

## A Microbial Survey of the Gastrointestinal Flora of Pigs in Dschang, West Province, Cameroon

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### ABSTRACT

Pig husbandry is of prime importance in Dschang and has suffered numerous plagues of gastrointestinal infections. The aim of this study was to assess pig rearing in Dschang and determine the gut microflora of these animals. Between February and May 2003, a questionnaire was used to collect data on pig management from 114 farmers resident in Dschang and its environs. Microbial analysis was carried out on 243 faecal samples collected from pigs reared on these farms. The faeces were processed using standard classical techniques. The questionnaire survey revealed that pigs were reared in an intensive manner with provision of suitable housing. Majority of the farmers were illiterate with very low income. Poor hygiene and diet, storage problems and lack of medical attention were found to militate against good pig production. The 243 pigs examined for microbial infection harboured one or more of the following organisms: *Escherichia coli* (98.4%), *Salmonella* spp. (44.9%), *Candida albicans* (42.8%), *Mucor* spp. (26.7%), *Proteus* spp. (17.7%), *Klebsiella* spp. (8.2%), *Pseudomonas* spp. (7.4%), *Enterobacter* spp. (5.4%), and *Rhizopus* spp. (2.1%). Multiple microbial infestation of the gut was more frequent (85.2%). Humans or other animals can be infected by these organisms if faeces are dumped or washed into water bodies used for drinking, or washed to contaminate grazing land, or when flies land on contaminated faeces then settle on food, or hands contaminated with faeces are used to prepare food. From this study, it can be concluded that pig farmers need to be educated on appropriate management systems to improve production, increase their income and prevent public health hazards. Antimicrobial therapy should also be considered paramount when the animals suffer gut pathology, since they harbour pathogens and coliform bacteria.

**Key words:** Pigs, Management, Faeces, Micro organisms.

### RESUME

L'élevage du porc est d'une importance capitale à Dschang mais il souffre de nombreuses infections gastro-intestinales. L'objectif de cette étude était d'évaluer l'élevage de porcs à Dschang et de déterminer la microflore intestinale de ces animaux. Entre février et mai 2003, un questionnaire a été administré pour collecter quelques données sur l'élevage des porcs chez 114 éleveurs résidant à Dschang et ses environs, et une analyse microbienne a été effectuée sur 243 échantillons fécaux des porcs de ces élevages. Les selles ont été analysées par les techniques classiques. Les 243 porcs examinés étaient infectés par un ou plusieurs des organismes suivantes : *Escherichia coli* (98.4%), *Salmonella* spp (44.9%), *Candida albicans* (42.8%), espèces de *Mucor* (26.7%), *Proteus* (17.7%), *Klebsiella* (8.2%), *Pseudomonas* (7.4%), *Enterobacter* (5.4%), et *Rhizopus* (2.1%). Les infections microbiennes multiples étaient les plus fréquentes (85.2%). Les hommes ainsi que d'autres animaux peuvent être infectés par ces organismes si les selles contaminées sont jetées ou lavées dans les eaux de boisson ou dans les pâturage. Les mouches et les mains souillées par les fèces infectées favorisent aussi la contamination par les aliments. Les eaux de ruissellement en provenance des champs et contenant des selles non traitées peuvent aussi contaminer le sol, les cultures, les animaux et les hommes. A partir des questionnaires administrés, il est apparu que les porcs sont élevés de manière intensive dans des logements appropriés. La majorité des éleveurs sont illettrés et possèdent de faible revenus. De mauvaises conditions d'hygiène alimentaire, le problème de stockage et le manque de suivi médical ont été identifiés comme facteurs limitants pour une bonne production porcine. Cette étude conclut que les éleveurs de porcs ont besoin d'être informés ou formés sur les systèmes d'exploitation appropriés pouvant permettre d'améliorer leur production, accroître leurs revenus et prévenir les risques de santé publique. Le traitement antimicrobien doit être fortement recommandé quand les animaux souffrent d'affectations gastro intestinales, causées par des bactéries coliformes et d'autres micro organismes pathogènes.

