

**Effects of physicochemical properties of fibrous feed on feeding behaviour and gut health  
of growing and finishing pigs**

by

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## **Declaration**

I, **Archibold Garikayi Bakare**, declare that this dissertation has not been submitted to any university and that it is my original work conducted under the supervision of Prof. M. Chimonyo.

All assistance towards the production of this work and all the references contained herein have been duly credited.

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## **List of abbreviations**

**ADF:** Acid detergent fibre

**ADFI:** Average daily feed intake

**ADG:** Average daily gain

**ADP:** Adenosine diphosphate

**AOAC:** Association of Analytical Chemists

**ATP:** Adenosine triphosphate

**BCM:** Barley-canola meal

**BD:** Bulk density

**CCN:** crossbred gilts fed on conventional diet

**CCO:** crossbred gilts fed on corn cob-based diet

**CD:** Crypt depth

**CF:** Crude fibre

**CK:** Creatine kinase

**CP:** Crude protein

**DDGS:** Distillers dried grains with soluble

**DE:** Digestible energy

**DF:** Dietary fibre

**DP:** Dietary protein

**EDTA:** ethylene-diamine-tetraacetic acid

**EE:** Ether extract

**FCR:** Feed conversion ratio

**GE:** Gross energy

**GH:** Grass hay

**GIT:** Gastro-intestinal tract

**GN:** Groundnut haulms

**HbG:** Glycated haemoglobin

**LC:** Lucerne hay

**MBW:** Metabolic body weight

**MC:** Maize cob

**MCN:** Mukota gilts fed on the conventional diet

**MCO:** Mukota gilts fed on corn cobs

**MS:** Maize stover

**NDF:** Neutral detergent fibre

**PU:** Dried citrus pulp

**RB:** Rice bran

**RTSD:** Relative time spent drinking to MBW of a pig

**RTSE:** Relative time spent on eating to MBW of pig

**SAS:** Statistical Analysis System

**SBP:** Sugar beep pulp

**SD:** Sawdust

**SH:** Sunflower hulls

**VH:** Villi height

**WB:** Wheat bran

**WHC:** Water holding capacity

**Effects of physicochemical properties of dietary fibre on feeding behaviour and gut health  
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**Abstract**

The broad objective of the study was to determine feeding behaviour and gut health of pigs fed different fibres at varying inclusion levels. Maize cob, maize stover, sunflower hulls, veld grass, sawdust, lucerne and dried citrus pulp were used in growing and finishing pig diets to provide a wide range of physicochemical properties. Time spent eating, drinking, lying down, sitting/standing and other activities was observed using video cameras. Blood samples were collected at the end of the trial for both growing and finishing pigs for analysis of glycated haemoglobin, albumin, globulin, total protein, creatine kinase, urea and uric acid. Intestinal segments were collected at the end of trial for growing pigs to determine mucosal architecture of the intestines. Digestible energy (DE), bulk density (BD), acid detergent fibre (ADF) and water holding capacity (WHC) were the most important variables predicting time spent on different behavioural activities in growing pigs ( $P < 0.001$ ). Water holding capacity, neutral detergent fibre (NDF) and ADF were the most important variables involved in predicting time spent on different behavioural activities in finishing pigs ( $P < 0.001$ ). Physicochemical properties influenced time spent on different behavioural activities and they provided relationships with time spent on different behavioural activities in both growing and finishing pigs. Glycated haemoglobin, albumin, globulin, total protein and uric acid were factors that influenced time spent eating in growing pigs ( $P < 0.05$ ). In finishing pigs, only serum total protein was selected

as the best predictor variable influencing time spent eating ( $P < 0.05$ ). The blood metabolites were correlated with time spent eating and drinking. They provided threshold values with time spent eating and drinking by pig. Hence, they can also be used as potential biomarkers that modulate neuronal pathways which reduce time spent eating and drinking. In this study, bulk density (BD) and NDF were the best predictor variables influencing villi height (VH) and apparent villi surface area (AVSA) in pigs ( $P < 0.05$ ). Grouped pigs fed fibrous diet spent more time eating, lying down, standing, walking and fighting ( $P < 0.05$ ). Skin lesions appeared the most on neck and shoulder region followed by chest, stomach and hind leg region, and finally head region ( $P < 0.05$ ). Fibrous diet did not reduce aggressive behaviours, rather aggressive behaviours emanated out of frustration when queuing at the feeder. It was concluded that physicochemical properties of fibrous diets and nutritionally-related blood metabolites influence feeding behaviour. Mucosal architecture was also influenced by physicochemical properties of the fibrous diets.

Keywords: Diurnal; Feeding behaviour; Mucosal structure; Pigs; Physicochemical properties.

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## Chapter 1: Introduction

### 1.1 Background

Pigs play an important role in alleviating animal protein demand for human consumption relative to most livestock species. This is mainly because of their fast growth rates, high feed conversion efficiencies and high prolificacy (Adesehinwa, 2008). With a widely held perception that pigs, unlike ruminants, do not subsist on fibrous feeds, thereby making them solely rely on whole grain as an energy source. An increase in population of pigs will, therefore, create competition for the limited feed resources (whole grains) with the ever-increasing human population (Ndindana *et al.*, 2002) and industries producing bio-fuels (Banse *et al.*, 2008). Thus, replacing whole grain with other energy sources might be a remedy to reduce competition.

Some of the potential substitutes for whole grain in feeds for pigs are crop by-products. These by-products include maize cob meal, maize stover, sunflower husks and soybean hulls, and wheat, barley and oat bran (Ndindana *et al.*, 2002; Wilfart *et al.*, 2007; Renteria-Flores *et al.*, 2008). They are easy to source, relatively inexpensive and readily consumed by pigs (Phuc *et al.*, 2000; Hogberg and Lindberg, 2004; Wilfart *et al.*, 2007). However, little is known about the influence of fibre sources and inclusion level on voluntary feed intake and behaviour of pigs. The lack of such information makes it difficult for stock-feed manufacturers to formulate feeds containing fibrous materials that will allow the optimum utilisation of nutrients (Whittemore *et al.*, 2001).

Fibre sources vary widely in their chemical and physical characteristics (King *et al.*, 2000; Dung *et al.*, 2002; Renteria-Flores *et al.*, 2008). Such variation is likely to influence voluntary feed intake and behaviour of the pigs. Water holding capacity (WHC), for example, influences bulkiness of feed. If the fibres have a high WHC, they absorb more water, making the fibres swell and occupy more space in the stomach. Consecutive stimulation of stretch receptors due to the distension of the gut leads to satiety, thus, affecting feeding behaviour as well as performance of pigs. Using more fibre sources will provide a wide range of physicochemical properties. This can increase level of accuracy for estimating relationships with time spent on different behavioural activities (feeding behaviour). Feeding behaviour is highly likely to be influenced by age of pig mainly because of different gut capacities (Kanengoni *et al.*, 2002).

When pigs are fed fibrous diets, they invoke certain behavioural activities that might be attributed to nutritional metabolites. Some of these nutritional metabolites include glucose, total proteins, albumin and urea. Glucose, for example, determines the time that pigs spent feeding. Transient declines in glucose levels usually precede meal initiation (Campfield *et al.*, 1996; De Leeuw *et al.*, 2004). Conversely, high levels of glucose in the circulatory system cause satiety, therefore, reduce the time that the pig spends feeding. Rate of absorption of these nutrients depends not only on the inclusion level of the fibre, but more importantly, on the physicochemical properties of the ingredient. Since dietary fibre sources have different physicochemical properties, it is highly likely that they have different fates and functional properties in the gastrointestinal tract. The differences may influence the concentrations of nutritional metabolites in blood. It is, therefore, important to determine whether there is a



relationship between nutritionally-related blood metabolites and time spent feeding by pigs fed on fibrous diets (Pambu-Gollah *et al.*, 2000; Ndlovu *et al.*, 2007).

Time spent on different behavioural activities provide information on preference of dietary fibre sources and also the internal state of an animal (Whittemore *et al.*, 2002). A negative effect on preference of dietary fibre sources and internal state of pig may be an indicator of poor welfare. Welfare of pigs can be assessed by offering pigs different fibres at varying inclusion levels to strike a balance between the loss in production and contentment of the pig.

Notwithstanding the role physicochemical properties of fibrous diets and blood metabolites play in controlling feeding behaviour, there is also a need to understand the mucosal structure of the intestines (villi height and crypt depth) of pigs. Feeding fibrous feeds is likely to change villi height and crypt depth (Jin *et al.*, 1994). Such changes affect the nutrient absorptive capacity of intestines, hence, normal behavioural activities. Information on how the physicochemical properties of fibrous diets affect structure of the intestines, however, remains sketchy and warrants further research.

## **1.2 Justification**

Competition for grain between humans and pigs is increasing. Such competition is instigated by the ever-increasing human population, and also crop failures due to persistent droughts and other environmental catastrophes. A possible remedy to reduce competition for grain can be the use of crop by-products such as maize cobs, maize stover, wheat straw, groundnut and soybean hulls, and wheat, barley and oat bran. Some authors argue that these crop by-products do not benefit

the pigs (Phuc and Lindberg, 2000; Chhay *et al.*, 2003), whilst others confirm their usefulness (Kanengoni *et al.*, 2002; Dégen *et al.*, 2007). When the fibres are used in diets of pigs, they should be included up to an optimum level where beyond that performance of pigs is not compromised. Optimum inclusion level of fibre can be estimated from physicochemical properties of fibre and also nutritionally-related blood metabolites which determine the time the pigs will stop eating due to satiation.

Maintenance of gut health is complex and relies on the diet offered to the pigs. Dietary fibres, for example, can interfere with the mucosa lining and gut microflora. Mucosa lining is highly likely to be influenced by physicochemical properties of fibres. Appropriate inclusion levels based on physicochemical properties should, therefore, be determined so as to maintain or improve gut health to prevent digestive dysfunction which may lead to poor growth. Changes in gut structure normally occur during the postnatal period and just after weaning. It is during this period when significant growth, morphological changes and maturation of the gastrointestinal tract is more pronounced (Xu, 1996; Trahair and Sanglid, 2002; Godlewski *et al.*, 2005). Hence, weaner pigs provide a perfect specimen for studying gut structure.

Following crop harvests, crop residues are normally burnt or thrown away. If these crop residues are to be incorporated into diets of pigs, a cheaper feed will be formulated. Feed compounders will cut feed costs; therefore, increasing the sustainability of pig enterprises.

### **1.3 Objectives**

The broad objective of the study is to determine the effect of physicochemical properties of fibrous diets on feeding behaviour and gut health of pigs.

The specific objectives are to:

1. Determine the effect of physicochemical properties of different fibre sources on time spent on different behavioural activities by growing pigs;
2. Determine the effects of nutritionally-related blood metabolites in growing pigs fed fibrous diets on time spent on different behavioural activities;
3. Determine the effect of physicochemical properties of fibrous diets on intestinal villi height and apparent villi surface area of growing pigs;
4. Predict the effect of physicochemical properties of different fibre sources on time spent on different behavioural activities by finishing pigs;
5. Determine relationships between nutritionally-related blood metabolites and different behavioural activities in finishing pigs fed on fibrous diets; and
6. Determine the effect of feeding fibrous diets on pig performance and aggressive behaviours in grouped pigs.

### **1.4 Hypotheses**

The following hypotheses will be tested:

1. Physicochemical properties of different fibre sources do not influence time spent on different behavioural activities by growing pigs
2. Blood metabolites do not influence time spent on different behavioural activities and nutritional status of growing pigs

3. Physicochemical properties of different fibre sources do not influence intestinal villi height and apparent villi surface area of growing pigs
4. Physicochemical properties of different fibre sources do not influence time spent on different behavioural activities by finishing pigs
5. Blood metabolites do not influence time spent on different behavioural activities and nutritional status of finishing pigs
6. Feeding fibrous diets does no influence pig performance and aggressive behaviours in grouped pigs.

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## **Chapter 2: Literature review**

### **2.1 Introduction**

To ensure viability of the pig enterprise, there is a need to identify cheap alternative feed resources. Introduction of a cheap alternative feed will require in-depth knowledge on their impact on growth performance, feeding behaviour, gut health and welfare. This chapter reviews information on common pig production systems, alternative feed sources, roles of dietary fibre on feeding behaviour, nutritionally-related blood metabolites and morphological characteristics of the gut of pigs.

### **2.2 Common pig production systems**

There are different types of breeds that are reared by farmers in the world. The pig breeds are reared under both extensive and intensive production. Choice of production system will determine the management practices to follow on the farm.

#### **2.2.1 Extensive production system**

The extensive production system is more common in communal farms (e. g. 13 % of the total population of pigs in South Africa) (DAFF, 2010). Although few pigs are reared under the system, it is now favoured by animal welfare activists because it increases the possibilities for the animals to graze the available natural pastures. Several researches on the extensive production system of rearing pigs have been conducted and have shown pigs to thrive well on the grasses (Edwards, 2003; Sterna and Andresen, 2003; Rodríguez-Estévez *et al.*, 2009). The production system also allows pigs to scavenge and wander freely to hunt for feed (Lekule and



Kyvsgaard, 2003). Furthermore, building costs for confined pig production have increased, making it economically rewarding to search for simpler, less capital-intensive systems.

### **2.2.2 Intensive production system**

The system is characterized by high capital investment requiring strong and expensive buildings and farmers buy feed for pigs until slaughter age. Most of the farmers have access to credit facilities which determine the level of herd size and capital intensiveness of the pig enterprise. Most of the pigs under intensive production system rely on maize-soya diets, resulting in serious competition for grains among humans and pigs for food, and bio-fuel producing industries (McCalla, 2009; Nonhebel and Kastner, 2011). The competition results in a rise in price of grain due to an increase in demand for the grain. For example, the price of grain in South Africa is shown in Table 2.1.

Optimum use of fibrous feedstuffs in pig diets can be a sustainable alternative to reduce the competition for grain. Their use under intensive production system can be an imitation of extensive production system where pigs have been observed to thrive well on grasses (Rodríguez-Estévez *et al.*, 2009).

### **2.3 Advantages and disadvantages of feeding fibrous feed materials**

The introduction of the fibrous material as feed for pigs may have some benefits and drawbacks. Knowledge of some of these provides valuable information to feed compounders on the type of fibre to use when formulating feeds.

**Table 2. 1: Ten year producer prices (US \$/tonne) of barley, maize, oats and sorghum in South Africa**

<b>Crop</b>	<b>Year</b>									
	<b>2002</b>	<b>2003</b>	<b>2004</b>	<b>2005</b>	<b>2006</b>	<b>2007</b>	<b>2008</b>	<b>2009</b>	<b>2010</b>	<b>2011</b>
Barley	113.8	189.4	208.3	183.1	234.3	196.1	278.4	227.9	126.6	314.1
Maize	129.5	123.4	129.7	99.2	145.9	205.9	201.6	156.2	137.4	212
Oats	90.1	145.4	163	145.7	145.6	232.3	248.7	155.3	296.8	273.8
Sorghum	142	191.6	202.6	70.9	177.1	210.6	214.7	179	189.1	231.5

### **2.3.1 Disadvantages of feeding fibrous diets**

Fibrous feed materials have a masking effect that hinders exposure of other nutrients to enzymatic digestion. Dietary nitrogen (Shi and Noblet, 1993; LeGoff and Noblet, 2001) and ether extract (LeGoff and Noblet, 2001) are made unavailable for digestion and absorption by fibres. Bindelle *et al.* (2005) reported that about 220 g/kg of nitrogen is bound to NDF in grasses. Unavailability of nitrogen due to binding by the fibres can result in low growth rates at high fibre inclusion levels (Glisto *et al.*, 1998; Mikkelsen *et al.*, 2004).

Fibrous feed material can either be soluble or insoluble. Soluble fibres increase viscosity of the digesta (Mosenthin *et al.*, 2001; Noblet and LeGoff, 2001). Increase in viscosity in the small intestine slows gut transit time due to suppressed intestinal contractions (Cherbut *et al.*, 1990). Intestinal contractions are necessary for mixing food in the gut, thus, increasing exposure to endogenous digestive enzymes.

Different fibres have different physicochemical properties. Water holding capacity of feed, for example, when in the stomach, swells occupying more space in the stomach. This causes distension of the stomach walls thereby inducing satiety. The pig will, therefore, not get all nutrients which meet requirements for growth. The extent to which the bulking effects of fibre can be incorporated in pig diets without compromising performance, behaviour, health and welfare of pigs is not known. Despite these disadvantages, inclusion of fibre in pig diets has several advantages.

### **2.3.2 Advantages of feeding fibrous diets**

Fibrous feed materials play a significant role in gastric emptying. Gastric emptying describes the time that feed takes to empty from the stomach and enter into the small intestines. Vestergaard (1997) showed reduced gastric emptying to prolong the feeling of satiety and thereby reduces the period of hunger. Reduced period of hunger will reduce stereotypies, aggression and competition for feed.

When the fibres are consumed by pigs, they are fermented in the large intestines (Glitsso *et al.*, 1998; Wenk, 2001). The fibres are fermented to form short chain fatty acids (SCFA), which generate about 17 % of the total digestible energy derived from the diet in growing pigs and over 25 % in sows (Shi and Noblet, 1993). Dietary fibre also creates an acidic environment in the gut which may inhibit the proliferation of pathogenic organisms, such as *Escherichia coli*, *Clostridium difficile* and *Salmonella* spp. (Cummings, 1983; May *et al.*, 1994; Montagne *et al.*, 2003).

Dietary fibre increases secretion of mucins in the gastro-intestinal tract (GIT) of animals (Montagne *et al.*, 2003). Mucus protects the GIT lining from invading organisms, and physical and chemical injury to the epithelium. It also reduces the incidence of gastric ulcers in non-ruminants (Lee and Close, 1987; Montagne *et al.*, 2003).

Having looked at some of the benefits of feeding the fibres, there is therefore, a need to select the type of fibre based on physicochemical properties which will not compromise performance and behaviour of pigs.

## 2.4 Alternative feed sources for pigs

Some of the potential feed sources that can be used to feed pigs are forages and, by-products from crops and timber industries. These include maize cob, maize stover, sunflower hulls, groundnut haulms, sawdust, rice bran, veld grass, lucerne, sawdust and citrus pulp. The fibre sources vary in their physicochemical properties (Table 2.2).

Maize cobs are remains left after removal of kernels from the cob. Maize stover, on the other hand, consists of the leaves and stalks of maize (*Zea mays*) plants left after removal of the cob. Groundnut haulms are pods left after removal of groundnut seeds. Sunflower hulls are by products from sunflower oil seeds after removal of kernels, whilst sawdust or wood dust is a by-product of cutting, grinding, drilling; it is composed of fine particles of wood. Veld grass and lucerne hay are plants that have been cut, dried, and stored for use as animal fodder.

Inclusion of the fibre sources in basal diets alters physicochemical properties of the feed. Adding fibre sources decreases crude protein (CP), ether extract (EE), ash and bulk density content of the diet (Kyriazakis and Emmans, 1995; Ndou *et al.*, 2012). On the contrary, crude fibre (CF), neutral detergent fibre (NDF), acid detergent fibre (ADF) and water holding capacity (WHC) content increases. Using different fibre sources will provide a wide range of physicochemical properties, which, invariably, contribute to bulkiness of feed, thus, influencing feeding behaviour of the pigs.

**Table 2. 2: Chemical composition (g/kg DM feed) and bulk characteristics of the fibrous feedstuffs**

<b>Components</b>	<b>GN</b>	<b>LC</b>	<b>MC</b>	<b>MS</b>	<b>RB</b>	<b>SD</b>	<b>SH</b>	<b>GH</b>	<b>PU</b>
Dry matter (DM) (g/kg)	882	811	948	947	810	956	941	940	896
Gross energy (MJ/kg DM)	22.9	17.7	17.8	16.8	19.9	19.1	19.7	17.4	17.3
Crude protein (g/kg DM)	117	189	52.2	53.4	190	44.1	59.5	60.2	70
Ether extract (g/kg DM)	21.3	12.2	7.5	5.1	19.9	16.9	63.9	15.9	24
Ash (g/kg DM)	27.9	86.6	40.8	61.5	72.5	6.2	33.1	70.8	69
<b>Bulk characteristics</b>									
Crude fibre (g/kg DM)	236	287	168	320	156.1	679	408	213	131
NDF (g/kg DM)	775	398	873	724	378	897	558	749	211
ADF (g/kg DM)	654	386	501	482	184	700	279	348	154
Bulk density (g DM/ml)	0.9	1.4	1.2	1	1.3	1.4	0.7	1.3	0.32
WHC (g water/g DM)	6.8	12.1	7.8	18.3	5.9	6.6	6.8	14	9.95

GN = groundnut haulms; LC = lucerne hay; MC = maize cob; MS = maize stover; RB = rice bran; SD = saw dust; SH = sunflower husks; GH = grass hay; PU = dried citrus pulp.

NDF = neutral detergent fibre; ADF = acid detergent fibre; WHC = water holding capacity.

**Source:** Ndou (2012); Gohl (1978); ADAS, 1992; Göhl (1981)

## **2.5 Behaviour of pigs**

Feeding behaviour can be defined as any action that is directed toward the procurement of nutrients. It can be recorded by using time as a unit of measurement. With the aid of video cameras, all the behavioural activities exhibited by the pig can be recorded for 24 hours. The use of video cameras paves way for acquiring unbiased behavioural data, that is, they offer an advantage in that they prevent disturbances to the pigs that occur due to the presence humans during recording. Feeding fibrous diets will affect time spent eating, stereotypic behaviours, time spent on different postural behaviours and aggressive behaviours in pigs kept in groups.

