucoid, small to long straight or curved rods.	Whitish, mucoid, entire, medium to large rods.	Greyish, confluent, mucoid, smooth, coccoid short rods.	Mucoid, irregular spreading colonies, slender rods.

*ND - Not done

The organisms in the family Enterobacteriaceae were differentiated from the other groups by their gram and Oxidase reactions.

Cytophaga is closely related to the Enterobacteria but easily differentiated by its proteus-type swarming growth on solid agar. Cytophaga are aerose negative and form long filamentous non-branishing rods.

RESULT

Ninety three bacterial isolations from beef, under two temperatre conditions (35°C and 4°C), were made. (see table 2).

TABLE 2

Summary of Isolation

Gram Reaction	Organism per Genera	Frequency of Isolation	Primary Isolation Temperature
Positive	Micrococcus	16	35°C
	Lactobacillus	10	35°C
	Bacillus	3	35°C
	Staphylococcus	3	35°C
Negative	Flavobacterium	10	4°C
	Pseudomonas	16	4°C and 35°C
	Aeromonas	8	35°C
	Acinetobacter	6	35°C
	Achromobacter/		
	Alcaligenes	4	4°C
	Cytophaga	6	35°C
	Enterobacteria	6	35°C
	Others - Unclassified	5	4°C and 35°C.

Micrococcus spp and *Lactobacillus* spp. constituted the majority among the gram positive bacteria isolated. *Pseudomonas* spp. *Flavobacterium* spp, and *Aeromonas* were the most commonly gram negative bacteria isolated. There were 5 isolations which were gram negative but because of their varying and inconclusive colonial and morphological characteristics and biochemical reactions did not fit into the Table of diagnostic criteria. Some of these unclassified bacteria grew at both 30°C and 4°C.

DISCUSSION

The modified template surface sampling method used in this work has been found to give a more accurate picture of the bacterial load of the meat because it relates the surface area to depth. Recently, aerobic spoilage bacteria have been found to be able to penetrate in significant numbers, with 61-64 per cent correlation between the surface down to a depth of 1.5cm (Alonge, 1980). This was why ordinary swab method was not used.

This work was limited to the Bodija Ministry abattoir alone because it is the only meat shop in Ibadan with a near ideal infrastructure for handling meat. It has acceptable, clean environment and working chiller rooms.

Incubation of duplicate plates at both 35°C and 4°C was to simulate subsequent storage environments. These are the average market meat stalls and household refrigerator temperatures respectively. Since only *Pseudomonas*, *Flavobacterium*, and *Achromobacter* Species were isolated at 4°C, these would constitute the major bacteria that would spoil meat stored in refrigerators. Meat left on market stalls, kitchen cupboards and inside non-refrigerated vehicles may therefore be spoiled by all the others including some *Pseudomonas* spp. which also grow in warm temperatures. This goes further to confirm what other workers (Ayres, 1960; Jay, 1972;

Smith, et al, 1975) in other countries, have found out that *Pseudomonas* spp. are the most important spoilage organisms of stored meat.

The presence of *Staphylococcus* spp. in the meat is inconclusive to indicate possible human contamination or ability to cause food poisoning as more work is indicated to determine whether one is dealing with the virulent *S. aureus* or the relatively avirulent *S. elbus* of the normal flora of the human skin, mouth and upper respiratory tract. Isolation of some organisms in the Enterobacteria group on meat definitely indicates faecal contamination which may be as a result of poor dressing techniques or contamination by meat handlers. Other bacteria may have been present but not recovered by the limited procedures used in this study. If plates had been incubated under anaerobic conditions, other spoilage bacteria could have been isolated.

In conclusion, sterile production of meat is impossible but the slaughter, dressing and handling methods must aim at minimising the number of potential spoilage organisms if the products are to have a reasonable storage life in the hands of the consumer.

LITERATURE CITED

- Alonge, D. O. 1980. Estimation of Meat Spoilage Level: Two Techniques for the Enumeration of Bacteria in Venison Meat. Nig. J. of Science Vol. 14.
- Ayres J. C. 1960. Temperature relationship and some other characteristics of the microbial flora developing on refrigerated beef. Food Res., 25: 1 – 18.
- Buchanan, R. E. and Gibbons, N. E. 1974 (Co-Editors): Bergey's Manual of Determinative Bacteriology (8th Edition) Publisher: The Williams and Wilkins Co., Baltimore, PP 99 – 444.
- Brown, A. D. & Weidemann, J. F. 1958 The Taxonomy of the Psychrophilic Meat-spoilage Bacteria: A reassessment. J. Appl. Bact. 21: (1), 11 – 17.
- CSIRO Meat Research Newsletter: Introduction to Meat Microbiology CSIRO, Queensland Australia No. 77/2 April 1977. PP. 10.
- Gardner, G. A. 1969. Physiological and Morphological characteristics of *Kurthia zopfii* isolated from meat products. J. Appl. Bacteriol. 32: 371 – 380.
- Gill, C. O. 1979 A Review of Intrinsic Bacteria in Meat. J. of Appl. Bact. 47. 367 – 378.
- Gill, C. O. and Lowry, P. D. 1981 A note on the identities of organisms causing Black spot spoilage of Meat. J. Appl. Bact. 51, 183 – 187.
- Gill, C. O., Newton, K. G. and Nottingham, P.M. 1979. Microbiology in the Meat Industry. Meat Industry Research Institute of New Zealand Publication No. 695: 6 – 9.
- Grau, F. H. 1974. Microbiology of Unpacked Meat. CSIRO Meat Research Laboratory Publication October, 1974: 5 – 6.
- Jay, J. M. 1972 Mechanism and detection of microbial spoilage in meats at low temperatures – a status report. J. Milk Food Technol. 35: 467 – 471.
- Nottingham, P.M. 1971 Microbiological quality control in the meat Industry. Meat Industry Research Institute of New Zealand Publication No. 217: 2 – 11.
- Smith, F. C., Adams, J.C. and Field, R.A. 1975. Predominant psychrotrophic bacteria on fresh and aged ground beef and antelope. J. Milk Food Technol., 516 – 517.
- Yokoya and Zulke 1975 Method of sampling meat surfaces. Appl. Microbiol. April 1975: 551 – 552.

THE A

The was inv
nate and
30°C te
source f
The pre
tern app
quite ad

Pse
gram-ne
can be c
as pyoc
fluoresc
(Stanier
vate can
when t
glutama
et al, 1
of *Pse*
et al, 1
fructos
synthes
(1976)
lised th
it to gl
et al (1
observ
far, it
when 1
of *P. fi*