**Mots Clés :** Porcs, Exploitation, Fecès, Microorganismes.

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## INTRODUCTION

Worldwide, pig husbandry is practised for the production of meat, pigskin, bristles and manure, and it has become a rapid source of income in some tropical countries because of the pig's high fecundity and growth rate (Williamson and Payne, 1989). In Dschang, fresh pork is a predominant source of protein, and provides revenue to the farmers. Furthermore, pigs are used during socio-cultural activities and their manure serves as fertilizer on vegetable farms. However, one of the major problems associated with pig production is the considerable morbidity and mortality in the animals caused by gastrointestinal infections (Kaufmann, 1996; Martineau, 1997). Generally, microbial diseases of the gastrointestinal tract are known to result from the ingestion of food or water containing pathogenic organisms or their toxin, and are influenced by various factors including poverty, malnutrition, overcrowding and natural reservoirs of pathogens among others (Wistreich and Lechtman, 1984). The necessity of pigs particularly in this subregion call for in-depth studies on their management in order to recommend measures which will enhance their health and productivity for greater societal and economic benefit.

Works have been carried out on the parasitic load (helminths) of pigs in Dschang (Tape, 1990; Mpoame *et al.*, 1995) but not on other organisms present in their gut. Knowledge of all the gut flora of the animals is necessary, not only to aid appropriate management, but also to elude public health hazards. Thus, the objective of this study was to determine microorganisms present in the faeces of pigs in Dschang with the view to suggesting healthy management systems for the animals to farmers and the public at large.

## MATERIALS AND METHODS

This study was carried out between February and May 2003 in Dschang, Menoua Division, which is situated in the Western Highlands of Cameroon. From inception, the aim of the study was explained to pig farmers resident in Dschang and its environs, and only farmers who indicated their interest in the work were selected. A total of 243 pigs and piglets present on 114 farms were included in the study.

### Questionnaire survey

A questionnaire was drawn up to collect data from the farmers which included details such as the date of sample collection, location of the farm, socio-economic status of the farmer (i.e. any other occupation, literacy and the financial status of the farmer) and

whether or not the farmer had formal knowledge of pig farming, number of animals on the farm, age and sex of each animal, type of rearing system, underlying clinical condition of the animals, health attention given to the animals, kind of feed and frequency of feeding, physiological state of each animal (pregnancy, lactating or normal), frequency of cleaning the piggery/animals, and methods of disposal of faeces. Such data were obtained a day before sample collection.

### Collection of faecal samples

Faecal samples were collected early in the morning (between 7 and 9.30am) or late in the afternoon (between 4 and 6.30pm) in already appropriately labelled sterile wide-necked glass bottles, avoiding urinary and environmental contamination. The time of sample collection coincided with the period of feeding and defaecation of the animals. The surroundings were thoroughly cleaned prior to sample collection. The animals were fed and then allowed to defaecate, after which the top layers only of the faecal samples were immediately picked up from the ground with an applicator stick. All these samples were immediately packed into a cold chain system (improvised ice box) and transported to the Applied Ecology Laboratory of the Department of Animal Biology of the University of Dschang for analysis.

### Laboratory examination of faecal samples

The colour, consistency and constituency of the stool samples were noted. Samples were described as either formed (solid), semi-formed (semi-solid) or unformed (liquid), and whether or not mucus, blood and worms were present.