### **2.5.1 Effect of feeding fibrous diets on feeding behaviour of pigs**

Due to qualitative restriction of diets offered to pigs, it has been observed that pigs increase intake or spent more time eating fibrous feeds. This is evident in researches conducted using gilts, pregnant sows, multiparous sows and castrated males (Table 2.3). Pigs are obliged to eat more of the fibrous diets to get nutrients for meeting their requirements for growth and maintenance. Nutritional requirements for growth are, however, not met due to gut fill which brings about satiation in pig. Gut fill is influenced by bulk properties of the feed presented to the pig. For example, increasing fibre levels decreases bulk density of feed (Kyriazakis and Emmans, 1995; Ndou *et al.*, 2013). When the feed is consumed it occupies more space in the stomach hence gut capacity is reached resulting in reduction in time spent eating. Water holding capacity, on the other hand, causes feed in the stomach to absorb water, swell and occupy more space causing distension of the stomach. Inclusion level of fibre also compromise on texture of the feed. As more fibres are included in the diet, the feed becomes coarse.

**Table 2. 3: Time spent eating by different types of pigs subjected to fibrous diets**

<b>Pig type</b>	<b>Fibre diet</b>			<b>References</b>
	<b>Low</b>	<b>Medium</b>	<b>High</b>	
Gilts (min/ 2h)	0.49	-	0.58	McGlone and Fullwood (2001)
Gilts (min/ meal)	24.9	-	56.7	Robert <i>et al.</i> (2002)
Pregnant Sows (min)	16.4	24.3	51.6	Ramonet <i>et al.</i> (1999)
Multiparous sows %/ 2h	16.4	-	31.0	Whittaker <i>et al.</i> , (1999)
Castrated male pigs	Low	-	High	Kallabis and Kaufmann (2012)



### **2.5.2 Effect of feeding fibrous diets on stereotypic behaviours of pigs**

Stereotypies are behaviours that are relatively invariant, regularly repeated, and without an obvious function (Odberg, 1978). Stereotypies may indicate that an environment is deficient in one or more aspects for an individual or species (Mason, 1991). They are induced by drugs, diet and also environment conditions animals are subjected to. With regards to diet offered to pigs, several studies have revealed fibres to reduce stereotypies (Whittaker *et al.*, 1999; Bergeron *et al.*, 2000, Danielsen and Vestergaard, 2001; Robert *et al.*, 2002). It has been suggested that part of the effect of fibrous diets on stereotyped behaviour may result from the increased time taken to eat the fibrous meal and the consequent shorter period of time available to perform such behaviour.

### **2.5.3 Effect of feeding fibrous diets on postural behaviours of pigs**

Feeding of fibrous diets has a bearing on the way pigs exhibit various postures. Table 2.4 shows proportions of time spent on various postures in different types of pigs fed fibrous diets. Pigs normally lie down to rest and sleep. This is very crucial, especially for juvenile pigs for it allows anabolic hormones such as growth hormones to be secreted, thus, allows growth of pigs. Juvenile pigs, therefore, have been reported to spend approximately 80 % of their time lying down (Ekkel *et al.*, 2003). Apart from metabolic processes which are instigators of sleep, other factors have also been reported, which make pigs to lie down. These factors include nature of diet the pig is being subjected to and environmental conditions (ambient temperature).

**Table 2. 4: Time spent on different postures by pigs fed fibrous diets**

Pig type	Posture activity	Fibre diet		References
		Low	High	
Gilts (min/ 2h)	Standing	3.43	3.52	McGlone and Fullwood (2001)
	Sitting	0.02	0.002	
	Lying down	105.5	107.6	
Gestating sows (day 40) %/day	Standing	75	77.6	Holt <i>et al.</i> (2006)
	Sitting	1	5.2	
	Lying down	24	17.2	
Gestating sows (day 80) %/day	Standing	60.6	60.2	Holt <i>et al.</i> (2006)
	Sitting	3.3	8.4	
	Lying down	35.1	31.4	
Pregnant sows (min / 2hr)	Standing	3.8	2.4	Whittaker <i>et al.</i> (1999)
	Lying down	93.9	116.6	

Day 40 - behavioural activities recorded after 40 days

Day 80 - behavioural activities recorded after 80 days

With regards to nature of diet being offered to pigs, satiety is induced by gut fill, especially in pigs that are fed high fibrous diets. High fibrous diets have high proportions of indigestible components (NDF, ADF and lignin). Pigs are, therefore, likely to spend more time lying down and resting in a bid to expend more energy towards digestion and clearing of some of indigestible components from the gut.

Lying down can be a state of boredom especially when there is limited space for allowing an animal to wander freely and also when the fibrous feed is less palatable (Pearce and Paterson, 1993). They also exhibit the postures of sitting and standing when they are showing levels discomfort (Pearce and Paterson, 1993). Rolandsdotter *et al.* (2009) describes the postures to be exhibited by the pigs to reduce the risk of shoulder lesions especially when there is hard floor.

#### **2.5.4 Effect of feeding fibrous diets on behaviours of grouped pigs**

Under commercial farming practices, group housing is more prominent. The grouping system of rearing pigs offers economic and management benefits. These benefits include suitability for all-in-all-out production which facilitates health management, the use of labour-saving machinery and bedding systems, and low capital investment in buildings and infrastructure (Turner *et al.*, 2003). Nevertheless, pigs reared in groups exhibit aggressive behaviours more often as they try to establish ranks while competing for limited resources (feed and water). Feeding of fibrous diets can bring about changes to such aggressive behaviours. Several studies have observed feeding of the fibrous diets by pigs to increase more time towards ingestion process (Martin and Edwards, 1994; Meunier-Salaun *et al.*, 2001).

Whittaker *et al.* (1999) pointed out that provision of supplementary fibrous feed can reduce the level of vulva biting in sows. More time spent eating results in less time spent on other behavioural activities (aggressive behaviours in particular). Apart from bulk properties of fibrous diets, nutritionally-related blood metabolites also play an important role in controlling feeding behaviour and are also an indicator of nutritional status of pigs.

## **2.6 Effect of feeding fibrous diets on nutrient metabolism**

Nutritionally-related blood metabolites interact with the central nervous system (CNS) to modulate different behavioural patterns in pigs. The nutritional-related blood metabolites can also give an immediate indication of the animals' nutritional status (Pambu-Gollah *et al.*, 2000). Some of the blood metabolites likely to modulate different behavioural patterns and nutritional status include glucose, serum proteins, urea and uric acid and creatine kinase.

### **2.6.1 Glucose**

Glucose influences the behaviours exhibited by pigs (De Leeuw, 2004; Lebret *et al.*, 2006). Glucose is the major source of energy for the body and has been observed to be a major contributor to satiety. Decline in glucose levels usually evoke meal initiation. On the contrary, increase in levels of glucose induces satiety. Mechanism by which glucose influences food intake is known as hyper-insulinemia. Hyper-insulinemia is a condition in which there are excess levels of insulin circulating in the blood than expected relative to the level of glucose. Because glucose is highly unstable in the circulatory system of the animal, glycated haemoglobin (HbG) is stable indicator of glucose status and can be used for determining time spent eating. Levels of HbG have been reported in white-tailed deers (Jenks *et al.*, 1991), camels (Al-Ali *et al.*, 1990),

ruminants (Alayash *et al.*, 1988; Shahbazkia *et al.*, 2010) and birds (Rendell *et al.*, 1985; Shahbazkia and Nazifi, 2008). Very few, if any, studies have been done to determine the relationship between HbG and time spent feeding on maize cob, maize stover, sunflower hulls, groundnut haulms, sawdust, veld grass, lucerne, sawdust and citrus pulp in pigs. Glycated haemoglobin is formed when blood glucose diffuse and bind irreversibly with amino groups of haemoglobin (haemoglobin N-terminal or lysine side chain amino groups). Glycated haemoglobin concentration can be used as measurement of the mean blood glucose level over the previous several weeks and is not affected by recent stress, drug use or physiological (Shahbazkia *et al.*, 2010). In a study conducted with Mukota (Zimbabwean indigenous pig) and Landrace × Mukota gilts fed diets with graded levels of maize cob (Mashatise *et al.*, 2005; Table 2.5), genotype and diet did not affect serum glucose concentrations. Other studies (Leclere *et al.*, 1993; Yoon *et al.*, 2010) have also observed increasing levels of fibre not to affect blood glucose concentrations.

### **2.6.2 Serum proteins**

Serum proteins can be used to measure protein status in animals. If protein is lacking in a diet, animal serum protein levels are lowered in the blood (Ndlovu *et al.*, 2007). Conversely, if dietary protein is high serum protein levels in blood are elevated. Inclusion of fibre in pig diets lowers protein levels. For example, indigestible components of fibrous material (NDF and ADF) have a masking effect that hinders nitrogen from being available for use by the animal (Bindelle *et al.*, 2005). Hence, serum protein levels are lowered. Serum protein levels can also be a consequence of reduced urinary excretion and/or protein catabolism caused by stress in pigs subjected to high fibrous diets (Zervas and Zijlstra, 2002).

**Table 2. 5: Effects of breed and fibrous diet on glucose concentration (mmol/L)**

Week	Treatments			
	CCO	MCN	CCN	MCO
0	4.34	4.46	5.06	4.84
2	4.44	4.88	5.36	4.80
4	4.74	4.24	4.88	4.26
6	4.32	4.74	4.30	4.34
8	4.95	4.08	5.00	4.80
10	4.54	4.32	4.33	4.76
12	4.92	4.21	5.02	4.78
14	4.90	4.44	4.87	4.76
SE	0.483	0.461	0.480	0.462
N	5	5	5	5

CCO: crossbred gilts fed on corn cob-based diet; CCN: crossbred gilts fed on conventional diet; MCO: Mukota (Zimbabwean indigenous pig) gilts fed on corn cobs; MCN: Mukota gilts fed on the conventional diet.

**Source:** Mashatise *et al.* (2005)

Apart from diet offered to the pigs, dehydration has also been reported to cause fluctuations in serum protein levels in the blood. High concentrations of the plasma proteins are an indication of dehydration due to increased plasma osmolality (Deaux *et al.*, 1970).

### **2.6.3 Urea and uric acid**

Urea is an organic compound which serves an important role in the metabolism of nitrogen-containing compounds by pigs. Concentration of urea in blood of pigs varies with breed and also nature of diet and time. In a study conducted with Mukota and Landrace × Mukota gilts fed diets with graded levels of maize cob (Mashatise *et al.*, 2005; Table 2.6), there was a significant interaction between genotype, diet and time on serum urea concentration. Higher urea levels were observed in Mukota gilts than in the crossbreds from week 8 up to the end of the trial.

If fibrous diets are offered to pigs, urea is used to synthesize microbial proteins. The microbes facilitate the breakdown of fibrous material in the large intestines (Columbus *et al.*, 2012). These fibres are thereafter converted to volatile fatty acids which serve as an energy source for the pig. If the diet does not have sufficient protein to produce microbes for breaking down fibres in the colon, the pig respond by breaking down its body tissue protein to produce urea. The urea cannot be excreted as waste. Instead, can be recycled and enters the gastro-intestinal tract from the saliva and walls of the intestines (Fuller *et al.*, 1998). Production of urea by the pig can be an adaptation to diet being offered, particularly fibrous diets.

**Table 2. 6: Effects of breed and fibrous diet on serum urea concentrations (mmol/L)**

<b>Week</b>	<b>Treatments</b>			
	<b>CCO</b>	<b>MCN</b>	<b>CCN</b>	<b>MCO</b>
0	2.98	4.32	3.02	3.82
2	3.84	5.84	4.88	4.66
4	3.58	3.97	3.68	3.09
6	4.42	3.24	5.14	4.10
8	2.73	3.77	4.24	4.51
10	2.78	4.70	4.12	4.53
12	2.76	4.87	3.98	4.56
14	2.74	5.56	3.99	4.58
SE	0.544	0.691	0.543	0.692
N	5	5	5	5

CCO: crossbred gilts fed on corn cob-based diet; CCN: crossbred gilts fed on conventional diet;

MCO: Mukota gilts fed on corn cobs; MCN: Mukota gilts fed on the conventional diet.

Source: Mashatise *et al.* (2005)



If pigs are offered diets high in fibrous material, they are expected to produce more saliva with urea to facilitate in the digestion process. This can be a sign that the animal is adapting to the diet being offered and performance is likely not to be dented. High concentrations of uric acid (hyper-uraemia) can occur when an animal has been starved, which result in massive tissue destruction (Chandra *et al.*, 1983).

#### **2.6.4 Creatine kinase**

Creatine kinase (CK) is a "leakage" enzyme present in high concentration in the cytoplasm of myocytes. The function of enzyme is to make ATP available for contraction by the phosphorylation of ADP from creatine phosphate by catalyzing the reversible phosphorylation of creatine by ATP to form phosphocreatine + ADP. Creatine kinase concentrations vary due to animal's diet, breed, muscle mass and sex (Hammond *et al.*, 1994; Ndlovu *et al.*, 2007). Animals with more muscle are likely to produce more creatine kinase in the blood. Muscle activity may also increase serum CK. For example, feeding fibrous diets increases time spent chewing, hence more CK levels are likely to be produced due to the chewing activity. Bonca *et al.* (2010) has also reported CK to be an indicator of stress in pigs with high concentrations indicating that an animal is stressed. Feeding pigs less palatable feeds (fibres) is likely to induce stress compared to palatable feeds. Absorption of nutrients from the gut, which subsequently determine concentration of glucose, serum proteins, urea and uric acid and creatine kinase depends on morphology of the intestinal walls. Morphology of the intestines, on the other hand, is altered by the diet consumed by the pig.

## **2.7 Effect of feeding fibrous diets on gut health of pigs**

Dietary fibre can influence gut health of pigs through several potential mechanisms. By increasing size and weight of the GIT (including the organs), changing the gut structure (villi height and crypt depth, increasing microbes in the GIT and also altering enzyme activities on the mucosa lining. The changes provide information on how the pig is adapting to the particular diet to better utilise the available nutrients for them survive. With regards to morphological characteristics of the GIT including the mucosa lining (Villi height and crypt depth), changes start to occur in newly born piglets when they are introduced to solid feeds (weaner phase) leading to the formation of specialized intestinal epithelium that takes part in digestion and absorption of nutrients (Barszcz and Skomia, 2011).

### **2.7.1 Effect of feeding fibrous diets on size and weight of the gastro-intestinal weight**

Several studies have reported hypertrophy of the gastrointestinal tract (GIT) of pigs when fed diets high in fibre compared to diets with low fibre content (Table 2.7). Increase in weight of the pancreas might be attributed to hyper-secretion of pancreatic enzymes in an attempt to compensate for digestion and absorption inefficiency (Wenk, 2001). At high inclusion levels of fibre, more enzymes are produced to breakdown coarse textured feed compare to lower fibre diets.

**Table 2. 7: Effect of feeding fibrous diets on organ weights of pigs**

Organ	Fibre sources									
	DDGS <sup>1</sup>		BCM <sup>2</sup>		BCM –ALF <sup>2</sup>		WB <sup>3</sup>		SBP <sup>3</sup>	
	Low	High	Low	High	Low	High	Low	High	Low	High
Liver	3.49	3.62	24.8	28	24.8	28	3.2	3.2	3.2	2.9
Pancreas	0.30	0.29	-	-	-	-	0.2	0.2	0.2	0.3
Stomach	0.95	1.02	-	-	-	-	0.8	0.9	0.8	0.9
Caecum	0.30	0.28	2.01	2.22	2.01	2.92	-	-	-	-
Small intestines	4.25	4.6	31.2	30.1	31.2	35.5	4.5	5.7	4.5	4.7
Large intestines	1.75	2.14	10.1	15.5	10.1	17.5	2.0	2.5	2.0	2.5
Units of measurements	% empty body		g /kg empty body weight				Relative weight (%)			

DDGS: Distillers dried grains with soluble; BCM - Barley-canola meal; BCM –ALF - Barley-canola meal and alfalfa; WB – wheat bran; SBP – sugar beep pulp;

References: 1. Agyekum *et al.* (2012)

2. Nyachoti *et al.* (2002)

3. Ma *et al.* (2002)

Ma *et al.* (2002), on the other hand, reported a decrease in liver weights in growing pigs fed on diets with sugar beet pulp. This suggests that organ weights are influenced by the source of the insoluble fibre used, which, subsequently, influences the ability of the pig to get enough energy for protein accretion (Pond *et al.*, 1988; Jørgensen *et al.*, 1996). Information on how the visceral organs change as pigs are subjected to the fibrous diets still remain vague, hence, warrants further investigations.

### **2.7.2 Effect of feeding fibrous diets on mucosal architecture of pigs**

Fibrous diets alter morphology of intestinal villi and crypt depth in pigs. Changes in morphology of intestinal villi and crypt depth are an indication that the pig is adapting to a diet (Caspary, 1992; Dong and Pluske, 2007). Increase in size of villi will enable more nutrients to be absorbed due to increased surface area from the lumen of the gut into the circulatory system, especially at higher inclusion level of fibre when diet is considered to be of low nutrient density. Fibre source and segment of the intestines influence size of the villi (Table 2.8). Agyekum *et al.* (2012) and Halimani *et al.* (2006) observed villi height to decrease as inclusion level of fibre increased.

Nyachoti *et al.* (2006), on the other hand, observed the villi height to increase with an increase in dietary fibre in the jejunum and ileum segment in weaner pigs. Jin *et al.* (1994) found no changes in size of the villi. Nevertheless, different conclusions were drawn on mucosa measurement of the intestine of the pigs subjected to different fibre sources, indicating that different fibres have different physicochemical properties. These physicochemical properties might influence size of villi in pigs, hence a single conclusion regardless of fibre source but based on physicochemical

**Table 2. 8: Effect of feeding fibrous diets on mucosal architecture of pigs**

Segment		Treatments									
		DDGS <sup>1</sup>		Decreasing DP and increasing DF <sup>2</sup>				<i>Colophospermum mopane</i> <sup>3</sup>			
		Low	High	Low	Medium	High	Very High	Low	Medium	High	Very High
Duodenum	VH $\mu$ m	509.9	505.8	583.0	550.3	528.8	532.6	738	556	691	714
	CD $\mu$ m	339.6	334.4	192.4	181.4	191.8	167.9	457	369	421	408
	VH:CD	1.52	1.52	3.10	3.06	2.79	3.25	1.6	1.5	1.6	1.8
Jejunum	VH $\mu$ m	470.4	453.1	482.1	518.8	450.5	541.7	742	536	669	687
	CD $\mu$ m	273.6	286.2	182.9	186.6	198.6	168.8	470	384	391	401
	VH:CD	1.75	1.64	2.67	2.84	2.27	3.26	1.6	1.4	1.7	1.7
Ileum	VH $\mu$ m	480.3	408.9	426.9	480.0	421.3	445.2	603	550	559	593
	CD $\mu$ m	307.3	298.4	168.9	170.0	183.4	160.3	434	418	384	387
	VH:CD	1.58	1.38	2.58	2.96	2.35	2.79	1.4	1.4	1.5	1.5

DDGS: Distillers dried grains with soluble; DP – dietary protein; DF – dietary fibre; VH – villi height; CD – crypt depth;

VH:CD - villi height-crypt depth ratio

References: 1. Agyekum *et al.* (2012); 2. Nyachoti *et al.* (2006); 3. Halimani *et al.* 2006

properties can be drawn. Very few, if any studies, have been conducted to relate physicochemical properties of the different fibre source and mucosal architecture.

### **2.7.3 Effect of body weight on mucosal architecture of pigs**

Apart from diet which influences size of villi, body weight has also been observed to have an effect. An increase in body weight results in an increase in villi height up to a point where no further increase will occur (Zambonino Infante *et al.*, 2013). Heavier pigs are likely to have villi greater in size compared smaller pigs.

## **2.8 Summary**

Fibre sources can be used as an alternative energy source to feed pigs. There is, however, a need to know the acceptability and adaptability by the pigs to the fibre sources. This can be done by assessing feeding behaviour, nutritional status and also morphometrical changes of the gastro-intestinal tract. Of particular importance when looking at feeding behaviour and gut morphometrical changes (i.e. villi height and crypt depth) are physicochemical properties of the fibre sources. Physicochemical properties of fibre sources are likely to have a greater influence on feeding behaviour and changes in villi height and crypt depth. Using physicochemical properties instead of using inclusion levels increase level of accuracy when predicting feeding behaviour gut morphometrical changes. The broad objective of the study was, therefore, to determine the effect of physicochemical properties of fibrous diets on feeding behaviour and gut health of pigs. This allows feed compounders to determine optimum inclusion levels of fibre in a diet based on physicochemical properties, which will not compromise on the behaviour and performance of pigs.

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### **Chapter 3: Influence of physicochemical properties of fibrous diets on behavioural reactions of individually housed pigs**

*(Published in Livestock Science: Appendix 1)*

#### **Abstract**

The objective of the study was to predict time spent on different behavioural activities of individually housed growing pigs from physicochemical properties of feeds. Maize cob, maize stover, sunflower hulls, veld grass, sawdust and lucerne were used to provide a wide range of physicochemical properties. The fibre sources were included at 0, 80, 160, 240, 320, 400 g/kg inclusion levels in pig diets. Time spent eating, drinking, lying down, sitting/standing and other activities was observed using video cameras. Pigs spent most of their time lying down (71.4 %) followed by time spent eating (23 %), drinking (3.2 %) and sitting/standing (2.4 %). Digestible energy (DE), bulk density (BD), acid detergent fibre(ADF) and water holding capacity (WHC) were the most important variables for predicting time spent on different behavioural activities ( $P < 0.001$ ). Bulk density and ADF produced linear responses with time spent eating and drinking ( $P < 0.001$ ). There was a quadratic response between time spent lying down and ADF content of feed ( $P < 0.001$ ). Water holding capacity was the most important physicochemical property of feeds for predicting number of visits made by the pig to the feeder. Total time spent on each visit by a pig per day was best predicted by ADF. In conclusion, physicochemical properties of diets alter behaviour of penned growing pigs.