Secondly, direct inoculation of each faecal sample was done on duplicate plates of MacConkey agar, one incubated at room temperature (for culture of *Yersinia* spp.) and the other at 37°C for 24 – 48 hours to culture other gram-negative bacteria. Further, several loopfuls of each sample were inoculated into Brain Heart Infusion broth (an enrichment medium for all organisms present in the samples) and incubated for 2 hours at 37°C. A loopful of the emulsified faeces was then subcultured onto MacConkey agar and Nutrient agar (a general purpose medium for microorganisms) and incubated at 37° for 24 – 48 hours, examining for growth after overnight incubation. A loopful of each stool sample in the broth was also inoculated onto duplicate plates of Sabouraud Dextrose agar (a selective medium for yeasts), one set incubated at 37°C and the other at room temperature for up to two weeks, examining plates for growth daily. All colonies which

**Table I:** Microbial infection of the gastrointestinal tract of pigs in Dschang, February – May 2003.

Microorganisms isolated	Total no. isolated (% Prevalence) n=243	Total no. of farms contaminated (% contamination ) n= 114
<i>Escherichia coli</i>	239 (98.4)	113(99.1)
<i>Salmonella spp.</i>	109 (44.9)	64 (56.1)
<i>Candida albicans</i>	104 (42.8)	64 (56.1)
<i>Mucor spp.</i>	65 (26.7)	39 (34.2)
<i>Proteus spp.</i>	43 (17.7)	20 (17.5)
<i>Klebsiella spp.</i>	20 (8.2)	18 (15.8)
<i>Pseudomonas spp.</i>	18 (7.4)	12 (10.5)
<i>Enterobacter spp.</i>	13 (5.4)	10 (8.8)
<i>Rhizopus spp.</i>	5 (2.1)	4 (3.5)

n = 243: total number of faecal samples analysed

n = 114: total number of farms sampled

grew on the agar media were identified according to standard techniques (Murray *et al.*, 1995). The Chi square test was used to compare frequencies and prevalences of infection at the 0.05 significant level.

**RESULTS**

All the farmers included in this study practised the intensive system of pig management with suitable housing for the animals. Generally, farmers constructed pens (about 2 meters high) with slatted nonspaced wooden floors raised about one meter from the ground, and with the provision of a dunging passage and a zinc roof which gave adequate shade to the animals. The piggeries were well ventilated and the animals well sheltered.

About 85% of the farmers fed the animals twice a day with a combination of commercial feed, household waste food and kitchen offal. Fifteen percent of the farmers fed the animals once a day with commercial feed but also intermittently gave kitchen refuse when available.

Majority of the farmers (95%) were illiterates with low income who never sought medical attention for the animals: 3.7% of the farmers occasionally and only 1.2% always asked veterinary advice as to the most effective ways of preventing and controlling diseases on the animals. Only 4.5% of the pigs studied were in gestation, 2.5% were sucklers while most (93%) were in the normal state, out of which 60.5% were piglets. None of the animals investigated manifested physical

symptoms of gastrointestinal infection. Less than 15% of the farmers cleaned the piggery once a day and all the animals under study were never cleaned. Farmers dumped untreated faeces either under a tree or a plantain plant away from the piggery and household or into water bodies.

Of the 243 pigs examined, 89 (36.6%) were males and 154 (63.4%) females, with ages ranging between one and thirty-one months. It was noted that 76% of the faecal samples collected were yellowish, 19.5% were brownish and 4.5% greenish. Formed faeces (79.4%) made up the bulk of the samples, 15.6% were semi-formed and 5% unformed. None of the stool samples examined contained blood or worms while 0.8% were mucoid.

Only gram-negative bacteria and fungi were isolated from the stool samples analysed (Table 1). The isolation rate for *Escherichia coli* (98.4%) was significantly different ( $P < 0.05$ ) from rates obtained for other common organisms, namely *Salmonella spp.* (44.9%), *Candida albicans* (42.8%) and *Mucor spp.* (26.7%). *E. coli*, *Salmonella spp.* and *C. albicans* were also most frequent in more than 50% of the farms. Single microbial colonization of the gut occurred only in 36 (14.8%) animals with *E. coli* infesting 34 (14%) and *Proteus spp.* 2 (0.8%). Multiple microbial infestation of the gut was more frequent (85.2%), particularly in farms with more than three animals, and *E. coli* and *Salmonella spp.* occurred together in most of the samples (>80%). A total of 81 (33.3%) specimens had two organisms present with the