**Keywords:** Dietary fibre; Feeding Behaviour; Growing Pigs



### 3.1 Introduction

The search for cheap alternative feed ingredients for pigs (*Sus scrofa*) is receiving increasing attention mainly because of the increased global demand for grains for both human and livestock consumption (McCalla, 2009). Alternative feed ingredients that can be used to feed pigs are crop by-products; maize cob, maize stover, sunflower husks and, also grasses and sawdust. These fibre sources are readily available, cheap and can be consumed by pigs (Owen and Ridgman, 1967; 1968; Ndindana *et al.*, 2002; Ferguson *et al.*, 2003). Fibres have been reported as diluents in previous studies (Whittemore *et al.*, 2002; Len *et al.*, 2007). Their use, depending on the inclusion level in a diet, improves welfare of pigs by reducing stereotypic behaviours, chronic colitis, dysentery and constipation (Filer *et al.*, 1986; Day *et al.*, 1996; Metzler and Mosenthin, 2008; Thomson, 2009).

As fibre content of diets increase, the dilution of the resultant diets alters the time that pigs spend on various behavioural activities. For example, pigs fed on fibrous diets have an increased motivation to eat (D'Eath *et al.*, 2009). Ramonet *et al.* (1999) reported that increasing fibre level in sow diets increases time spent eating. High-fibre diets also increased time spent eating in non-pregnant nulliparous gilts (Robert *et al.*, 1997). Behavioural reactions of growing pigs subjected to the fibrous diets, however, remain unclear. Apart from assessing feed intake and growth performance, behavioural responses of the pigs need to be considered in the determination of appropriate fibre inclusion levels.

Physicochemical characteristics of fibrous feeds might play a key role in influencing behaviour of pigs. The physicochemical properties of fibrous feeds include water holding capacity (WHC), bulk density (BD), acid detergent fibre (ADF), neutral detergent fibre (NDF) and crude fibre (CF). The physicochemical characteristics vary across fibre sources

(Ndou, 2012). Fibres with a high WHC, for example, will absorb more water, swell and occupy more space in the stomach. Consecutive stimulation of stretch receptors due to distension of the gut will lead to satiety (Lepionka *et al.*, 1997; De Leeuw, 2004). Satiety is the state of being satisfactorily full and unable to take on more feed. This affects feeding behaviour and, consequently, performance of growing pigs.

Although physicochemical properties have been demonstrated to predict feed intake (Kyriazakis and Emmans, 1995; Ndou *et al.*, 2012), their effect on time spent on different behavioural activities in growing pigs is still unclear. For example, Whittemore *et al.* (2002) conducted a study with few fibrous sources with a narrow range of physicochemical properties. Use of more fibrous feedstuff at varying inclusion levels provides a wider variation of physicochemical properties to generate more accurate predictions of behavioural activities exhibited by growing pigs. For close monitoring of pigs and to avoid the effects of association which can affect normal behavioural activities when in groups, it is necessary to house pigs individually. Time spent on different behavioural activities might be a first step in providing an insight on how specific physicochemical characteristics of different fibre sources affect satiety levels in pigs. The objective of the study was, therefore, to predict the influence of physicochemical properties of feeds on time spent on different behavioural activities in growing pigs; the hypothesis stated that there is no relationship between the physicochemical properties and time spent on different behavioural activities.

## **3.2 Materials and methods**

### **3.2.1 Pigs and housing**

A total of 124 clinically-healthy, castrated, male growing pigs were used. The care and use of the animals was performed according to the Certificate of Authorization to Experiment on

Living Animals provided by the University of KwaZulu-Natal (Reference Number: 061/12/ Animal)(Appendix 2). Initial body weight for the pigs used in the experiment was  $22.4 \pm 3.12$  kg. Each pig was used as an experimental unit. They were reared in individual pens measuring  $1.5 \times 1$  m and had a slatted floor. The space in the pens allowed the pigs to move around and have limited exercise. The pens contained a plastic tube feeder (Big Dutchman Lean Machine®) and a low-pressure nipple drinker which provided clean water ad-libitum. The individual pens were not in total isolation because a pig had visual contact with the pig in adjacent pens and could hear and communicate with each other. Pigs were reared in individual pens for five weeks including the adaptation period of one week. The ambient temperature and relative humidity were recorded automatically throughout the trial using HOBO data loggers (Onset Computer Corporation, Pocasset, MA, USA). The average temperature and humidity were  $21.15 \pm 2.74^{\circ}\text{C}$  and  $42.64 \pm 1.56$  %, respectively.

### **3.2.2 Diets**

A premium commercial feed (Express Weaner, Meadow feeds Ltd) with a low level of crude fibre (CF) was used as the basal feed. Six fibre sources, namely maize cob, maize stover, sunflower hulls, grass hay, lucerne and saw dust were selected to dilute the basal diet. Maize cobs used in the experiment were remains left after removal of kernels from the cob. Maize Stover, on the other hand, consisted of the leaves and stalks of maize (*Zea mays*) plants left after removal of the cob. Sunflower hulls were by-products from sunflower oil seeds after removal of kernels whilst sawdust or wood dust were by-product of cutting, grinding, drilling; that composed of fine particles of wood. Grass hay and lucerne were plants that have been cut, dried, and stored for use as animal fodder. All the fibre sources were used in the experiment to increase the range of physicochemical properties. Maize cob, maize stover, sunflower hulls, grass hay and lucerne hay were ground to pass through a 3 mm screen. The

fibre sources were then included at different inclusion levels of 0, 80, 160, 240, 320 and 400g/kg in diet of pigs. It was assumed that the pigs would increase their feed intake proportionately to the degree of dilution of the basal feed with the fibre source. Hence, all pigs would get nutrients that met requirements for growth. Each of the thirty-one diets (i.e. one control diet and 30 fibre-based diets) was fed to four randomly selected pigs in their own pens.

### **3.2.3 Analyses of physicochemical properties of diets**

Tables 3.1, 3.2 and 3.3 show composition of ingredients and physicochemical properties of diets used in the experiment. Ash was determined by combusting the sample in a furnace for 4 h at 550°C according to method 942.05 of AOAC (2005). The dry matter (DM) was determined in an oven at 65°C for 48 h according to the method 2001.12 of AOAC (2005). Crude protein (CP) was determined by using a Truspec N analyser (Leco, MI, USA) based on Dumas Combustion method 990.03 (AOAC, 2005). Crude fibre (CF) was determined according to the method 2001.12 of AOAC (2005). Neutral detergent fibre (NDF) and acid detergent fibre (ADF) were analysed using filter bags by means of a fibre analyzer (Ankom 220; Ankom Technology Corp., Macedon, NY, USA). Gross energy (GE) was determined using a bomb calorimeter. Digestible energy (DE) of feeds was calculated as:  $DE = 949 + (0.789 \times GE) - (43 \times \% \text{ ash}) - (41 \times \% \text{ NDF})$  (Noblet and Perez, 1993). Bulk density of the feeds was measured according to the water displacement method described by Peterson and Baumgardt (1971). The method was based on the Archimedes Principle of determining the volume of a known mass of feed ingredient.

**Table 3. 1: Composition of ingredients in a basal diet**

<b>Ingredients</b>	<b>Composition (%)</b>
Yellow maize whole	42.6
Full fat soya	17.6
Whole wheat	10.0
Wheat bran middlings	10.0
Soyabean oil cake	8.4
Sunflower oil cake	7.5
Fish meal	2.0
Vitamin mineral mix	1.9

**Table 3. 2: Nutritional composition of diets with different inclusion levels of Lucerne, Sawdust, Sunflower Hulls, Grass hay, Maize cob and Maize stover**

Inclusion level (g/kg)	Fibre source	Nutritional composition				
		DM (g/kg)	<sup>1</sup> calc DE (MJ/kg) <sup>1</sup>	CP (g/kg DM)	EE (g/kg DM)	Ash (g/kg DM)
0	B	989.3	13.9	195.7	52.9	61.1
80	LC	989.3	13.4	192.6	49.2	63.1
	SD	989.2	12.3	117.4	51.5	53.7
	SH	989.3	13.5	166.9	54.1	56.6
	GH	989.5	13.1	186.4	51.7	60.4
	MC	989.3	13.2	185.7	51.2	59
	MS	990.1	13.8	178.6	50.1	62.3
160	LC	990	12.6	187.6	45.2	65.1
	SD	989.7	11.2	164.5	47.7	48.3
	SH	989.4	12.8	145.7	55.4	53.8
	GH	990	12.3	145.3	49.3	61.9
	MC	990.4	12.1	168.1	45.9	54.9
	MS	990	13.0	169.8	44.9	64.2
240	LC	990	11.8	174.3	41.5	67.2
	SD	989.7	10.2	145.8	45.55	44.1
	SH	989.6	12.2	134.6	55.7	51.6
	GH	990.5	11.2	135.5	40.7	63.9
	MC	990.5	11.0	152.8	45.2	53.2
	MS	989.9	12.0	147.8	39.3	65.3
320	LC	989.9	10.9	168.5	39.2	69.2
	SD	989.7	9.2	131.3	42.35	41.6
	SH	989.7	11.6	132.1	56.6	48.5
	GH	990.5	10.2	120.8	36.6	64.9
	MC	990.2	10.3	139.4	41.3	52.1
	MS	989.8	11.0	143	37.1	66.4
400	LC	989.9	10.2	165.9	33.3	71.3
	SD	989.8	8.1	117.5	36.7	39.2
	SH	989.8	11.3	123.5	57.3	44.9
	GH	990.8	9.2	112.9	32.4	65.9
	MC	991.1	9.3	116.4	39.9	46.7
	MS	989.8	10.1	141.3	31.9	66.5

B – Basal diet; LC – Lucerne; SD – Sawdust; SH – Sunflower Hulls; GH – Grass hay; MC – Maize cob; MS – Maize stover; DM – Dry matter; GE – Gross energy; CP – Crude protein; EE – Ether extract; calc DE – calculated digestible energy

$$^1\text{calc DE} = 949 + (0.789 \times \text{GE}) - (43 \times \% \text{Ash}) - (41 \times \% \text{NDF}) \text{ (Noblet and Perez, 1993)}$$

**Table 3. 3: Composition of physical properties of diets with different inclusion levels of Lucerne, Sawdust, Sunflower hulls, Grass hay, Maize cob and Maize stover**

Inclusion level (g/kg)	Fibre source	Physical properties				WHC (g water/g DM)
		CF(g/kg DM)	NDF(g/kg DM)	ADF(g/kg DM)	Density (g DM/ml)	
0	B	26.1	192.3	88.4	1.45	3.76
80	LC	46.1	218.1	101.2	1.54	3.65
	SD	77.4	257.1	128.2	1.54	3.41
	SH	55.6	223.1	94.5	1.49	3.16
	GH	40.1	230.3	121.8	1.51	3.76
	MC	36.4	230.6	111.5	1.52	3.17
	MS	48.6	192.3	110.7	1.49	4.15
160	LC	67.6	261.1	126.4	1.46	4.26
	SD	129.7	318	177.7	1.49	3.95
	SH	86.3	269.8	110.7	1.44	3.48
	GH	55.1	284.2	166.7	1.5	4.28
	MC	47.9	294.4	147.4	1.47	3.57
	MS	72.2	228.1	142.9	1.31	5.24
240	LC	87.9	301.9	150.7	1.39	5.04
	SD	182.1	378.8	227.4	1.43	4.27
	SH	116.9	316.9	126.2	1.33	3.76
	GH	70.2	337.6	214.3	1.47	5.14
	MC	59.3	355.3	181.2	1.42	4.08
	MS	95.8	279.8	175.2	1.3	6.48
320	LC	108.9	343.9	175.5	1.32	5.56
	SD	234.4	439.8	277.3	1.4	4.79
	SH	147.6	363.3	142.1	1.31	4.06
	GH	85.2	391.2	259.4	1.31	6.45
	MC	70.8	401.3	218.4	1.25	4.41
	MS	119.4	332.3	207.4	1.12	7.49
400	LC	129.9	385.7	200.2	1.27	6.52
	SD	286.8	500.8	326.6	1.31	5.21
	SH	178.2	409.6	157.8	1.21	4.38
	GH	100.3	444.1	304	1.2	7.13
	MC	82.2	457.4	250.5	1.22	4.75
	MS	143	383.2	239.7	1.1	8.31

B – Basal diet; LC – Lucerne; SD-Sawdust; SH-Sunflower hulls; GH-Grass hay; MC-Maize cob; MS-Maize stover

CF – Crude fibre; NDF – Neutral detergent fibre; ADF – Acid detergent fibre; WHC – water holding capacity

Water holding capacity (WHC) was determined by centrifugation (Robertson *et al.*, 1981). With the method, diets with inclusion levels of different fibres were soaked for 48 h in distilled water (1g /500 ml) and samples containing 0.25 - 0.5g dry fibre were then transferred to tarred 25 ml polypropylene centrifuge tubes which contained distilled water (15 ml). After 1 h the contents of the tubes were centrifuged at 6000 g for 15 min, excess water decanted and the tubes inverted and left to drain for 30min. The fresh weight of the fibre was determined before freeze drying to determine the dry weight and hence WHC as g water/g fibre. Three replicates for each sample were analysed.

### **3.2.4 Recording of pig behaviour**

Pigs were allocated at random and maintained on to their trial diets. They were allowed to adapt to the feed offerings and the feeders for seven days, in line with previous studies on dietary fibre (Kyriazakis *et al.*, 1991; Ferguson *et al.*, 1999). After the adaptation phase, behavioural activities were observed on each of the experimental pigs for 16 separate days, involving 8 h experimental protocol per day using overhead video cameras. Video cameras were mounted in such a way that each camera focused on six pigs in separate pens. The use of video cameras was to prevent disturbances from observers to the pigs whilst they are exhibiting their day-to-day normal behavioural activities. In addition, number of visits to the feeder and duration of each visit on the feeder were recorded. Number of visits in the study is defined as movement of pig to the feeder to eat. The behavioural activities recorded were the time spent on eating, drinking, standing/sitting, lying down and other activities (object biting and licking). Description of the behavioural activities is shown in Table 3.4. Videos recorded to monitor behavioural activities of pigs were scored by the same person throughout for the duration of the experiment which lasted for four weeks.



**Table 3. 4: Ethogram of behaviours recorded for pigs in individual pens**

<b>Behaviour</b>	<b>Description of behaviour</b>
Eating	Time spent with the head in the feeder or very close to feeder (includes nosing feeder)
Drinking	Time spent inserting mouth at the nipple drinker
Standing/ sitting	Standing idle with body equally supported by all four legs/ Standing on fore-legs, hind quarter on the floor
Lying down	Lying with shoulder or sternum in contact with floor
Other	When the focal animal is not involved in any of the listed behaviours (non postural and/or feeding behaviours)

Proportion of time spent on different behavioural activities was calculated as time spent on a particular behaviour exhibited by the pig over the total time for all behavioural activities for the whole day. Body weights of the pigs were recorded once a week for four weeks. Feed intake of the different types of fibres was recorded after every week for four weeks. Weights of feed refusals and spillages were subtracted from the total weight of the feed that was put into the feeder to determine feed intake.

Body weight usually affects behavioural activities exhibited by the pig. Time spent on each behavioural activity by a pig was, therefore, expressed as relative time spent to body weight of a pig. The formula used was:

$$\frac{t}{B}$$

Where;  $t$  is time spent on a particular behavioural activity by each pig and  $B$  is the body weight of each pig.

### 3.2.5 Statistical analyses

The PROC CAPABILITY procedure of SAS (2008) was used to check for normality of the data for time spent on each behavioural activity. Thereafter, logarithmic transformation was used to normalise the data for time spent on different behavioural activities since it was not normally distributed. A  $6 \times 6$  factorial analysis was used to test for significance of main effects and interactions on time spent on each behavioural activity using the General Linear Model procedure of SAS software (SAS 2008). The model used was as follows:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha \times \beta)_{ij} + e_{ijk};$$

Where;

$Y_{ijk}$  = Response variable (for time spent on different behavioural activities);

$\mu$  = overall mean response;

$\alpha_i$  = effect of fibre source ( $i$  = maize cobs (MC), grass hay (GH), Lucerne hay (LH), maize stover (MS), sawdust (SD) and sunflower hulls (SH));

$\beta_j$  = effect of inclusion level ( $j$  = 0, 80, 160, 240, 320 and 400g/kg);

$(\alpha \times \beta)_{ij}$  = interaction of effect of fibre source and effect of inclusion level;

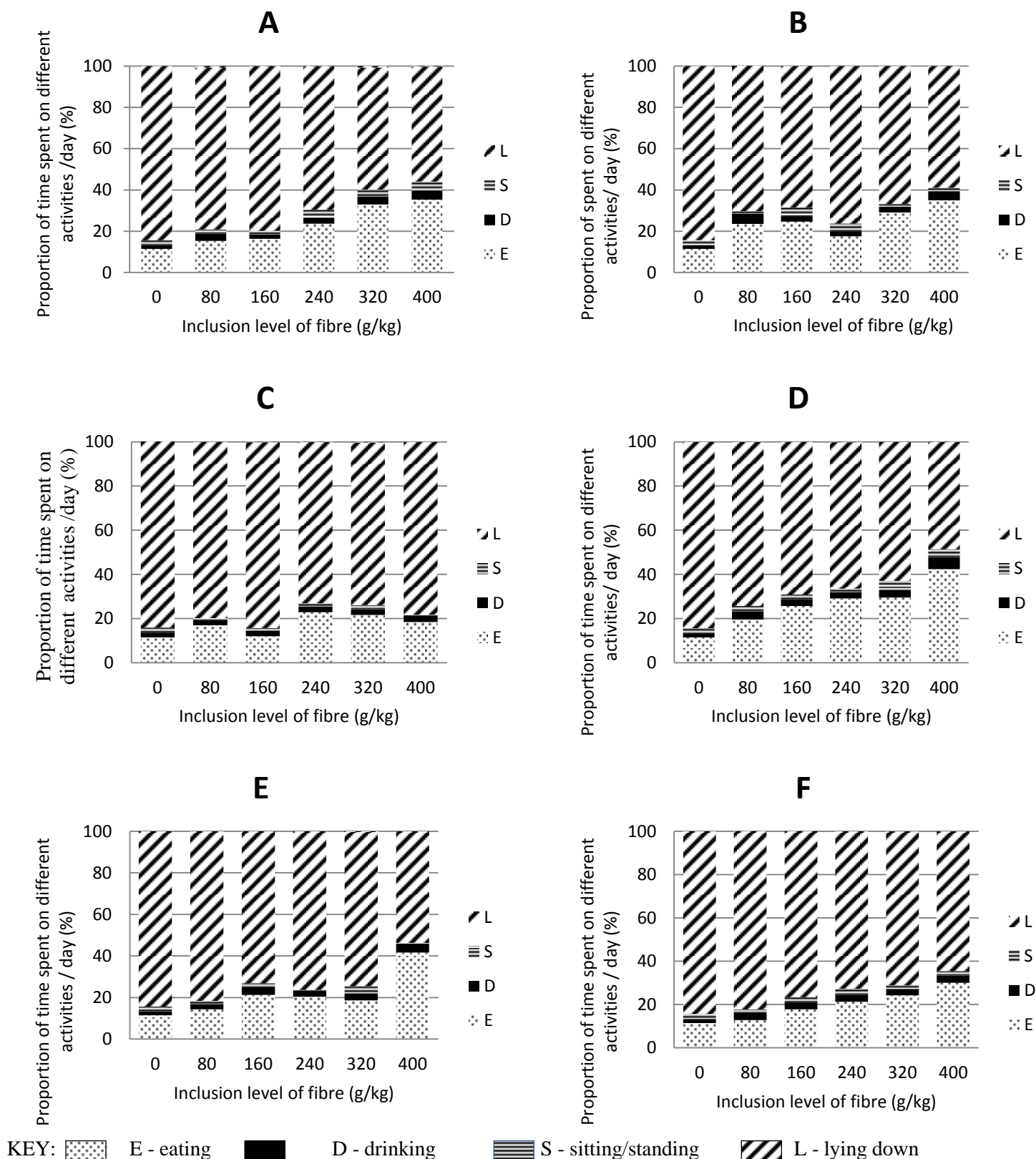
$e_{ijk}$  = residual error.

The STEPWISE procedure of SAS (2008) was used to determine best predictor variables for time spent on different behavioural activities, average daily feed intake (ADFI), average daily gain (ADG), number of visits to the feeder and duration of each visit by to pig on the feeder. Variables incorporated in the model as predictor variables for time spent on different behavioural activities were; DM, DE, CP, EE, ash, CF, NDF, ADF, BD and WHC. Relationships between time spent on different behavioural activities, ADFI, average daily gain, number of visits to the feeder and duration of each visit by to pig on the feeder; and properties of feeds were determined using the quadratic response surface model (SAS 2008).

### **3.3 Results**

#### **3.3.1 Effect of fibre source and inclusion on time spent on different behavioural activities by growing pigs**

Effect of inclusion level and interaction of fibre source and inclusion level was significant ( $P < 0.05$ ). However, effect of fibre source was not significant. Time spent on different behavioural activities varied with inclusion level of fibre per day ( $P < 0.05$ ). Pigs spent most of their time lying down followed by eating, drinking and sitting/standing for all the fibre sources per day (Figure 3.1).



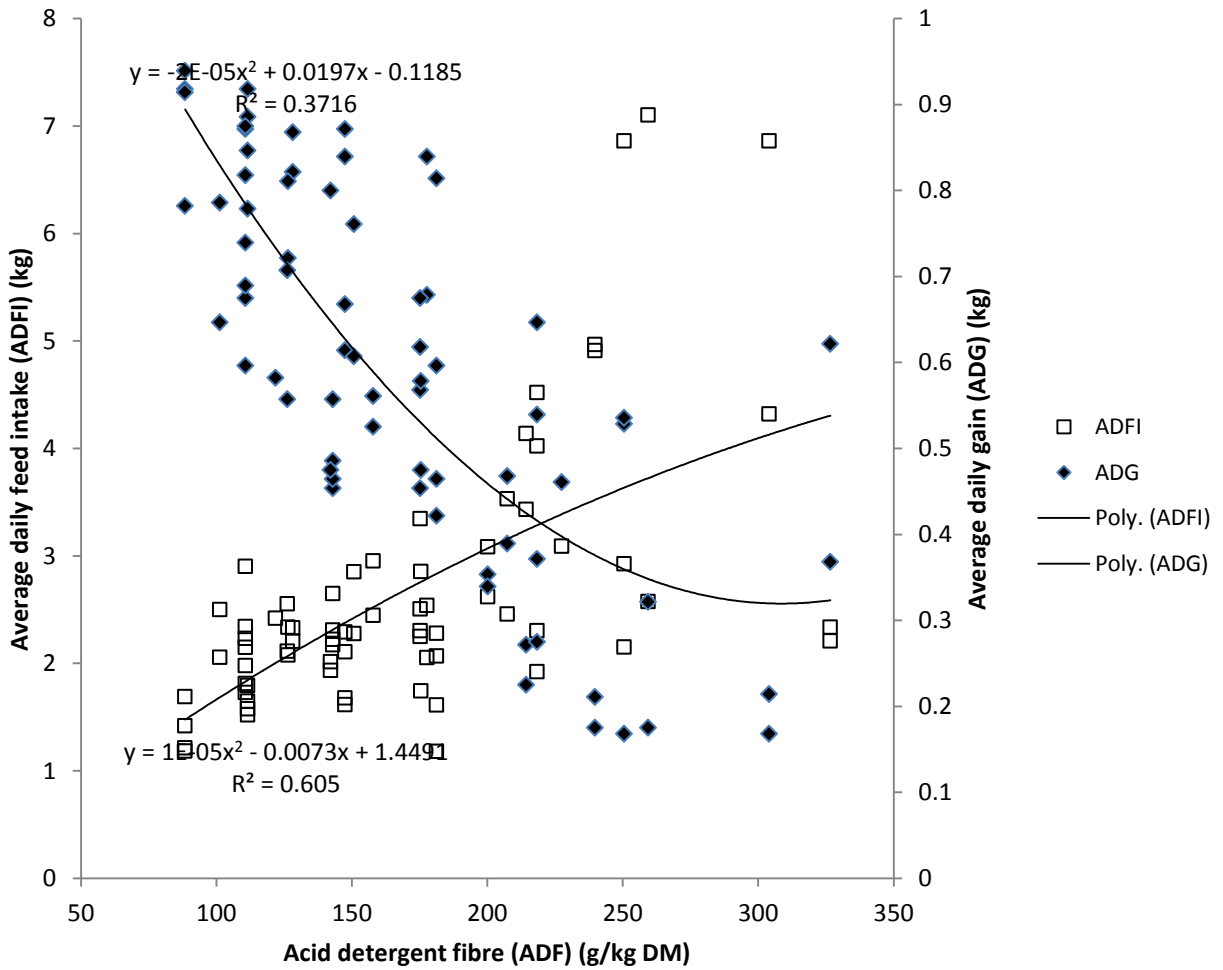
Time spent eating was observed to increase steadily with inclusion level of fibre. Conversely, time spent lying down decreased with inclusion level of fibre. Other behaviours which did not involve feeding activities (eating and drinking) and postural activities (lying down, sitting and standing) were infrequent in pigs subjected to treatment diets.

### **3.3.2 Effects of physicochemical properties of fibrous diets on average daily feed intake and average daily gain of growing pigs**

Physicochemical properties of the feed were used to predict ADFI, ADG and time spent by pigs on different behavioural activities. Acid detergent fibre was the most important predictor variable for determining ADFI and ADG. Figure 3.2 shows the relationships between ADF and ADFI, and ADF and ADG. As ADF content in feed increased, a significant increase ( $P < 0.05$ ) in ADFI was noted. A different trend was, however, noted for ADG. A significant decrease ( $P < 0.05$ ) in ADG was noted as ADF content of diets increased to a level where it becomes constant.

### **3.3.3 Effects of physicochemical properties of fibrous diets on time spent on different behavioural activities by growing pigs**

Table 3.5 shows regression equations predicting time spent on different behavioural activities of pigs from physicochemical properties of diets. Linear responses were noted for time spent eating and drinking from, DE, BD and ADF ( $P < 0.001$ ). A linear response was observed for time sitting/standing from BD as best predictor variable ( $P < 0.05$ ). A quadratic response, on the other hand, was observed for time spent lying down from ADF as best predictor variables ( $P < 0.001$ ).



**Figure 3. 2: Relationship between average daily gain (ADG) and acid detergent fibre (ADF) of feed, and average daily feed intake (ADFI) and Acid detergent fibre (ADF)**

**Table 3. 5: Relationships of best predictor variables (bulk properties) of relative time spent on different behavioural activities to relative body weight**

Behavioural activity	Regression	Equation	R <sup>2</sup>	P-value
E	Linear	$E = -20.54(28.20)BD + 26.71(18.86)$	0.45	<.0001
E	Linear	$E = 0.031(0.015)ADF - 1.33(1.37)$	0.48	<.0001
D	Linear	$D = -1.22(1.43)BD + 1.37(0.958)$	0.41	<.0001
D	Linear	$D = 0.0012(0.000722)ADF - 0.091(0.0662)$	0.49	<.0001
L	Quadratic	$L = -0.00011(0.000047)ADF^2 + 0.055(0.019)ADF + 4.60(1.70)$	0.15	<.0001
S	Linear	$S = -8.98(6.53)BD + 7.104(4.37)$	0.11	0.0010

E - time spent eating; D– time spent drinking; L –time spent lying down; S – time spent sitting and standing

ADF - Acid detergent fibre; BD - bulk density

### **3.3.4 Effects of physicochemical properties of fibrous diets on number of visits to the feeder and duration of each visit on the feeder by growing pigs**

Water holding capacity was the most important variable for predicting the number of visits made by the pig to the feeder per day ( $P < 0.05$ ). Figure 3.3 shows the relationship between mean number of visits to the feeder per day and water holding capacity. A significant decrease ( $P < 0.001$ ) in number of visits to the feeder by a pig per day was observed.

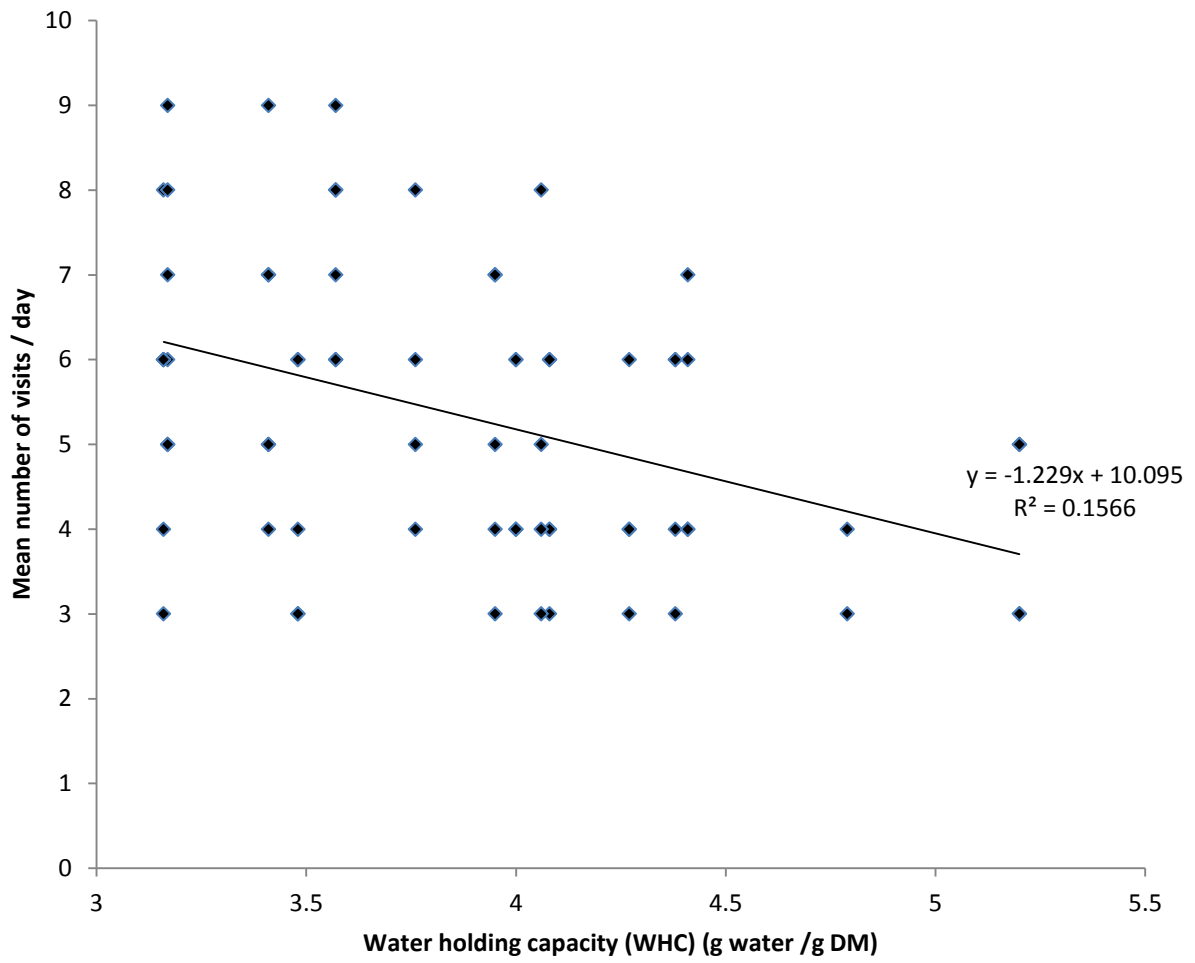
A maximum of nine visits per day were made when water holding capacity was low in a diet. The visits were reduced to minimum of three visits per day at higher water holding capacity. For total time spent on each visit by a pig per day, ADF was the most important predictor variable ( $P < 0.05$ ). Duration of each visit on the feeder per day increased as content in diets increased ( $P < 0.001$ ) (Figure 3.4).

Other physicochemical properties that were not used to predict ADFI, ADG, time spent by the pigs on different behavioural activities, number of visits to the feeder and duration of each visit at the feeder were not statistically significant ( $P > 0.05$ ).

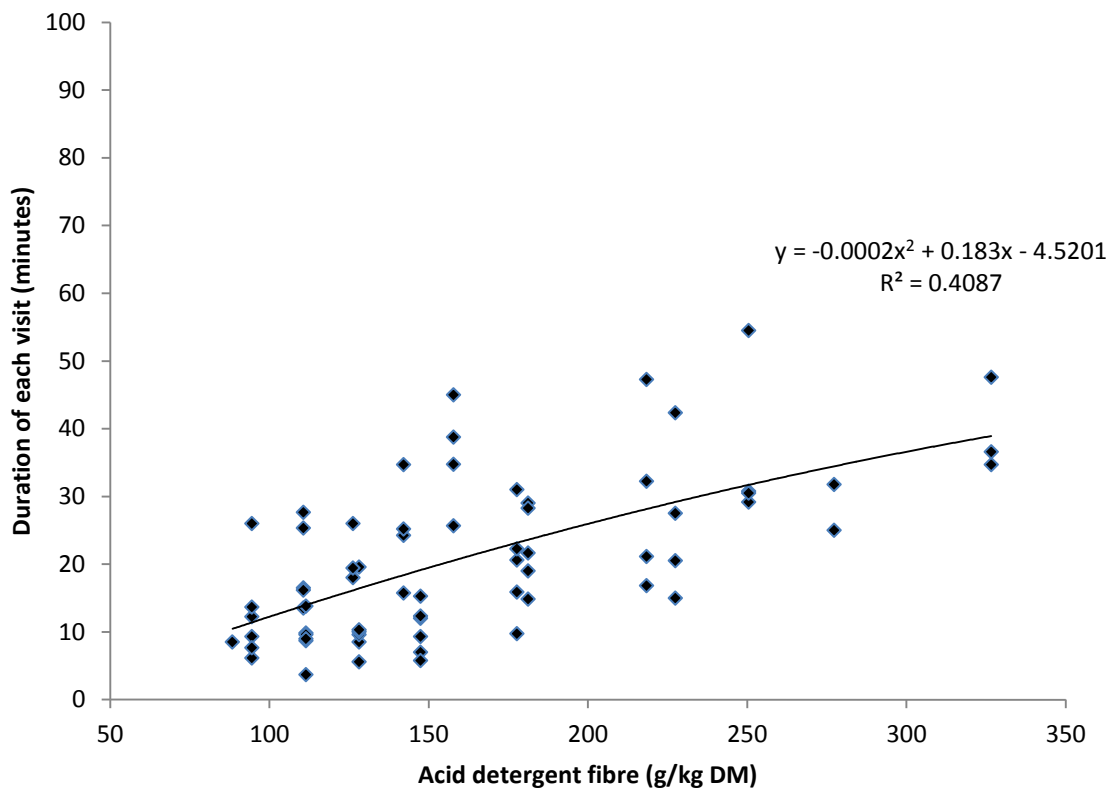
### **3.4 Discussion**

A preliminary study was conducted for the feeding behaviour trial where video cameras were used for 24h. Most of the pigs in the study were lying down after 1600h and activities started to increase after 0800h the next morning when they were offered feed. Observations were, therefore, made between 0800h and 1600h. The observation that pigs are inactive during the night hours is also in agreement with study conducted by Whittemore *et al.* (2002).





**Figure 3. 3: Relationship between mean number of visits to the feeder and water holding capacity (WHC)**



**Figure 3. 4: Relationship between duration of visit on the feeder and acid detergent fibre**

The observation that time spent eating increased with inclusion level of fibre corresponds with previous work in gilts (Rushen *et al.*, 1999), dry sows ( Brouns *et al.*, 1997) and pregnant gilts (McGlone and Fullwood, 2001), which showed that foods high in bulk made the pigs spend more time eating. Fibres are known as diluents in diets of pigs and inclusion of fibres reduces energy density of a diet (Robert *et al.*, 1997; Rijnen *et al.*, 2003), resulting in the pig to compensate for the reduced density at higher inclusion levels by eating more to meet nutrient requirements for growth.

When energy density is low, pigs are expected to eat more. Conversely, at high energy density of feed, the time spent eating is reduced. Gut capacity due to feed bulk was supposed to be revealed from the decrease in time spent eating as fibre level was increased. The observed linear increase in time spent eating might also may be attributed to more time that was channelled to mastication as the diet became coarser with increasing fibre inclusion level (Wenk, 2001). Increased chewing may be necessary to breakdown coarse feed and properly mix feed with saliva to facilitate swallowing. Time spent masticating in the current study was, however, not measured and warrants further investigations.

Acid detergent fibre accounted for the effects on ADFI and ADG. Acid detergent fibre is an indigestible feed component in diets consumed by the pigs. The indigestible component increases transit time of digesta in the gut causing early satiety resulting from gastric signals in response to the elongation of the stomach wall. Due to distension of the gut and indigestible feed material (ADF), the pig will not acquire enough nutrients which meet requirements for growth from eating a diluted feed. This can account for the decrease in average daily feed intake and, hence, low weight gain at higher fibre inclusion levels. The low weight gain at higher inclusion levels could also be attributed to protein in feed that is

made available for use by the pig. Nitrogen not available for use by the pig would be bound by fibre (22 %) resulting in low weight gains at high inclusion level of fibre (Bindelle *et al.*, 2005). Low ADG at higher inclusion level of fibre results in poor health status of the animal due to qualitative feed restriction.

The observed increase in time spent drinking with inclusion level of fibre was expected. There is a positive relationship between feed intake and quantity of water consumed by growing pigs (Jackson, 2007). Ratio of water to feed intake is estimated to be 3:1 (3 litres of water/kg feed). Also, an adaptive mechanism that can be employed by a pig to digest fibrous feed material is to produce prodigious quantities of saliva (Dryden, 2008). Saliva facilitates the breakdown fibres and contains urea, which can be used by the microbes in the lower gut of a pig. A large proportion of saliva constitutes water and, to produce large quantities of saliva, the pig will need to increase water consumption. Saliva production was, however, not measured in the current study.

In the current study involving growing pigs, decrease in time spent lying down resulted from an increase in time spent eating. Lying down might be a sign of satiety in growing pigs receiving low fibrous diets. The pigs must have acquired enough nutrients to satisfy their appetite. As pigs grow, their ability to utilise fibrous feeds is enhanced (Bindelle *et al.*, 2005), making sows to utilize dietary fibre more efficiently than growing pigs. Time spent lying down compared to other activities (eating, drinking and sitting/standing) may be a result of unavailability of space for the pig to manoeuvre freely. In the present study, the design of the individual pens did not permit such free movements. As a result, pigs spent more time lying down compared to other activities.

Time spent sitting/standing as observed in the study is an indication of pigs showing levels of discomfort due to gut fill. This results in pigs to change different positions and was, therefore, trying to cope with stress and to come up with a comfortable position to rest (Pearce and Paterson, 1993). The observation that time spent sitting/standing increased with a decrease in BD of feed might explain levels of alertness to sound and movement pigs' exhibit as they will be anticipating a much more palatable feed. Apart from properties of dietary fibres, pigs also exhibit the activity to reduce the risk of shoulder lesions caused by hard floors in pens (Rolandsdotter *et al.*, 2009).

Growing pigs adhere to a feeding pattern that provides them with sufficient feed which can meet their nutrient requirements for growth (Nielsen *et al.*, 1996). This might be the case in the current study where pigs receiving high fibrous diets had few visits of long duration. The lengths of visits at the feeder are motivated by hunger as the pig will be trying to get nutrients which meet requirement for growth.

Considerable number of visits of short duration in low fibrous diets compared to high fibrous diets, on the other hand, might be attributed to property diets. Low fibrous diets are easily digested and absorbed for use by the pig. This stabilises glucose levels and enhance satiety on the short term. Long durations per visit per day at high ADF content of diets may be due texture of the feed, which results in a pig spending more time masticating. All this creates more time for the ingestion process and less time for other activities.

### **3.5 Conclusions**

Time spent on different behavioural activities of growing pigs was significantly influenced by the inclusion levels of fibre in their diet. Pigs spent more time eating and less time lying

down at higher inclusion level of fibres. Bulk density, DE, ADF and WHC were the most important bulk properties for predicting time spent on different behavioural activities. Inclusion of fibre in diets, therefore, alters behaviour of individually housed growing pigs and could have an impact on their welfare. It is also necessary to determine the relationship between nutritionally-related metabolites and time spent eating. Physicochemical properties of diets and nutritionally-related blood metabolites work in synergy to induce behavioural responses.

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## **Chapter 4: Feeding behaviour and nutritional status of growing pigs fed on diets containing graded levels of maize cobs**

*(Submitted to Asian-Australasian Journal of Animal Sciences)*

### **Abstract**

The objective of the study was to determine the relationships between time spent eating and nutritionally-related blood metabolites in growing pigs fed on fibrous diets. Eighteen growing pigs (initial body weight  $14.2 \pm 1.20$  kg) were used in a five week study period. The pigs were individually penned and subjected to diets containing 0, 80, 160, 240, 320 and 400 g of maize cob/kg DM. Time spent eating and drinking was observed by use of video cameras. Blood was collected at the end of the trial for analysis of glycosylated haemoglobin, total protein (TP), urea, uric acid, albumin and creatinine kinase. Stepwise regression selected glycosylated haemoglobin, albumin, globulin, total protein and uric acid as factors that influenced time spent eating ( $P < 0.05$ ). Albumin, globulin and total protein were the best predictor variables for time spent drinking ( $P < 0.05$ ). Time spent eating increased linearly with an increase in uric acid, total protein, albumin and globulin concentrations and started decreasing at higher fibre concentrations ( $P < 0.05$ ). A negative relationship was observed between time spent eating and glycosylated haemoglobin concentration. Curvilinear responses were noted between time spent drinking and concentration of the nutritional metabolites (albumin, globulin and total protein) ( $P < 0.05$ ). Using broken-stick analyses, to indicate satiety levels from time spent eating and drinking. Uric acid, TP, albumin and globulin as the descriptors of time spent eating, produced threshold values at 14.4 (SEM 2.91) (mg/dl) / kg, 0.55 (SEM 0.042) (g/dl) / kg, 0.28 (SEM 0.024) (g/dl) / kg and 0.28 (SEM 0.125) (g/dl) / kg, respectively. For time spent drinking, threshold values were obtained at 0.23 (SEM 0.012) (g/dl) / kg (albumin), 0.28 (SEM 0.031) (g/dl) / kg (globulin) and 0.40 (SEM 0.021) (g/dl) / kg (TP). In conclusion, the nutritionally-related blood metabolites

provided quadratic relationships with time spent eating and drinking for growing pigs. Threshold values from the relationships can be used to predict satiety levels in pigs subjected to the fibrous diets.

**Key words:** Growing pig, Feeding behaviour, Dietary fibre, Nutritional status

#### **4.1 Introduction**

In chapter 3, physicochemical properties of fibre influenced time spent eating. The physicochemical properties are supposed to cause distension of stomach walls, thus, inducing satiety. Physical and chemostatic mechanisms have been reported to control feed intake in animals (Quiniou *et al.*, 2000; De Leeuw *et al.*, 2004; Hsia and Lu, 2004). Integrating these mechanisms will help in constructing a single model, which can be used by feed compounders to predict intake of feed (Forbes, 2007). It is not clear whether the pattern for the changes in feed intake and time spent eating/drinking are similar. The time spent eating feed is important for producers to predict pig behaviour and aggression of pigs among pen-mates.