**Table 2:** Prevalences of faecal microorganisms of pigs in Dschang according to sex.

Microorganisms isolated	Male	Female
	n=89 Positive faecal samples (%)	n= 154 Positive faecal samples (%)
<i>Escherichia coli</i>	86 (96.6)	153(99.4)
<i>Salmonella spp.</i>	32 (36.0)	73 (47.4)
<i>Candida albicans</i>	33 (37.1)	71 (46.1)
<i>Mucor spp.</i>	22 (24.7)	43 (27.9)
<i>Proteus spp.</i>	20 (22.5)	23 (14.9)
<i>Klebsiella spp.</i>	7(7.9)	13 (8.4)
<i>Pseudomonas spp.</i>	8 (9.0)	10 (6.5)
<i>Enterobacter spp.</i>	5 (5.6)	8 (5.2)
<i>Rhizopus spp.</i>	3 (3.4)	2 (1.3)

n = total number of animals sampled per sex group

association of *E. coli* and *Salmonella spp.* (14.8%) in preponderance. Triple microbial infestations were observed in 95 (39.1%) faecal samples with *E. coli*, *Salmonella spp.* and *C. albicans* 29 (11.9%) on the lead. Quadruple infestations were identified in 31 (12.8%) stool specimens and *E. coli*, *Salmonella spp.*, *C. albicans* and *Mucor spp.* 18 (7.4%) was the predominant association.

Significant differences were not apparent in the microbial flora of the faeces, which differ in colour, consistency and constituency ( $P>0.05$ ). The physiological state and sex of the animals did not also influence the microbial flora (Table 2). *Rhizopus spp.* were isolated

only in animals less than 6 months of age, while species of *Proteus*, *Klebsiella* and *Enterobacter* were not isolated in animals above 19 months (Table 3).

## DISCUSSION

Pigs in Dschang generally are reared under intensive management, especially as cropping of the land occurs throughout the year, thus greatly limiting the space available for animal production (Manjeli *et al.*, 1992; Mpoame *et al.*, 1995; Tegua *et al.*, 1997). Also, the fact that urban and rural councils warn against stray animals is a contributing factor to the intensive system of management under practice. In the present study, some factors which were found to adversely affect good

**Table 3:** Prevalences of faecal microorganisms of pigs in Dschang according to age groups.

Microorganisms isolated	Age groups			
	1 – 6 months (n =147)	7– 12 months (n = 65)	13 – 18 months (n = 20)	19 – 31 months (n = 11)
	Number (%) of animals infected			
<i>Escherichia coli</i>	143 (97.3)	65(100)	20(100)	11(100)
<i>Salmonella spp.</i>	66 (44.9)	24(36.9)	11(55.0)	4(36.4)
<i>Candida albicans</i>	66 (44.9)	24(36.9)	8(40.0)	6(54.9)
<i>Mucor spp.</i>	37 (25.2)	18(27.8)	6(30.0)	4(36.4)
<i>Proteus spp.</i>	33 (22.4)	9(13.8)	1(5.0)	0(0.0)
<i>Klebsiella spp.</i>	12 (8.2)	7 (10.8)	1(5.0)	0(0.0)
<i>Pseudomonas spp.</i>	11 (7.5)	4 (6.2)	1(5.0)	2(18.2)
<i>Enterobacter spp.</i>	8 (5.4)	4 (6.2)	1(5.0)	0(0.0)
<i>Rhizopus spp.</i>	5 (3.4)	0 (0)	0(0)	0(0.0)

n = total number of animals sampled per age group

pig production were poor sanitary conditions in piggeries, non-cleaning of the animals, lack of medical attention for most animals, irregular feeding and watering of the animals, feeding animals with stale food or kitchen wastes kept for up to four days, and improper storage (under damp conditions) of commercial feed by some farmers thereby encouraging the proliferation of organisms which can subsequently become infectious to the animals. Also, most farmers encountered were illiterates and had very little knowledge of good management systems for pigs.