Several studies have reported transient decline of glucose, for example, to precede meal initiation (Campfield *et al.*, 1996; De Leeuw *et al.*, 2004). Conversely, high glucose levels induce satiety. The extent of fibre inclusion determines the rate of digestion and absorption of glucose and other nutrients (Wenk, 2001; Metzler and Mosenthin, 2008). Glycosylated haemoglobin is stable indicator of glucose status and can be used for determining time spent eating (Rendell *et al.*, 1985; Alayash *et al.*, 1988; Al-Ali *et al.*, 1990; Shahbazkia and Nazifi, 2008; Shahbazkia *et al.*, 2010). Other metabolites that can be used to determine time spent eating as well as nutritional status of pigs are total protein (TP), albumin, globulin, urea, uric

acid and creatine kinase (CK). The metabolites can also reflect concentrations over the previous weeks and this depends on age, infection to diseases and breed of pig (Atinmo *et al.*, 1976; Chen *et al.*, 1995; Mashatise *et al.* 2005). Maize cob was used in the current study because it is the most abundant, cheap and readily available fibre source in Southern Africa.

The objective of the study was, therefore, to determine whether nutritionally-related blood metabolites from feeding fibrous diets can be used to predict time spent eating and drinking in growing pigs. It was hypothesized that nutritionally-related blood metabolites are potential biomarkers that adequately describe the satiety levels in pigs fed maize cob based diets.

## **4.2 Materials and methods**

### **4.2.1 Pigs and housing**

Eighteen randomly selected clinically healthy castrated male growing pigs were used in the experiment. Initial body weight for the pigs was  $14.3 \pm 1.20$  kg. All pigs were housed in individual pens measuring  $1.5 \times 1$  m and containing a plastic tube feeder (Big Dutchman Lean Machine®) and a low-pressure nipple drinker providing water *ad-libitum*. The space in the pens allowed the pigs to turn around and exercise freely. Ambient temperature and relative humidity were recorded automatically during the trial using HOBO data loggers (Onset Computer Corporation, Pocasset, MA, USA). The average temperature and humidity was  $21.2 \pm 2.74^{\circ}\text{C}$  and  $42.6 \pm 1.56$  %. Care and use of the pigs were in compliant with internationally accepted standards for welfare and ethics in animals and was specifically approved by the University of KwaZulu-Natal Animal Ethics Research Committee (Reference number: 061/12/Animal).

#### **4.2.2 Diets**

A commercial feed (Express weaner, Meadow feeds Ltd, South Africa) with a low level of DF was used as control diet (basal diet). Maize cob was used as a fibre source to dilute the basal diet. Maize cobs used in the experiment were remains left after removal of kernels from the cob. The cobs were ground to pass through a 3 mm screen and thereafter were included at different inclusion levels of 80, 160, 240, 320 and 400 g/kg in diets of pigs. It was assumed that the pigs would increase their feed intake proportionately to the degree of dilution of the basal feed with the fibre source. Hence, all pigs would get nutrients that meet requirements for growth.

#### **4.2.3 Analyses of physicochemical properties of diets**

Methods used for analyses of the diets used in the trial have been described in chapter 3. Table 4.1 shows chemical and physicochemical properties of the different diets.

#### **4.2.4 Pig behaviour**

Pigs were allowed to adapt to their new environment, the feed offered and to using the feeders for a period of one week. Thereafter, behavioural activities were observed for 8 h a day on four days of the week using overhead video cameras. Video cameras were mounted from the ceiling in such a way that each camera focused on six pigs. Disturbances to the pigs were prevented to allow pigs to exhibit their day-to-day normal behavioural activities. The behavioural activity recorded was time spent on eating a particular fibrous diet and drinking. Time spent eating was recorded when the pig's head was within the feeder and the head moving as if its chewing whilst time spent drinking was when the pig's snout was in contact with the nipple drinker and neck moving in a gulping movement.

**Table 4. 1: Chemical and physicochemical properties of diets**

Inclusion level (g/kg)	DM (g/kg)	calc DE (MJ/kg) <sup>1</sup>	CP (g/kg DM)	EE (g/kg DM)	Ash (g/kg DM)	CF(g/kg DM)	NDF(g/kg DM)	ADF(g/kg DM)	BD (g DM/ml)	WHC (g water/g DM)
0	989.3	13.9	195.7	52.9	61.1	26.1	192.3	88.4	1.45	3.76
80	989.3	13.2	185.7	51.2	59	36.4	230.6	111.5	1.52	3.17
160	990.4	12.1	168.1	45.9	54.9	47.9	294.4	147.4	1.47	3.57
240	990.5	11	152.8	45.2	53.2	59.3	355.3	181.2	1.42	4.08
320	990.2	10.3	139.4	41.3	52.1	70.8	401.3	218.4	1.25	4.41
400	991.1	9.3	116.4	39.9	46.7	82.2	457.4	250.5	1.22	4.75

DM –Dry matter; CP – Crude protein; EE – Ether extract; calc DE –calculated digestible energy; CF – crude fibre; NDF – Neutral detergent fibre; ADF – Acid detergent fibre; BD- bulk density; WHC – water holding capacity

<sup>1</sup>calc DE = 949 + (0.789 x GE) - (43 x %Ash) - (41 x %NDF) (Noblet and Perez, 1993)

#### 4.2.5 Blood collection and analyses

After 5 weeks of feeding on the diets, blood samples were taken from jugular vein using vacutainer blood collection tubes. It was assumed that concentration of nutritionally-related blood metabolites collected at the end of the experiment resembled concentration from previous days. One set of tubes used for collecting blood had ethylene-diamine-tetraacetic acid (EDTA) for TP, albumin, globulin, urea, uric acid levels, CK examination. The other set of tubes had no EDTA for HbG examination. Blood samples collected in tubes with no EDTA were centrifuged at 3 000 rpm for 5 min, and serum separated was stored at  $-20^{\circ}\text{C}$  prior to analysis of TP, albumin, globulin, urea, uric acid levels and CK. Blood samples for examination of HbG were stored in a temperature range of between  $2 - 8^{\circ}\text{C}$ . Total protein, albumin, globulin, urea, uric acid, CK and HbG levels were measured using an automated chemistry analyzer with reagents kits from Labtest Diagnostics (Labmax Plenno, Labtest, Lagoa-Santa, Brazil).

#### 4.2.6 Calculations and statistical analyses

The data for each pig on time spent eating and drinking, and concentration of nutritionally related blood metabolites were adjusted by the metabolic body weight (MBW) to give relative measurements. Metabolic body weight of each animal was calculated using the formula:

$$\text{Metabolic body weight} = \mathbf{BW}^{0.75}$$

Where; **BW** - body weight of a pig



Following the Proc capability procedure of SAS (2008) to check for normality of the data, data for relative time spent on eating to MBW of pig (RTSE) and relative time spent drinking to MBW of a pig (RTSD) was not normally distributed. It was then log transformed to normalize it. Data was then back-transformed when reporting. A one-way factorial analysis with a common control treatment was used to test for significance of inclusion level on concentration of nutritionally-related metabolites using the General Linear Model procedure of SAS (SAS 2008). The quadratic response surface model (SAS 2008) was used to determine the relationship between inclusion level and average daily feed intake (ADFI), relationship between inclusion level and average daily gain (ADG), relationship between inclusion level and behavioural activities, relationship between feed conversion ratio (FCR) and inclusion level of fibre, and relationship between inclusion level and concentration of nutritionally related metabolites. Stepwise regression in SAS (2008) was used to identify the best nutritionally-related blood metabolites which influence RTSE and RTSD. The broken-stick model as described by Robbins *et al.* (2006) was then used to determine the thresholds values indicating satiety levels from relationship between RTSE and RTSD, and nutritionally-related blood metabolites using NLIN procedure (SAS, 2008).

## **4.3 Results**

### **4.3.1 Effect of feeding graded levels of maize cob on average daily feed intake, average daily gain, and feed conversion ratio and, time spent eating and drinking by growing pigs**

Average daily feed intake and ADG decreased linearly with inclusion of maize cob ( $P < 0.05$ ; Table 4.2). There were no differences for FCR of pigs as inclusion level of maize cob was increased. Both RTSE and RTSD increased linearly with inclusion level of maize cob ( $P < 0.05$ ; Table 4.3). More time was spent eating at higher inclusion levels than in diets with low fibre inclusion levels. The same trend was also observed for time spent drinking by the pigs.

**Table 4. 2: Effect of maize cob inclusion level in diets on average daily feed intake (ADFI), average daily gain (ADG) and feed conversion ratio (FCR) in growing pigs**

Performance parameters	Inclusion level (g / kg)							<i>P-value</i>	
	0	80	160	240	320	400	SE	Linear	Quadratic
ADFI (kg/day)	1.31	1.39	1.11	1.31	1.01	0.83	0.10	*	NS
ADG (g/day)	0.324	0.279	0.263	0.289	0.224	0.152	0.061	*	NS
FCR	4.04	4.98	4.22	4.53	4.51	5.46	0.520	NS	NS

\*P < 0.05; NS - not significant (P > 0.05); SE – pooled standard error

**Table 4. 3: Effect of maize cob inclusion level in diets on time spent eating and drinking in growing pigs**

Activity	Inclusion level (g/kg)						SE	P- value	
	0	80	160	240	320	400		Linear	Quadratic
RTSE (min/MBW/day)	3.46	4.29	4.91	6.12	12.34	12.89	1.10	**	*
RTSD (min/MBW/day)	0.69	0.77	0.93	1.01	1.24	1.09	0.15	*	NS

\*P < 0.05; \*\*P < 0.01; NS - not significant (P > 0.05); SE- pooled standard error

RTSE - relative time spent on eating to MBW of pig; RTSD - relative time spent on eating to MBW of pig

### **4.3.2 Effect of feeding graded levels of maize cob on concentration of nutritionally-related blood metabolites**

Table 4.4 shows relationships between concentration of nutritionally-related metabolites and fibre inclusion levels in diets. There were positive linear responses found between the blood metabolites (TP, albumin, globulin, urea and uric acid) and fibre inclusion level in diets of pigs ( $P < 0.05$ ). A quadratic response was found between CK and inclusion level of fibre in diets of pigs ( $P < 0.05$ ). Glycosylated haemoglobin levels, on the other hand, decreased with inclusion levels of fibre in diets of the pigs ( $P < 0.05$ ).

### **4.3.3 Predicting time spent eating and drinking from nutritionally-related blood metabolites**

Using stepwise regression HbG, albumin, globulin, TP and uric acid were the blood metabolites selected to influence RTSE. Table 4.5 shows the relationships between the metabolites and RTSE. Relative time spent eating to MBW of pigs showed curvilinear responses with an increase in concentration of the blood metabolites ( $P < 0.05$ ). Relative time spent eating to MBW of pigs increased linearly with an increase in uric acid, TP, albumin and globulin concentrations up to a level where it starts decreasing at higher concentrations. A negative relationship was noted between RTSE and HbG concentration in blood of pigs fed graded levels of fibre ( $P < 0.05$ ). Blood metabolites selected as best predictor variables for RTSD were albumin, globulin and TP. Curvilinear responses were produced between RTSD and concentration of the nutritional metabolites (albumin, globulin and TP) ( $P < 0.05$ ; Table 4.6).

**Table 4. 4: Effect of maize cob inclusion level in diets on blood metabolites in growing pigs**

Blood metabolites	Inclusion level of maize cob (g/kg)							Regression	
	0	80	160	240	320	400	SE	Linear	Quadratic
TP (g/dl) / kg MBW	0.40	0.41	0.47	0.43	0.50	0.58	0.04	*	NS
Albumin (g/dl) /kg MBW	0.20	0.21	0.21	0.22	0.29	0.26	0.01	*	NS
Globulin (g/dl) / kg MBW	0.20	0.20	0.25	0.20	0.25	0.32	0.03	*	NS
CK (U/L) / kg MBW	64.3	81.1	73.9	83.6	85.1	176.3	11.2	**	*
Urea (mg/dl) / kg MBW	0.50	0.74	0.88	0.72	1.28	1.35	0.22	*	NS
Uric acid (mg/dl) / kg MBW	5.98	8.90	11.62	9.41	10.13	16.20	1.29	*	NS
HbG (%) / kg MBW	0.067	0.065	0.061	0.067	0.055	0.041	0.007	*	NS

\*P < 0.05; \*\* P < 0.01; NS - not significant (P > 0.05); SE- pooled standard error

TP - total protein; CK - creatine kinase; HbG - glycosylated haemoglobin; MBW – metabolic body weight;

**Table 4. 5: Relationships between time spent eating and blood metabolites in growing pigs (estimate  $\pm$  pooled standard error)**

Blood metabolites	Components of a regression			Relationship		
	$ax^2$	$bx$	$c$	Linear	Quadratic	$X_c$
Uric acid (mg/dl) / kg MBW	$-0.11 \times 10^{-2} \pm 0.06 \times 10^{-2}$	$0.14 \pm 0.02$	$0.21 \pm 0.09$	**	*	$14.41 \pm 2.91$
TP (g/dl) / kg MBW	$-10.52 \pm 2.93$	$12.18 \pm 3.11$	$-2.18 \pm 0.80$	**	**	$0.60 \pm 0.04$
Globulin (g/dl) / kg MBW	$-13.13 \pm 3.28$	$9.14 \pm 2.19$	$-0.90 \pm 0.33$	NS	**	$0.32 \pm 0.13$
Albumin (g/dl) / kg MBW	$-24.78 \pm 4.13$	$18.17 \pm 2.94$	$-2.31 \pm 0.45$	NS	**	$0.29 \pm 0.02$
HbG (%) / kg MBW	$17.71 \pm 4.46$	$-10.88 \pm 2.61$	$1.13 \pm 0.14$	NS	**	-

\* $P < 0.05$ ; \*\*  $P < 0.01$ ; NS - not significant ( $P > 0.05$ );  $X_c$  – threshold values

TP - total protein; HbG - glycosylated haemoglobin; MBW – metabolic body weight; **a** - regression coefficient of  $x^2$ ; **b** - regression coefficient of  $x$ ; **c** - intercept

**Table 4. 6: Relationships between time spent drinking and blood metabolites in growing pigs (estimate ± pooled standard error)**

<b>Blood metabolites</b>	<b>Components of a regression</b>			<b>Relationship</b>	
	<b>ax<sup>2</sup></b>	<b>bx</b>	<b>c</b>	<b>Quadratic</b>	<b>X<sub>c</sub></b>
TP (g/dl) / kg MBW	-2.01 ± 0.65	2.19 ± 0.70	-0.53 ± 0.18	**	0.41 ± 0.02
Albumin (g/dl) / kg MBW	-3.12 ± 0.95	2.23 ± 0.67	-0.24 ± 0.10	**	0.19 ± 0.01
Globulin (g/dl) / kg MBW	-2.29 ± 0.73	1.50 ± 0.49	-0.11 ± 0.07	**	0.26 ± 0.03

\*\* P < 0.01; NS - not significant (P > 0.05); X<sub>c</sub> – threshold values

TP - total protein; MBW – metabolic body weight; **a** - regression coefficient of x<sup>2</sup>; **b** - regression coefficient of x; **c** - intercept

The broken-stick model was used to determine the thresholds values indicating satiety levels from relationship between time spent eating and drinking, and nutritionally-related blood metabolites. As shown in Table 4.5, using uric acid, TP, albumin and globulin as the descriptors of RTSE, threshold values were reached. For RTSD, threshold values were also obtained, as shown in Table 4.6.

#### **4.4 Discussion**

Growing phase in pig production is important because it is the phase where nutritional requirements of feed are most needed for growth. Hence, feed compounders are expected to gain an insight on how the growing pigs are able to utilise nutrients from the formulated fibrous diets to prevent retarded growth. The observation that time spent eating increased with inclusion level of maize cobs was expected. Inclusion of fibres in diets has been reported to reduce energy density of feed (Noblet and van Milgen, 2004). This results in pigs spending more time eating diets higher in fibre to compensate for the reduced energy density in order to meet requirements for growth (Robert *et al.*, 1993; Rijnen *et al.*, 2003). Several studies have also reported time spent eating to be strongly correlated to time spent drinking (Dybkjær *et al.*, 2006; Shaw *et al.*, 2006). In the current study, the time spent drinking increased in the same way as time spent eating. Perhaps, drinking water was a way of stimulating appetite as the pig would be trying to consume feed which will be changing in palatability with inclusion of fibre in a diet.

Indigestible components in the diet (NDF and ADF) increase with inclusion level of fibre (Ndou *et al.*, 2013). These indigestible components have a masking effect that hinders exposure of other nutrients to enzymatic digestion. For example, Bindelle *et al.* (2005) reported that about 220 g/kg



of nitrogen is bound to NDF in grasses. Unavailability of nitrogen due to binding by the fibres might have resulted in low weight gain at high fibre inclusion levels in the study (Glisto *et al.*, 1998; Mikkelsen *et al.*, 2004). Nitrogen is essential for synthesizing amino acids for tissue building. Decrease in weight in pigs fed on graded levels of fibre has also been reported in earlier studies (Ndindana *et al.*, 2002).

Increase in plasma total protein and albumin with inclusion level of fibre was not expected. Concentrations of these plasma proteins are expected to be related to the dietary protein (Bergen and Potter, 1975). The elevated urea and plasma protein levels might be a consequence of reduced urinary excretion and/or protein catabolism caused by stress in pigs subjected to high fibrous diets (Zervas and Zijlstra, 2002).

The decrease in glucose concentrations with inclusion level of fibre reflect the negative energy balance the pigs were in. Inclusion of fibres in a diet might have resulted in some nutrients not to be digested and absorbed, mainly due to accelerated transit time of feed along the gut (Wenk, 2001; Metzler and Mosenthin, 2008). Glycosylated haemoglobin was used because it is stable and is not affected by stress and consumption of meals. The observation that CK levels increase with fibre inclusion level was expected. Creatine kinase is an indicator of stress in pigs (Kim *et al.*, 2004). As more fibre was included in the diet, the pigs became more stressed.

One of the adaptive mechanisms pigs employ to digest fibrous feeds is to recycle urea (Mosenthin *et al.*, 1992). Urea increased in the blood of pigs as fibre levels in a diet increased. The observed higher plasma urea levels in pigs fed diets high in fibre could be attributed to

imbalances in dietary amino acid content which led to increased amino acid catabolism (wasting of tissue). Also, fibre is known to influence endogenous amino acid losses. This, therefore, impacts the appearance of amino acids (or urea) in the portal blood and subsequently at the site of protein synthesis.

The observation that time spent eating produced quadratic relationships with HbG, albumin, globulin, TP and uric acid concentrations in blood was expected. The behavioural responses are a result of negative energy and protein balance in the body of pigs (Le Bellego *et al.*, 2001). Glucose, for example, decreased with inclusion level of fibre in the diet. The decrease in glucose concentration might be an indication that the pigs were unable to extract nutrients from high fibrous diets. If glucose falls below the normal levels in the circulatory system, pigs respond by spending more time eating with inclusion level of fibre to restore normal blood glucose levels. Uric acid concentration is an indication of protein catabolism in the body of pigs. Metabolic status of the pigs could have forced the pigs to increase time spent eating in order for them to get enough protein which meet requirements for growth. Although the pigs increased time spent eating, they were still not getting required nutrients from the feed resulting in the body to waste tissue to increase protein and uric acid concentrations in blood. The broken-stick model indicated break points for inducing satiety from the metabolites to fall at 14.4 (mg/dl) / kg (uric acid), 0.55 (g/dl) / kg (total protein), 0.28 (g/dl) / kg (albumin) and 0.28 (g/dl) / kg (globulin). The break points fall within a range of 320 to 400g/kg inclusion of maize cob. This corresponds to study conducted by Ndou *et al.* (2013), where break point for inclusion level of sunflower hulls indicating satiety due to gut capacity was at 320 g/kg in diet of pigs.

Total plasma protein, globulin and albumin concentrations are often used to assess the hydration status of animals with high levels indicating dehydration due to increased plasma osmolality (Deaux *et al.*, 1970) as more water in the circulatory system will be used to produce saliva to be used during mastication process (Jaber *et al.*, 2004; Averos *et al.*, 2007). The pigs might have drunk insufficient water in relation to the feed consumed as incremental levels of fibre were included in the diets. The observation that time spent drinking increased at a decreasing rate could be attributed to body responding to the increased plasma osmolality by triggering anti-diuretic hormone (ADH) secretion, thus, allowing the kidneys not to excrete water as waste through urine. Water intake and the concentration of anti-diuretic hormone were, however, not measured. Break points obtained particularly for albumin (0.23 (g/dl) / kg) and globulin (0.28 (g/dl) / kg) shows that there is a positive correlation between time spent eating and time spent drinking.

#### **4.5 Conclusions**

Glycosylated haemoglobin, TP, globulin, albumin and uric acid were the best predictors for relative time spent on eating to MBW of growing pigs. Total protein, albumin and globulin concentrations, on the other hand, were the best predictors for relative time spent drinking to MBW of a pig. The nutritionally-related blood metabolites were correlated with time spent eating and drinking. The nutritionally related blood metabolites provided threshold values with relative time spent on eating to MBW of pig and relative time spent drinking to MBW of a pig. They are potential biomarkers that modulate neuronal pathways that reduce time spent eating and drinking. To better understand how the nutrients are absorbed, there is a need to understand the

mucosal architecture of the intestines (villi height and crypt depth). Estimating villi height and crypt depth is important to understand adaptation of pigs fed on fibrous diets.

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## **Chapter 5: Predicting histomorphometry of small intestines of growing pigs fed maize cob diet**

*(Submitted to Journal of Animal Science)*

### **Abstract**

The objective of the study was to predict villi height and apparent villi surface area from physicochemical measurements of maize cob based diets. Eighteen growing male pigs (initial bodyweight  $14.3 \pm 1.20$  kg) were used in the experiment. Pigs were individually penned and subjected to diets containing 0, 80, 160, 240, 320, 400 g/kg maize cob meal. Feed and water were provided ad-libitum. Using stepwise regression, bulk density (BD) and neutral detergent fibre (NDF) were the best predictor variables influencing villi height (VH) and apparent villi surface area (AVSA) ( $P < 0.05$ ). Villi height produced quadratic and linear responses with BD and NDF, respectively ( $P < 0.05$ ). The equations are:  $VH = 211.3(BD) - 591.0(BD) + 442.4$  and  $VH = 0.03(NDF) + 22.8$ . Conversely, AVSA produced quadratic and linear responses with NDF and BD, respectively ( $P < 0.05$ ). The equations are:  $AVSA = 0.00036(NDF) - 0.012(NDF) + 7.25$  and  $AVSA = - 47.12(BD) + 45.03$ . In conclusion, BD and NDF of a feed can be used to predict VH and AVSA of growing pigs fed maize cobs.

**Key words:** Feed bulk; Histomorphometric; Maize cob meal; Mucosa; Physicochemical properties.

## 5.1 Introduction

Concentration of nutritionally-related blood metabolites varied with fibre source and inclusion level (Chapter 4). It is possible that such variations are owed to by mucosal changes in the gastro-intestinal tract (GIT) of the pigs. The fibrous diets have been reported to interact with both the micro-flora and the mucosa of all sites of the small intestines, which are important in the assimilation of nutrients and a major component for optimal gut health (Montagne *et al.*, 2003). The gastro-intestinal tract (GIT), which is the first organ system directly affected by the fibrous diets displays both acute and long-term adaptations (Ferraris and Carey, 2000). Such changes occur to the visceral organs and mucosal architecture of the intestines.

Feeding pigs on diets that increase visceral organ mass results in an increase in energy expenditure, which is concomitant with greater rates of cellular turnover (McBride and Kelly, 1990). Cellular turnover could be assessed by degree of variations in the intestinal morphology of the pigs subjected to fibrous diets. When formulating pig diets rich in dietary fibre, feed compounders should not compromise on the way nutrients are absorbed and converted to protein in pigs.

The ability of pigs to absorb nutrients lies in mucosal structure of the small intestines (Hedemann *et al.*, 2006; Woyengo *et al.*, 2011). Mucosal structure relates to villi height (VH) and crypt depth (CD) and is influenced by feeding level and composition of diets. There are conflicting reports on the influence of dietary fibre on the mucosa. High fibre inclusion levels were reported to increase VH and CD (Ngoc *et al.*, 2012) and to also decrease VH (Agyekum *et al.*, 2012). Some reports have argued that fibre does not influence mucosal structure (Jin, 1992; Jin *et al.*, 1994). Different

fibre sources elicit varied impacts on the mucosa. Maize cobs have a relatively low water holding capacity and does not depress feed intake greatly even at inclusion levels of up to 30 % (Ndindana *et al.*, 2002; Ndou *et al.*, 2013). Although inclusion of maize cobs in diets of pigs alters characteristics of the diets, their effects on VH and AVSA are not yet clear. The relationship between diet characteristics and, VH and AVSA provides a theory basis in formulation of diets for the weaner pigs. Villi height and AVSA determine the absorptive capacity of nutrients for use by pigs, which determine their overall performance (Nabuurr *et al.*; 1993; Vente Spreeuwenberg and Beynen, 2003). The objective of the study was to determine the feed characteristics that best describe relationship with the histomorphometry of small intestines (VH and AVSA) of growing pigs. It was hypothesized that VH and AVSA pigs will increase as levels of maize cobs in diets of pigs is increased.

## **5.2 Materials and methods**

### **5.2.1 Pigs and housing**

A total of 18 clinically healthy castrated male growing pigs, weighing  $14.3 \pm 1.20$  kg were used in the experiment. The pigs were treated in accordance with the University of KwaZulu-Natal guidelines for the care and use of laboratory animals (Reference number: 061/12/Animal). All pigs were penned individually in metabolism crates measuring  $1.5 \times 1$  m and containing a plastic tube feeder (Big Dutchman Lean Machine®) and a low-pressure nipple drinker. The diets and drinking water were provided ad-libitum throughout the whole experimental period. HOBO data loggers (Onset Computer Corporation, Pocasset, MA, USA) were used to record automatically ambient temperatures and relative humidity during the trial. The average temperature and humidity was 21 °C and 42.6 %, respectively.

### **5.2.2 Diets and feeding management**

The diets and feeding management have been described in Chapter 4.

### **5.2.3 Analyses of physicochemical properties of diets**

Methods used for analyses of the diets in the trial have been described in Chapter 3. Table 4.1 shows chemical and physicochemical properties of diets.

### **5.2.4 Visceral organ measurements**

After four weeks on the experimental diet, pigs were euthanised with intravenous injection using sodium thiopental. The pigs were slit opened on the abdomen and entire intestines and organs (liver and pancreas) in the abdominal cavity were removed. The intestines were untwined and length of the small and large intestines was recorded. Weights of the liver and pancreas, empty weights of both small and large intestines, and stomach of the pigs were also recorded.

### **5.2.5 Mucosal structural measurements**

The small intestines of each pig were divided into duodenum, jejunum and ileum segments. From each intestinal segment 10 cm pieces were cut and immediately fixed in 10 % neutral formalin prior to preparation and analysis. The intestinal samples were sectioned and stained with haematoxylin-eosin stain. The sections were observed at 100 × magnification using a light microscope equipped with an ocular micrometer. The VH, CD, villus to crypt ratio, villus apical width and AVSA were recorded. Between eight and 11 randomly chosen and well-oriented villi were selected for the measurements. Villi height was measured from the tip to the base of the villus, whilst CD was measured from the tip of the crypt to the point where it meets the

muscularis mucosa (Velayudhan *et al.*, 2008).

Apparent villus surface area was estimated using the formula (Iji *et al.*, 2001) (Appendix 3):

$$AVSA = \frac{(a+b)}{2} \times c$$

Where;

*a* is villus apical width; *b* is villus basal width; and *c* is villus height.

### 5.2.6 Calculations and statistical analyses

The data collected for visceral organ and mucosal architecture measurements were divided by the metabolic body weight to give relative measurements. This would take account of any differences in mean weight between treatment diets. Metabolic body weight of each animal was calculated using the formula:

$$\text{Metabolic body weight (MBW)} = \text{BW}^{0.75},$$

Where; BW - body weight of the pig

Effects of feeding fibrous diets on visceral organs size and length, and mucosal architecture measurements were analysed using the general linear models procedure of SAS (2008). The following models were used:

For visceral organ measurements:

$$Y_{ij} = \mu + \text{TRT}_i + e_{ij}, \text{ where;}$$

$Y_{ij}$  = response variable (visceral organ measurements);

$\mu$  = the overall mean response common to all observations;

$TRT_i$  = the effect of the fibre inclusion level;

$e_{ij}$  = the residual error.

For mucosal architecture measurements:

$Y_{ij} = \mu + TRT_i + Seg (TRT)_{ij} + e_{ij}$ , where;

$Y_{ij}$  = response variable (AVSA, CD, VH and VH : CD ratio);

$\mu$  = is the overall mean response common to all observations;

$TRT_i$  = the effect of the fibre inclusion level;

$Seg (TRT)_{ij}$  = intestinal segment nested in treatment;

$e_{ij}$  = the residual error.

Mean separation was done using the PDIFF procedure of SAS (2008) for factors that were significant ( $P < 0.05$ ). Stepwise regression in SAS (2008) was used to identify feed characteristic (DM, GE, CP, EE, NDF, ADF and WHC) which best influence intestinal VH and AVSA. Thereafter, quadratic response surface model (SAS, 2008) was used to determine the relationships between feed characteristics selected from stepwise regression and, VH and AVSA of the small intestines. Pearson's correlation coefficients between VH and AVSA were also determined (SAS, 2008).

## **5.3 Results**

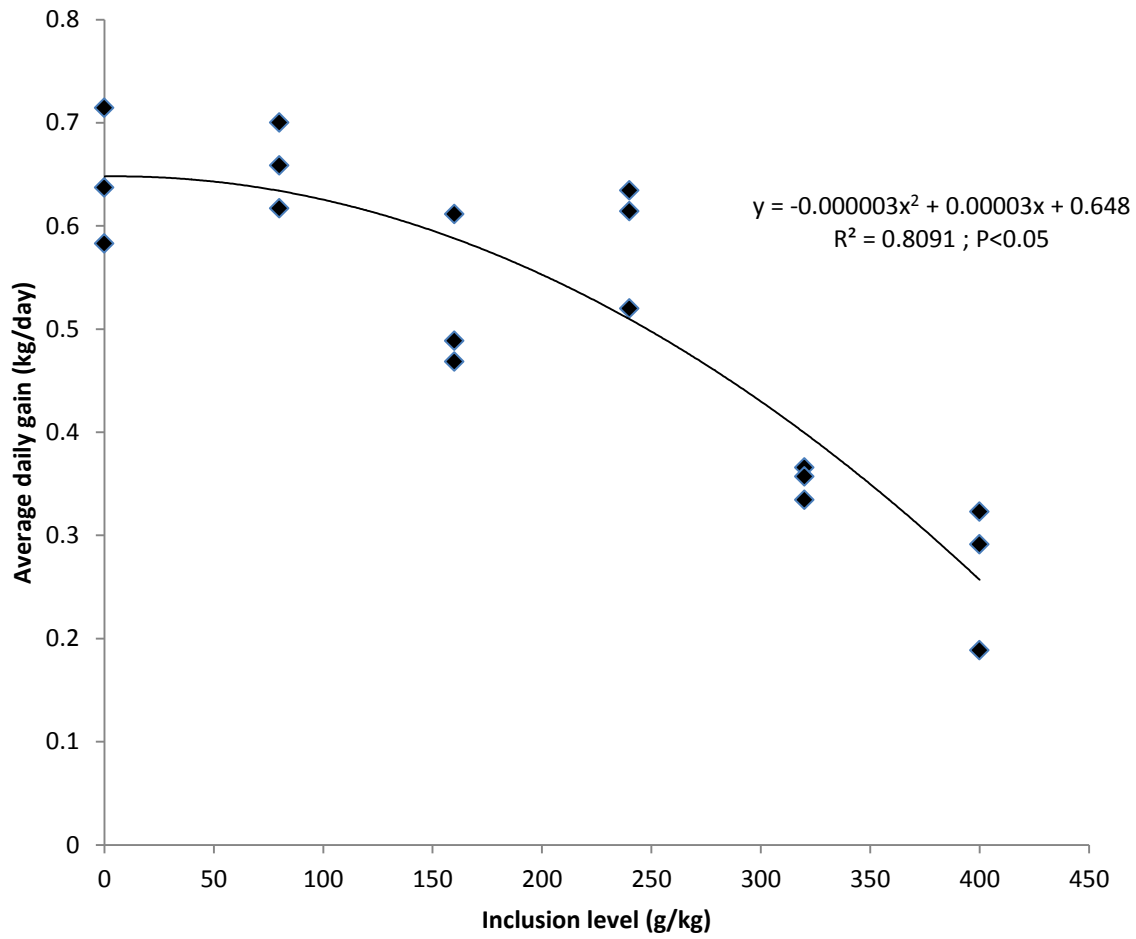
### **5.3.1 Effects of feeding graded levels of maize cob on average daily gain**

All pigs were clinically healthy and had almost similar weights at the beginning of the experiment. Pigs that received diets high in fibre content had low average daily weight gains (ADG) compared to those receiving low fibre diets ( $P < 0.05$ ) (Figure 5.1).

### **5.3.2 Effects of feeding graded levels of maize cob on organ weights and lengths of intestines**

Table 5.1 shows the means and relationships of relative organ weights and lengths of intestines of pigs fed diets with varying inclusion levels of fibre, respectively. Relative weights of the livers of pig decreased linearly with inclusion level of fibre ( $P < 0.05$ ). Conversely, relative weights of the pancreas and the stomachs increased linearly with inclusion level of fibre. There were no significant differences for relative weights of the small and large intestines of pigs fed diets with incremental levels of fibre. Relative lengths to metabolic body weight of pigs for the small intestines and large intestines increased ( $P < 0.05$ ) with inclusion level of fibre, respectively.





**Figure 5. 1: Relationship between average daily gain and inclusion level of fibre**

### **5.3.3 Effects of feeding graded levels of maize cob on histological measurements of the mucosa of the intestines**

Table 5.2 shows means and relationships of the histological measurements of the mucosa of the intestines. Villi height increased linearly with inclusion level of fibre in the duodenum, jejunum and ileum segments of the small intestines ( $P < 0.05$ ). The same trend was also observed for AVSA, which increased with inclusion level of fibre ( $P < 0.05$ ). Crypt depth was observed to increase with inclusion level of fibre in all the segments of the small intestines ( $P < 0.05$ ). There were no differences in villi height to crypt depth ratio of pigs fed incremental levels of fibre in the jejunum and ileum ( $P > 0.05$ ). Villi height to crypt depth ratio increased linearly with inclusion level of fibre in the duodenum segment of the small intestine ( $P < 0.05$ ).

**Table 5. 1: Visceral organ weights and lengths of growing pigs fed diets with incremental levels of fibre**

Organs	Inclusion level (g/kg)							Linear		Quadratic	
	0	80	160	240	320	400	SE	Regression coefficient	SE	Regression coefficient	SE
<b>Weights (g / MBWkg)</b>											
Liver	96.05	94.76	84.53	81.90	78.46	72.97	6.592	-0.66	0.638	NS	-
Pancreas	5.02	5.74	5.32	6.95	7.30	6.94	0.404	0.08	0.047	NS	-
Stomach	18.80	19.37	19.78	21.24	24.18	24.15	1.708	0.07	0.168	NS	-
Small intestine	102.21	84.88	95.65	86.83	95.70	100.48	5.403	NS	-	NS	-
Large intestine	52.33	49.26	56.94	49.70	48.62	67.26	6.390	NS	-	NS	-
<b>Lengths (cm / MBWkg)</b>											
Small intestine	126.14	105.10	107.49	103.23	128.15	137.31	8.117	-2.26	0.838	0.07	0.020
Large intestine	22.63	25.30	28.05	25.08	26.45	30.27	2.101	0.14	0.124	NS	-

**SE – standard error**

**Table 5. 2: Histological measurement ( $\mu\text{m} / \text{MBWkg}$ ) of mucosa of the intestinal segments in growing pigs fed diets with incremental levels of fibre**

Histo	Inclusion level (g/kg)						Linear		Quadratic	
	0	80	160	240	320	400	Reg.	SE	Reg.	SE
Duodenum										
VH	25.4 ± 1.68	32.6 ± 1.63	33.6 ± 1.40	28.3 ± 1.73	29.5 ± 1.79	37.0 ± 1.54	0.18	0.089	NS	-
CD	26.0 ± 2.25	33.4 ± 2.19	35.8 ± 1.88	35.0 ± 2.33	37.8 ± 2.41	55.9 ± 2.07	-0.018	0.012	NS	-
VCR	0.97 ± 0.08	0.8 ± 0.07	1.0 ± 0.06	0.8 ± 0.08	0.8 ± 0.07	0.7 ± 0.068	-0.0088	0.0081	NS	-
AVSA	5.9 ± 0.40	6.6 ± 0.39	8.0 ± 0.33	6.7 ± 0.41	7.3 ± 0.43	9.2 ± 0.36	0.018	0.0460	NS	-
Jejunum										
VH	24.3 ± 2.27	30.0 ± 1.80	33.0 ± 2.10	32.2 ± 1.51	29.6 ± 1.85	38.5 ± 1.71	0.22	0.216	NS	-
CD	30.6 ± 1.99	28.9 ± 1.58	32.9 ± 1.84	32.9 ± 1.32	33.9 ± 1.62	39.6 ± 1.50	-0.071	0.182	NS	-
VCR	0.80 ± 0.08	1.0 ± 0.07	1.1 ± 0.08	1.0 ± 0.06	0.9 ± 0.07	1.0 ± 0.06	NS	-	NS	-
AVSA	6.5 ± 0.50	5.7 ± 0.40	6.7 ± 0.46	7.5 ± 0.33	7.6 ± 0.41	9.5 ± 0.38	-0.014	0.0464	0.0024	0.0011
Ileum										
VH	26.2 ± 1.30	27.7 ± 1.48	35.7 ± 1.07	26.3 ± 1.21	34.7 ± 1.17	38.0 ± 1.56	-0.16	0.069	NS	-
CD	24.2 ± 1.90	24.2 ± 2.16	26.2 ± 1.57	25.9 ± 1.77	33.1 ± 1.71	36.9 ± 2.28	-0.12	0.097	0.011	0.0049
VCR	1.3 ± 0.10	1.2 ± 0.11	1.5 ± 0.08	1.0 ± 0.09	1.1 ± 0.09	1.1 ± 0.12	NS	-	NS	-
AVSA	6.2 ± 0.43	5.3 ± 0.50	7.4 ± 0.36	7.4 ± 0.40	7.5 ± 0.39	10.7 ± 0.52	0.015	0.005	NS	-

VH – villi height; CD- crypt depth; VCR- VH : CD ratio; VSA-villi surface area;

Reg. – regression coefficient; SE – standard error; NS – not significant

#### **5.3.4 Correlations between different histological measurements of the small intestines**

Table 5.3 shows correlations of the different histological measurements of mucosa of the small intestines. There was a moderate positive correlation between VH and AVSA ( $P < 0.001$ ). Moderate negative correlations were observed for VH and VCR, and CD and VCR ( $P < 0.01$ ). Apparent villi surface area and CD showed a weak negative correlation ( $P < 0.001$ ).

#### **5.3.5 Effects of physicochemical properties of fibrous diets on histological measurements of the small intestines**

Using stepwise regression, bulk density (BD) and neutral detergent fibre (NDF) were the best predictor variables influencing villi height (VH) and apparent villi surface area (AVSA) ( $P < 0.05$ ). Villi height produced quadratic and linear responses with BD and NDF, respectively ( $P < 0.05$ ; Table 5.4). Conversely, AVSA produced quadratic and linear responses with NDF and BD, respectively ( $P < 0.05$ ; Table 5.4).

### **5.4 Discussion**

From previous studies involving histomorphometric measurements of the intestines (Nyachoti *et al.*, 2000; Marion *et al.*, 2002; Ding *et al.*, 2011), more animals were used to get satisfactory results with small standard deviations within treatments. However, with the current ethical issues which prohibit the unnecessary sacrifices of animals especially when statistical power is high, 3 pigs were therefore subjected to each treatment diet in the study. The number was sufficient enough to produce plausible results for the histomorphometric measurements.

**Table 5. 3: Correlation coefficients between CD and VCR, VCR and AVSA, CD and AVSA; VH and VCR, VH and CD, VH and AVSA**

	<b>VCR</b>	<b>CD</b>	<b>AVSA</b>	<b>VH</b>
<b>VCR</b>	-	-0.67*	-0.097NS	-0.41*
<b>CD</b>		-	-0.25*	-0.29*
<b>AVSA</b>			-	0.44*

\* P < 0.05; NS - not significant (P > 0.05);

VH – villi height; CD- crypt depth; VCR- VH to CD ratio; AVSA- apparent villi surface area

**Table 5. 4: Relationship between physicochemical properties and, villi height (VH) and apparent villi surface area (AVSA)**

y	Components of regression equation			Linear	Quadratic
	$ax^2$	bx	c		
VH	211.3 ± 58.49 (BD) <sup>2</sup>	-591.0 ± 159.06 (BD)	442.4 ± 107.42	*	*
VH		-0.03 ± 0.0047 (NDF)	22.8 ± 1.59	*	NS
AVSA	0.000036 ± 0.0000148 (NDF) <sup>2</sup>	-0.012 ± 0.00969 (NDF)	7.25 ± 1.493	*	*
AVSA		-47.12 ± 39.957 (BD)	45.03 ± 26.985	*	NS

\* P < 0.05; NS - not significant (P > 0.05);

**a** - regression coefficient of  $x^2$ ; **b** - regression coefficient of x; **c** - intercept

NDF – Neutral detergent fibre; BD-Bulk density

The body weight gain of pigs decreased with inclusion level of fibre. The results are in agreement with previous studies (Le Goff *et al.*, 2002; Ndou *et al.*, 2013) which reported a decrease in ADG in growing pigs fed diets high in fibre levels. The observation that feeding diets with incremental levels of fibre produce variable effects on individual visceral organ mass was expected in the study. Increase in relative weight of the pancreas and stomachs agree with previous study (Agyekum *et al.*, 2012). Increase in weight of the pancreas might be attributed to hyper-secretion of pancreatic enzymes in an attempt to compensate for digestion and absorption inefficiency (Pond *et al.*, 1988). At high inclusion levels of fibre, more enzymes are produced to breakdown coarse textured feed compare to lower fibre diets. Decrease in weight of liver, on the other hand, was observed in pigs in the current study. When pigs were fed on lucerne containing diets, an increase in weight of liver was reported (Pond *et al.*, 1988; Jørgensen *et al.*, 1996; Nyachoti *et al.*, 2000). On the contrary, previous study (Ma *et al.*, 2002) reported decreases in liver weights in growing pigs fed on diets with wheat straw. Such conflicting findings suggest that organ weights are influenced by the source of the insoluble fibre used.