The animal gut contains organisms that make up the normal microbiota. Under normal conditions, these organisms depend on each other and restrict each other. They play a nutritional role and restrict the multiplication of pathogens, but under stressed conditions, the animal can become infected since the gut microbial balance can be affected (Sholten *et al.*; 1999). Such factors include a high pathogen load in the gut environment, which could come from feed and water or from destruction of the micro-ecology balance by using broad-spectrum antimicrobials. Stress conditions can also be influenced by factors such as change of sheds (poor housing), overcrowding and poor diet (grossly contaminated feed, polluted water, irregular feeding and malnutrition), among others. Changes in gut microflora may cause problems in digestibility and absorption of dietary nutrients (Sholten *et al.*, 1999).

The presence of high numbers of pathogens and coliform bacteria in the gut environment of the animals under study was worrisome and suggested that the animals were under some stress conditions which, if not checked, could become problematic. Coliform bacteria and some pathogens produce toxins, which are harmful for the gastrointestinal mucosa and different strains of *E. coli* are known to cause diarrhoeal generally termed colibacillosis in animals (Horst, 1989; Gray, 1995). *Salmonella* species such as *S. typhimurium*, *S. enteritidis*, *S. agona* and *S. anatum* cause salmonellosis in animals, which is reportedly the most common widespread and important zoonotic infection in temperate regions (Sewel and Brocklesby, 1990). *C. albicans* can cause mucocutaneous candidiasis after a normal broad spectrum antibiotic therapy which distort the normal floral balance of the gut. *Mucor* and *Rhizopus* species can opportunistically be involved in zygomycosis in the gut. Pseudomonads and enterobacteria like *Proteus*, *Klebsiella* and *Enterobacter* species can cause tissue necrosis under stress conditions (Sewel and Brocklesby, 1990). In the US, the swine industry has suffered severe economic losses due to bacterial

infections, and *E. coli* was the reported leading cause of sickness and death in newborn and unweaned pigs (Sholten *et al.*, 1999). Profitable animal production demands efficient husbandry of healthy animals, as disease remains one of the profit limiting factors. Thus, the diagnosis and control of bacterial and fungal infections in these animals should also be considered paramount. However, it should be noted that the use of antibiotics to promote growth in animals is discouraged as it leads to the development of resistant strains. Fortunately, studies are underway to use herbs, spices and fermented liquid feeds to fight enteropathogens challenge in farm animals (Sholten *et al.*, 1999). Once these feeds are standardized worldwide, the problem of gastrointestinal infections in these animals may be resolved to a great extent (Sholten *et al.*, 1999).

It is interesting to note that microbial infestation of pigs can have enormous implications to public health. Human contamination with faecal flora of animals can result in infestations such as enteric fever caused by *Salmonella typhi* and *S. paratyphi*, and gastroenteritis due to *E. coli*. Humans can also come down with diseases like candidiasis and wound, urinary tract, respiratory and eye infections depending on the site of infection and the specific organism involved (Cheesbrough, 1984). The public can be infected by organisms in faeces through many ways (Cheesbrough, 1984; Duguid *et al.*, 1985; Achia and Boris, 1989). Infection can occur if:

- faeces –contaminated hands are used to prepare especially children's food, thus, farmers should maintain a high level of personal hygiene, particularly thorough hand-washing after handling or caring for the animals and before preparing food,
- flies land on contaminated faeces then settle on food or water which is to be consumed; thus, food and water should be protected from flies and animals,
- faeces contaminated fresh pork during slaughter and it is not properly washed and/or not well roasted or cooked before consumption,
- contaminates faeces are dumped or washed into water bodies used for drinking. In Dschang, springs and wells used for drinking are subject to possible contamination particularly by runoff or seepage. Thus, drinking water, especially that given to children, should be boiled or treated otherwise,
- untreated faeces contaminates vegetables which, when eaten raw (without proper washing) or improperly cooked, can be problem-