The weights of the small and large intestines relative to MBW of pigs were not affected by the fibrous diets. This is consistent with previous studies (Jin *et al.*, 1994). Explanation to this might lie in the chemical composition of the feed offered to the pigs. At higher fibre inclusion levels where energy will be low, the pig will not be getting enough energy for protein accretion (Pond *et al.*, 1988; Jørgensen *et al.*, 1996). Weights of the intestines are, therefore, maintained or to some extent decreased depending on the amount of energy the pigs are able to utilise from the fibrous feed offered. The observed increase in length of large intestines with inclusion level of fibre



might be attributed to the prolonged presence of fibre in the gut. This stimulates an increase in length, which facilitates the development of bacterial mass for fermentation of fibres (Eastwood, 1992).

Villi height and AVSA were positively correlated. Inclusion level of fibre in diets of pigs, therefore, did not alter shape of villi, particularly villi width. The observation that VH and AVSA increase along the gut agrees with previous study (Jin *et al.*, 1994). A possible explanation for the reduced villi length and villi surface area in the duodenum segment of the small intestines may be due to abrasive effects of fibrous diets offered to the pigs on the most apical part of the villi (Agyekum *et al.*, 2012). The abrasive effect of dietary fibre is reduced as feed moves along the gut. Most of the absorption of nutrients occurs in the ileum segment (Ngoc *et al.*, 2012). Therefore, increase length and surface area of villi in the segment is to allow an increase in absorption of nutrients.

Increase in villi height and villi surface area is regarded to be an adaptive mechanism to improve the absorption of nutrients from the gut (Caspary, 1992; Langhout, 1998; Yasar and Forbes, 1999). In growing pigs, inclusion of 10 % wheat straw has been found to increase VH and deeper crypts in the jejunum and ileum, and increased cell division and crypt depth in the large intestine (Jin *et al.*, 1994). In the current study, VH and AVSA increased with inclusion level of fibre in diets. High fibre diets have reduced density of nutrients; hence length and surface area of villi increase absorptive functions in order to capture as much nutrients as possible for nutritional requirements to be met. The crypt region in the intestines is a site of cell regeneration.

A plethora of studies have reported depth of the crypt to increase in response to degeneration of cells of the villi.

The current study has shown that diet quality as measured by bulk values influenced villi morphology. Bulk density of a feed can be described in terms of particle size of feed ingredients. If a feed has got a high bulk density, it means it has got finely ground feed particles. Conversely, a diet low in bulk density has coarse feed particles. Feed with larger particle size have lower rate of passage through the gastrointestinal tract (Warner, 1981), which results in a greater contact between the food and the intestinal mucosa, thereby, increasing villus height (Dahlke *et al.*, 2003). This will result in greater absorption of available nutrients from the digesta. This conforms to our current finding in growing pigs where BD had a significant role in influencing height of villi.

Neutral detergent fibre constitutes hemi-cellulose, cellulose and lignin, and is considered as a complete description of fibre in a diet. Earlier report (Bindelle *et al.*, 2005) showed that increasing the level of NDF in the diet generally decreases digestibility of nutrients. This might account for increased VH and AVSA as NDF increased. Neutral detergent fibre might have triggered a natural increase in absorption and utilisation of nutrients by increasing VH and AVSA as nutrients become more limiting.

## **5.5 Conclusions**

Feeding incremental levels of fibre alters visceral organ development and histomorphometric changes of small intestines in growing pigs. Bulk density and NDF were the best predictor

variables influencing VH and AVSA in segments of the small intestines. Bulk density and NDF provided linear and quadratic relationships with VH and AVSA of growing pigs, respectively. The feeding behaviour of finishing pigs subjected to the fibrous diets also needs to be assessed. This is mainly because of differences in digestive capacities between adult and juvenile pigs

## 5.6 References

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## **Chapter 6: Effects of physicochemical properties of fibrous diets on feeding behaviour of finishing pigs**

*(Submitted to South African Journal of Animal Science)*

### **Abstract**

The objective of the present study was to predict time spent on different behavioural activities by finishing pigs using the physicochemical measurements of fibrous feed. Eighty four Topigs barrows with  $80.5 \pm 4.7$  kg BW at the beginning of the experiment were used. Maize cob, sunflower hulls, Lucerne hay and dried citrus pulp were the fibre sources used. These were included at 0, 80, 160, 240, 320, 400 g/kg in a basal diet. Time spent eating, drinking, lying down, sitting, standing and other activities was observed using video cameras. Neutral detergent fibre and ADF were best predictor variables for time spent eating. They both produced a curvilinear response with time spent eating ( $P < 0.05$ ). Time spent drinking was observed to increase linearly as ADF increased in diets of pigs ( $P < 0.001$ ). Water holding capacity produced a quadratic response with time spent lying down ( $P < 0.05$ ), whilst ADF and NDF, on the other hand, produced negative linear responses with time spent lying down ( $P < 0.001$ ). Time spent standing increased as ADF increased ( $P < 0.001$ ). Water holding capacity and ADF were best predictor variable influencing number of visits made by pigs to the feeder ( $P < 0.05$ ). Water holding capacity influenced duration of visits to the feeder ( $P < 0.05$ ). It can be concluded that time spent on different behavioural activities by finishing pigs is influenced by fibre source and inclusion level.

**Keywords:** Dietary fibre; Feeding behaviour; Finishing pigs; Water holding capacity



## 6.1 Introduction

The ability of pigs to utilize fibrous feeds is influenced by age (Bindelle *et al.*, 2008; Ndou *et al.*, 2013). Adult pigs have higher digestive capacity and higher microbial activity due to the development of their large intestines and caecum than juvenile pigs (Jørgensen *et al.*, 1996; Freire *et al.*, 2000; Renteria-Flores *et al.*, 2008). Feeding behavioural patterns in adult and juvenile pigs fed fibrous diets are, therefore, likely to differ.

Feeding behaviour are any actions that are directed towards the procurement of nutrients and can be influenced by physicochemical properties of the fibrous feeds. Examples of the physicochemical properties are; water holding capacity (WHC), bulk density (BD), acid detergent fibre (ADF) and neutral detergent fibre (NDF). The physicochemical properties vary across fibre sources (Ndou, 2012). Their bulking properties stimulate stretch receptors on the stomach walls due to distension leading to a gradual decrease in time spent eating (Lepionka *et al.*, 1997; De Leeuw, 2004).

Chapter 3 highlighted that physicochemical properties of the fibrous diets influence time spent on different behavioural activities in growing pigs. Very few, if any studies, have been conducted to determine the influence of physicochemical properties of fibrous diets on time spent on different behavioural activities in finishing pigs. The use of different fibre sources provides a wide range of physicochemical properties. More data points are, therefore, produced, which will enhance accurate estimation of the relationship between time spent on different behavioural activities and physicochemical properties of the different fibre sources. Time spent on different behavioural activities might is a first step in providing an insight on how specific

physicochemical characteristics of different fibre sources affect satiety levels in pigs. The objective of the study was, therefore, to predict time spent on different behavioural activities from physicochemical properties of fibrous diets in finishing pigs. It was hypothesized that physicochemical measures of a feed adequately describe time spent on different behavioural activities by finishing pigs.

## **6.2 Material and methods**

### **6.2.1 Pigs and housing**

Eighty four clinically healthy male pigs were used in the experiment. Bodyweight of the pigs at the beginning of the experiment was  $80.5 \pm 4.7$  kg. Ethical approval was obtained from the University of KwaZulu-Natal Animal Ethics Sub-Committee (Reference Number: 061/12/Animal). Pigs were randomly allocated to individual pens measuring  $2.0 \times 1.1$  m with concrete floors. Walls of the pen were not solid and pigs had contact with and could see each other. The pen was equipped with a plastic self-feeder trough (Big Dutchman Lean Machine®) designed to minimise spillages and a low-pressure nipple drinker providing water *ad-libitum*. The ambient temperature and relative humidity were recorded automatically throughout the trial using HOBO data loggers (Onset Computer Corporation, Pocasset, MA, USA).

### **6.2.2 Diets and feeding**

A commercial feed (Grower - Finisher, Meadow Feeds Ltd) was used as the basal feed. The fibre sources used to dilute the basal feed were maize cob (MC), sunflower hulls (SH), Lucerne hay (LH) and dried citrus pulp (PU). The idea of using the 4 fibre sources was to expand the range of physicochemical properties. The fibre sources were first ground to pass through a 3 mm screen.

They were then included at different inclusion levels of 0, 80, 160, 240, 320 and 400 g/kg in the basal diet. A total of 21 treatment diets were, therefore, formulated by diluting the basal diet with each of the fibre sources. Ingredients used in the formulation of basal diet contained 500 g/kg yellow maize, 158 g/kg soya bean, 20.2 g/kg soybean oil cake, 163 g/kg wheat bran, 85 g/kg sunflower oil cake, 25 g/kg molasses syrup and 48.8 g/kg additives. The pigs were allocated to treatment diets on an ad-libitum basis for 31 days, including 10 days of adaptation.

### **6.2.3 Analyses of chemical composition and physical properties of diets**

Methods used for analyses of the diets in the trial have been described in chapter 3. The chemical composition and bulk properties of the fibrous diets are shown in Table 6.1.

### **6.2.4 Behavioural measurements**

The behavioural activities for each pig were continuously recorded for 8h per day (0800h – 1600h) once a week for three consecutive weeks. Most of the pigs in the study were inactive (lying down) after 1600h and became active after 0800h the next morning when they were offered feed. The behaviours were recorded with the aid of video cameras mounted on the ceiling in such a way that each camera focused on six pigs while preventing disturbances to the pigs whilst they are exhibiting their day-to-day normal behavioural activities. The behavioural activities recorded were time spent eating, drinking, standing, sitting, lying down and other activities (object biting and licking). The behavioural activities have been described in table 3.4.

Behaviour was recorded by the same person throughout the whole experiment which lasted for four weeks. Number of visits to the feeder and duration of each visit on the feeder were also recorded.

**Table 6. 1: Chemical composition and bulk characteristics of diets**

<b>Fibre source</b>	<b>Inclusion level (g/kg)</b>	<b>DM (g/kg)</b>	<b><sup>1</sup>calc DE (MJ/kg)a</b>	<b>CP (g/kg DM)</b>	<b>EE (g/kg DM)</b>	<b>Ash (g/kg DM)</b>	<b>NDF (g/kg DM)</b>	<b>ADF (g/kg DM)</b>	<b>BD(g DM/ml)</b>	<b>WHC (g water/g DM)</b>
Basal diet	0	90.14	10.75	247.8	3.73	5.52	430.93	68.39	1.83	4.6
MC	80	90.05	10.56	218.5	4.88	5.28	454.44	170.57	1.75	4.5
MC	160	90.91	10.56	207.9	5.45	5.04	470.3	185.59	1.65	5.71
MC	240	90.35	9.52	199.1	3.4	4.55	477.09	236.12	1.64	4.8
MC	320	90.79	9.51	168.7	3	4.36	505.41	325.75	1.47	5.94
MC	400	90.69	9.42	145.2	2.91	4.38	506.83	256.96	1.44	5.94
LH	80	87.59	9.84	178.8	5.09	5.75	450.62	126.02	1.75	4.06
LH	160	88.23	8.64	177.8	5.14	5.6	537.03	183.5	1.69	5.61
LH	240	88.07	8.01	179.4	3.46	6.8	548.06	205.97	1.65	5.28
LH	320	88.59	9.54	176.2	4.49	6.62	476.99	285.88	1.47	7.32
LH	400	88.25	8.91	173.1	3.61	6.41	471.05	303.55	1.44	6.6
PU	80	87.75	9.36	180.2	4.48	5.07	475.35	165.91	1.86	4.07
PU	160	91.61	10.03	160.6	4.94	5.34	434.68	202.89	1.77	6.39
PU	240	88.53	10.23	147.6	2.75	5.13	411	229.59	1.75	6.34
PU	320	88.07	10.47	143.5	4.36	5.17	404.67	306.99	1.73	6.36
PU	400	87.95	11.05	139.6	3.25	5.23	362	277.17	1.73	7.42
SH	80	88.02	8.40	168.7	4.68	4.84	541.69	164.32	1.76	4.58
SH	160	87.78	8.91	158.9	4.45	4.71	511.54	310.24	1.69	4.2
SH	240	88.17	9.65	151.6	4.26	4.38	475.99	285.43	1.61	4.08
SH	320	90.09	8.94	134.6	4.6	3.9	535.81	452.44	1.54	4.87
SH	400	88.78	8.49	124.6	3.13	4.18	565.81	457.06	1.47	4.8

LH – Lucerne hay; SH – Sunflower hulls; MC –Maize cob; PU – dried citrus pulp.

DM – Dry matter; CP – Crude protein; EE – Ether extract; calc DE – calculated digestible energy; NDF – neutral detergent fibre; ADF – acid detergent fibre; BD – bulk density; WHC – water holding capacity.

<sup>1</sup> calc DE = 949 + (0.789 × GE) - (43 × % Ash) - (41 × % NDF) (Noblet and Perez,1993).

Time spent on different behavioural activities was divided by body weight of a pig. Results are, therefore, reported as relative time spent on different behavioural activities to body weight of the pig.

Body weight and feed intake were monitored weekly, and the results were used to calculate average gaily gain (ADG) and average daily feed intake (ADFI). Feed intake was determined every week by weighing the feed supplied and feed left. Feed spillages were collected by placing a plastic tray under each trough and were taken into consideration when doing calculations for intake.

#### **6.2.5 Statistical analyses**

To test for significance of fibre source, inclusion level and interactions on time spent on each behavioural activity, ADFI and ADG, General Linear Model procedure of SAS software (SAS, 2008) was used. Relationship between time spent eating and ADFI was analysed using PROC CORR procedure of SAS (2008). The STEPWISE procedure of SAS (2008) was used to determine best predictor variables for time spent on different behavioural activities. Variables entered into the model as predictor variables for time spent on different behavioural activities were: Dry matter, DE, CP, EE, ash, NDF, ADF, BD and WHC. Variables selected as best predictors of relative time spent eating to body weight of a pig were subjected to further analysis to determine the relationship existing with time spent eating using quadratic response surface model (SAS, 2008). Relationships between number of visits to the feeder and duration of each visit, and inclusion level were determined using the quadratic response surface model (SAS, 2008).

## **6.3 Results**

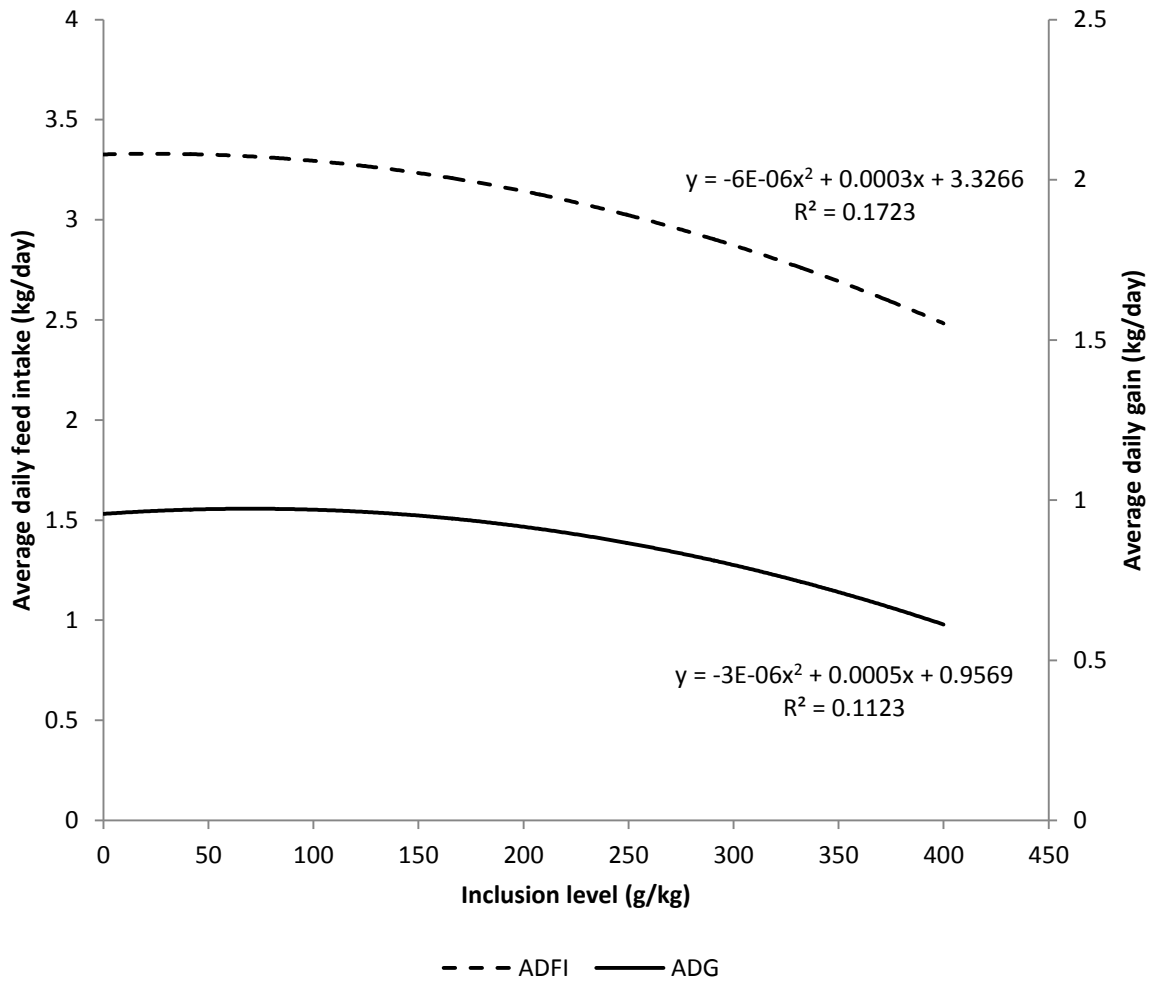
### **6.3.1 Effect of feeding different fibre sources at different inclusion levels on pig**

#### **performance and time spent eating and drinking**

Figure 6.1 shows the effect of inclusion level of fibres on ADFI and ADG of finishing pigs. Average daily feed intake and ADG decreased as inclusion level of fibres increased ( $P < 0.05$ ). Time spent eating for all fibre source increased with inclusion level (Table 6.2). Maize cob and LH showed a linear increase in time spent eating with inclusion level of fibre ( $P < 0.05$ ). Conversely, PU and SH showed both linear and quadratic relationships ( $P < 0.05$ ). There was no relationship observed for time spent drinking as inclusion level PU increased ( $P > 0.05$ ; Table 6.2). Other fibrous sources (MC, LH and SH) showed positive linear responses between time spent drinking and inclusion level of fibre ( $P < 0.05$ ). There was a strong negative correlation between time spent eating and ADFI ( $r = -0.54$ ;  $P < 0.05$ ).

#### **6.3.2 Effect of feeding different fibre sources on time spent on different postures (lying down, standing and sitting) and other behavioural activities**

Table 6.3 shows the effect feeding fibrous diets on time spent lying down, standing and other activities by finisher pigs. Time spent lying down decreased as inclusion level of LH, MC and SH increased ( $P < 0.01$ ). No differences were observed for time spent lying down as inclusion level of PU increased ( $P > 0.05$ ). A positive linear relationship was observed between time spent standing and inclusion level of MC ( $P < 0.01$ ). There were no relationships for time spent standing and inclusion level of other fibrous sources (PU, LH and SH) ( $P > 0.05$ ). Time spent on other behavioural activities (chewing bars, scratching) was noted to decrease as inclusion level LH increased ( $P < 0.05$ ).



**Figure 6. 1: Effect of inclusion level on average daily feed intake (ADFI) and average daily gain (ADG) of finishing pigs**

**Table 6. 2: Effect of fibre source and inclusion level on time spent eating (E) and drinking (D) by finisher pigs**

Activity	Fibre source	Inclusion level (g/kg)						P-value	
		0	80	160	240	320	400	Linear	Quadratic
E	PU	0.91	0.84	0.73	0.91	1.13	1.74	**	*
	LH	0.89	1.05	1.12	1.17	1.68	2.04	**	NS
	MC	0.88	0.72	1.20	1.35	2.01	2.43	***	NS
	SH	0.91	0.69	1.38	1.35	3.14	4.46	***	**
D	PU	0.17	0.14	0.15	0.20	0.24	0.22	NS	NS
	LH	0.16	0.20	0.30	0.18	0.41	0.38	*	NS
	MC	0.16	0.21	0.29	0.20	0.34	0.32	*	NS
	SH	0.18	0.15	0.26	0.20	0.33	0.56	**	NS

\*P < 0.05; \*\* P < 0.01; \*\*\* P < 0.001; NS - not significant (P > 0.05)

LH – Lucerne hay; SH – Sunflower Hulls; MC –Maize cob; PU – dried citrus pulp



**Table 6. 3: Effect of fibre source and inclusion level on time spent lying down, standing and other activities by finisher pigs**

Activity	Fibre source	Inclusion level (g/kg)						P-value
		0	80	160	240	320	400	Linear
Lying down	PU	3.52	2.98	3.21	3.03	2.94	3.55	NS
	LH	3.35	3.15	2.56	2.88	2.28	2.10	**
	MC	3.32	3.07	2.88	2.82	2.24	2.12	**
	SH	3.33	2.52	2.77	2.82	2.10	0.82	**
Standing	PU	0.08	0.28	0.21	0.43	0.44	0.33	NS
	LH	0.62	0.33	0.75	0.24	0.46	0.56	NS
	MC	0.46	0.41	0.50	0.92	0.99	0.69	*
	SH	0.74	0.51	0.69	0.37	0.60	0.24	NS
Other activities	PU	0.06	0.02	0.01	0.06	0.08	0.04	NS
	LH	0.06	0.04	0.00	0.02	0.02	0.03	*
	MC	0.00	0.02	0.03	0.14	0.03	0.04	NS
	SH	0.03	0.03	0.01	0.23	0.04	0.42	NS

\*P < 0.05; \*\* P < 0.01; NS - not significant (P > 0.05)

LH – Lucerne hay; SH – Sunflower Hulls; MC –Maize cob; PU – dried citrus pulp

### **6.3.3 Effect of physicochemical properties of fibrous diets on time spent on different behavioural activities**

Table 6.4 shows relationships of best predictor variables (bulk properties) of relative time spent on different behavioural activities to body weight of finishing pigs. Neutral detergent fibre and ADF were the best predictor variables for time spent eating. Both NDF and ADF showed a curvilinear response with time spent eating ( $P < 0.05$ ). As NDF and ADF increased, time spent eating also increased. Time spent drinking was observed to increase linearly as ADF increased in diets of pigs ( $P < 0.001$ ). Water holding capacity, ADF and NDF were the best predictor variables for time spent lying down. Water holding capacity produced a quadratic response with time spent lying down ( $P < 0.05$ ) whilst ADF and NDF, on the other hand, produced negative linear responses with time spent lying down ( $P < 0.001$ ). Time spent standing increased as ADF increased in diets of pigs ( $P < 0.001$ ).

### **6.3.4 Effect of physicochemical properties of fibrous diets on number of visits to the feeder and duration of each visit**

Water holding capacity and ADF influenced number of visits made by pigs to the feeder ( $P < 0.05$ ; Figure 6.2). Number of visits to the feeder decreased linearly as WHC and ADF increased in diets of pigs. Water holding capacity influenced duration of each visit on the feeder ( $P < 0.05$ ; Figure 6.3).

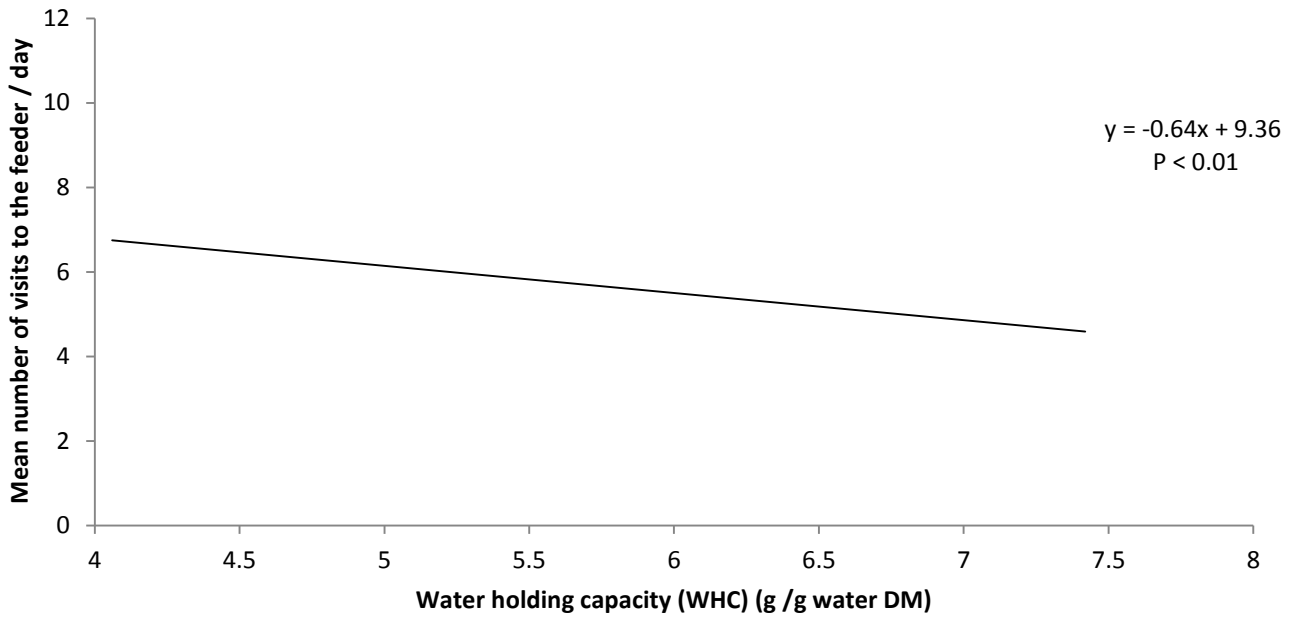
**Table 6. 4: Relationships of best predictor variables (bulk properties) of relative time spent on different behavioural activities to body weight of finishing pigs**

Activity	Physicochemical property	Components of a regression			P-value	
		$ax^2$	$bx$	$c$	Linear	Quadratic
Eating	NDF	$7.39 \times 10^{-5} \pm 2.84 \times 10^{-5}$	$-0.064 \pm 0.027$	$14.91 \pm 6.179$	**	*
	ADF	$2.68 \times 10^{-5} \pm 6.98 \times 10^{-6}$	$-0.006 \pm 0.004$	$1.20 \pm 0.466$	***	**
Drinking	ADF	$1.30 \times 10^{-6} \pm 1.47 \times 10^{-6}$	$0.0005 \pm 0.001$	$0.10 \pm 0.098$	***	NS
Lying down	WHC	$1.52 \times 10^{-1} \pm 7.36 \times 10^{-2}$	$-1.726 \pm 0.822$	$7.47 \pm 2.225$	NS	*
	ADF	$-1.35 \times 10^{-5} \pm 7.57 \times 10^{-6}$	$0.003 \pm 0.004$	$2.96 \pm 0.506$	***	NS
	NDF	$-1.68 \times 10^{-6} \pm 2.28 \times 10^{-5}$	$-0.005 \pm 0.021$	$5.46 \pm 4.961$	***	NS
Standing	ADF	$6.61 \times 10^{-7} \pm 3.50 \times 10^{-6}$	$0.002 \pm 0.002$	$0.01 \pm 0.233$	***	NS

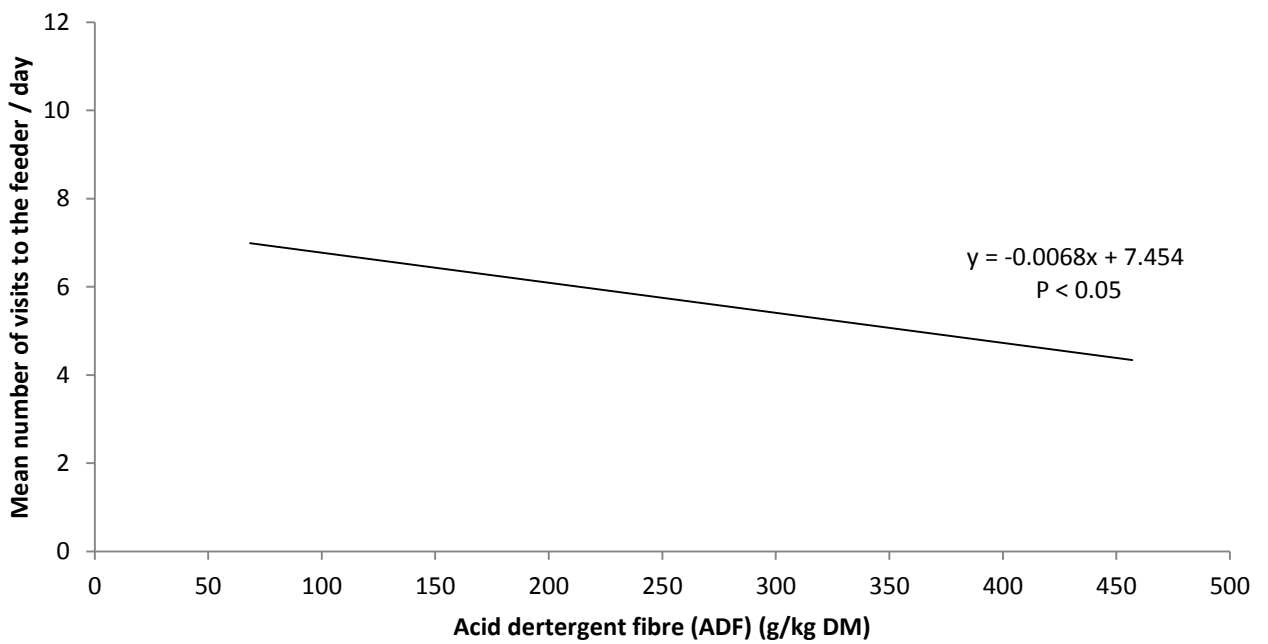
\*P < 0.05, \*\* P < 0.01, \*\*\* P < 0.001, NS - not significant (P > 0.05);

NDF – neutral detergent fibre, ADF – acid detergent fibre, WHC – water holding capacity;

**a** - regression coefficient of  $x^2$ ; **b** - regression coefficient of  $x$ ; **c** - intercept

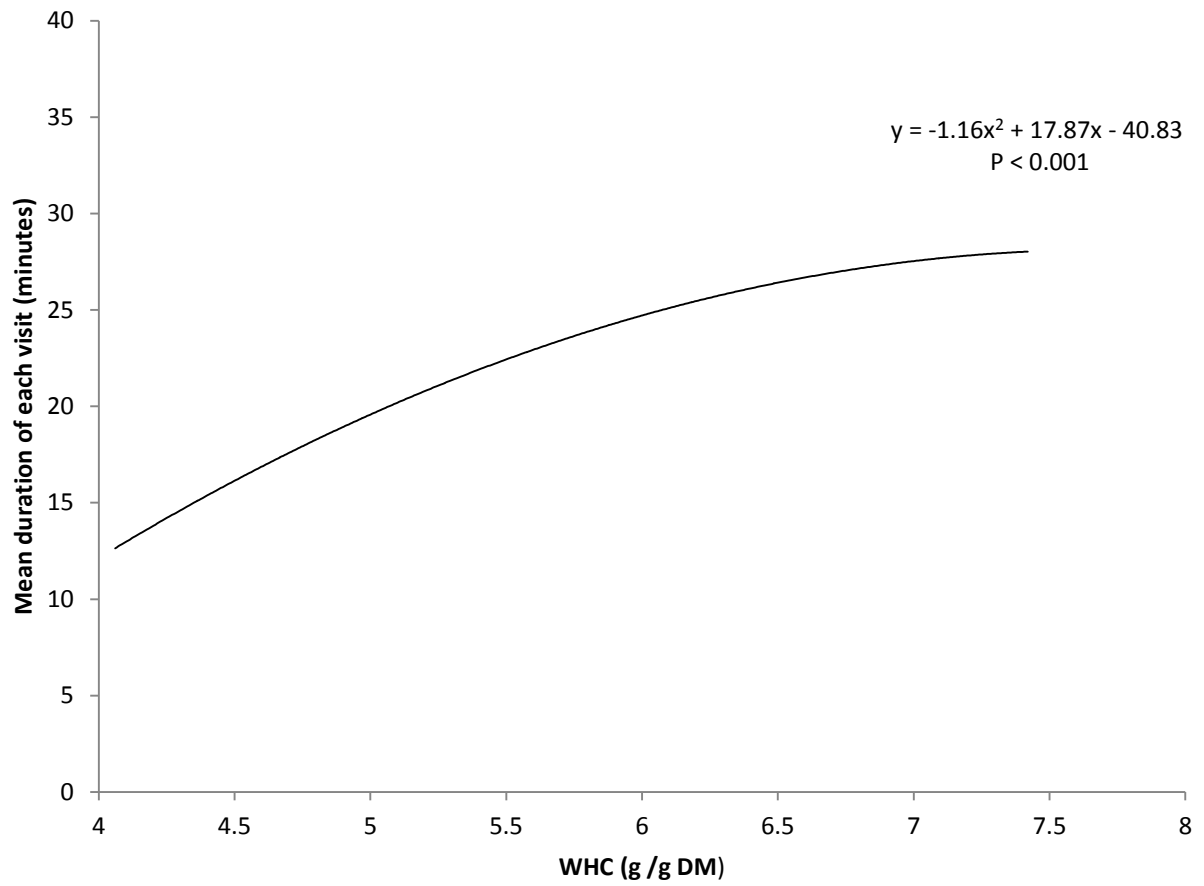


(a)



(b)

**Figure 6. 2: Relationship between (a) Water holding capacity (WHC) (b) acid detergent fibre (ADF) and number of visits to the feeder**



**Figure 6. 3: Relationship between water holding capacity and mean duration of each visit on the feeder**

## 6.4 Discussion

Physicochemical properties of a feed describe time spent on different behavioural activities in pigs. In the current study, it was expected that the behavioural activities, particularly time spent eating, would reflect gut capacity in finishing pigs due to the bulking effects of the physicochemical properties. Gut capacity in this case is noticed when time spent eating started to decrease. Inclusion levels of fibres for optimal productivity were, therefore, obtained based on physicochemical properties of feed and also time spent eating.

Plant species have different fibrous matrices and composition (Ndou *et al.*, 2013). Such variation in composition of the plant species might account for differences obtained for time spent on different behavioural activities (eating, drinking, lying down, standing, sitting down and other behavioural activities) in the current study.

The observation that time spent eating increased with inclusion level of fibre was expected. This is in agreement with previous studies which also showed pigs to increase intake or time spent eating with increasing levels of fibre (Kyriazakis and Emmans, 1995; Brouns *et al.*, 1997; Rushen *et al.*, 1999; Ndou *et al.*, 2013). Progressive inclusion of fibres tends to dilute a high digestible energy diet to a low digestible energy diet. The low levels of digestible energy promote an increase in feed intake to compensate for the reduced energy density in feed (Rijnen *et al.*, 2003).

Acid detergent fibre and NDF can be described as indigestible components of a feed. The selection of ADF as best predictor for time spent eating in finishing pigs was expected. More time spent eating with an increase in ADF content is attributed to an increase in chewing activity at higher fibre inclusion level in diets (Wenk, 2001). Adult pigs have much more

developed masseter muscles and are expected to chew better than growing pigs (Herring, 1977; Herring and Wineski, 1986; Xiaofeng *et al.*, 1994). Hence, time spent eating was for adult pigs was reduced at high ADF content in diets. Continual increase in time spent eating in finisher pig could also be attributed to bite sizes made by the pigs. Larger bite sizes will mean more time spent chewing and also mixing of feed in the mouth. Bite sizes made by pigs in the current study were, however, not measured and warrants further investigations.

Decrease in time spent lying down came entirely from an increase in time spent eating in the present study with finisher pigs. Lying down might be a sign of satiety in growing pigs receiving low fibrous diets. Time spent standing, on the other hand, could explain levels of alertness to sound and movement which pigs exhibit as they anticipate a much more palatable feed from stockmen.

The introduction of PU in diets of finishing pigs increased range of WHC values. According to De Leeuw (2004) and Lepionka *et al.* (1997), high WHC result in the swelling of feed in the stomach which cause distension of the stomach walls. This will lead to satiety, which consequently reduce time spent eating. This can possibly explain the decrease in mean duration of each visit as WHC increased in the study. Kyriazakis and Emmans (1995) have also shown high water holding capacity to maintain the physical satiate for longer time, thus, lowering the stimulus to feed. Maintenance of physical state for longer time might have also resulted in increased time spent lying down in pigs. Hence, number of visits to the feeder is reduced especially in feeds high in WHC. Although indigestible fractions of feed (NDF and ADF) were not selected using stepwise regression, they could have probably influenced duration of visit on the feeder. With duration on the feeder increased owing to more time spent chewing the indigestible material.

Decrease in number of visits to the feeder in the study came entirely from an increase in duration of visit on the feeder. Increasing inclusion level of fibres dilutes the diets; hence, at higher inclusion levels of fibre in a diet pigs spend more time at the feeder in a bid to get more feed with nutrients which meet their requirements.

## **6.5 Conclusions**

It can be concluded that time spent on different behavioural activities is influenced by fibre source and inclusion level. Water holding capacity (WHC), ADF and NDF are physicochemical properties selected by stepwise regression to predict time spent on different behavioural activities, (eating drinking, lying down, standing and other activities). The physicochemical properties provided relationships with time spent on different behavioural activities.

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## **Chapter 7: Growth performance, nutritional status and behaviour of finishing pigs fed graded levels of fibrous diets**

### **Abstract**

The objectives of the study were to determine effect of feeding fibrous diets on growth performance, time spent eating and nutritional status of the finishing pigs and to determine blood metabolites that best describe time spent eating. A total of 84 clinically healthy male pigs were used in the experiment. Bodyweight of the pigs at the beginning of the experiment was  $85 \pm 10.1$ kg. Treatment diets used contained four fibre sources, namely maize cob (MC), sunflower hulls (SH), lucerne hay (LH) and dried citrus pulp (PU). The fibres were included at different inclusion levels of 0, 80, 160, 240, 320 and 400 g/kg in a basal diet of finisher pigs. Average daily feed intake (ADFI) decreased as level of PU, LH, MC and SH increased ( $P < 0.05$ ). A linear decrease in ADG as level of PU and SH increased was also observed ( $P < 0.05$ ). There was a quadratic increase in serum total concentration (TP) as level of PU and LH increased ( $P < 0.05$ ). A positive linear relationship was, however, observed between TP concentration and inclusion levels of MC ( $P < 0.05$ ). There was a linear decrease in CK concentration as levels of PU, LH and MC increased ( $P < 0.05$ ). Time spent eating increased with inclusion level of fibre ( $P < 0.05$ ). Serum total protein was selected as the best predictor variable influencing relative time spent eating to body weight of pig (RTSE) ( $P < 0.05$ ). The equation for the relationship was:  $RTSE = -111.88(TP) + 57.67(TP) - 6.34$ . It can be concluded that PU, LH, MC and SH influenced average daily feed intake, average daily gain, time spent eating and nutritionally-related blood metabolites of the finishing pigs. Serum total protein was the best predictor variable for predicting time spent eating.

**Keywords:** Behaviour; Blood metabolites; Finishing pigs; Fibres; Nutritional status

## 7.1 Introduction

Use of alternative fibrous feeds to feed pigs is receiving increasing attention because it reduces competition for grain with humans. Furthermore, feed costs are reduced largely by reducing the proportion of grain that is usually more expensive (Mashatise *et al.*, 2005). There is, however, a need to determine how the pigs will respond in terms of growth performance, behaviour and nutritional status when offered the new diet. The information will enable feed compounders to formulate a diet from fibrous material which will allow finishing pigs to ingest sufficient nutrients to optimise performance.

Nutritional status of pigs may be monitored by blood metabolites such as urea, uric acid, plasma proteins, glucose (glycated haemoglobin) and creatine kinase. Urea, uric acid, plasma proteins, glycated haemoglobin and creatine kinase have been measured in several fields of studies (Fischer *et al.*, 2000; Shahbazkia *et al.*, 2010; Liotta *et al.*, 2012). Feeding behaviour, on the other hand, can be determined by using time as a unit of measurement. Nutritionally-related blood metabolites are also useful proxies which determine when the pig will stop eating the feed. In chapter 4 using growing pigs, blood metabolites have been observed to influence time spent eating. There is also a need to know if the metabolites can also influence time spent eating in finishing pigs. Very few, if any studies, have been documented on how the metabolites influence time eating in finishing pigs. The objectives of the study were, therefore, to:

1. Determine effect of feeding fibrous diets on growth performance, time spent eating and nutritional status of the finishing pigs; and
2. Determine blood metabolites that best describe time spent eating such that level of satiety of finishing pigs can be predicted using the measurements.

The hypotheses tested were:

1. There are no differences in growth performance parameters, time spent eating and nutritional status of the finishing pigs fed graded levels of fibre and
2. Time spent eating increase linearly to the point where an optimum level is reached and no further increase will occur. This point should be related to some metabolites in blood of the finishing pigs.

## **7.2 Materials and methods**

### **7.2.1 Pigs and housing**

A total of 84 clinically healthy male pigs were used in the experiment. Initial body weight for the pigs at the beginning of the experiment was  $85 \pm 10.1$ kg. Ethical approval was obtained from the University of KwaZulu-Natal Animal Ethics Sub-Committee (Ref 061/12/Animal). Pigs were randomly allocated to individual pens measuring  $2.0 \times 1.1$  m with concrete floors. Walls of the pen were not solid as the pigs could see each other. The pen was equipped with a plastic self-feeder trough (Big Dutchman Lean Machine®) designed to minimise spillages and a low-pressure nipple drinker providing water ad libitum. The ambient temperature and relative humidity were recorded automatically throughout the trial using HOBO data loggers (Onset Computer Corporation, Pocasset, MA, USA). The average temperature and humidity was  $21.15 \pm 2.74^{\circ}\text{C}$  and  $42.64 \pm 1.56$  %.

### **7.2.2 Diets and feeding management**

Diets and feeding management have been described in Chapter 6.

### **7.2.3 Analyses of chemical composition and physical properties of diets**

Methods used for analyses of the diets in the trial have been described in Chapter 3. The chemical composition and bulk properties of the fibrous diets are shown in Table 6.1.

#### **7.2.4 Measurements**

All pigs were weighed after every seven days. Feed intake was determined as pigs were being weighed. This was done by weighing feed supplied in and feed left each week. Behavioural activities were observed with video cameras for 24 hrs. Most of the pigs in the study were inactive after 1600h and become active after 0800h the next morning when they were offered feed. Hence, behavioural activities were recorded for eight hours a day (0800h – 1600h) once a week for three consecutive weeks. Video cameras were mounted in such a way that each camera focused on six pigs while preventing disturbances to the pigs whilst they are exhibiting their day-to-day normal behavioural activities. The all occurrence sampling method, which involve continuous recording all behavioural activities was used. Number of visits to the feeder and duration of each visit on the feeder were recorded. The behavioural activities recorded were the time spent eating (time spent with the head in the feeder) and drinking (time spent with mouth on the nipple drinker). Behaviour was recorded by the same person throughout the whole experiment which lasted for four weeks. Time spent on different behavioural activities was divided by body weight of the pig. Results are, therefore, reported as relative time spent on different behavioural activities to body weight of the pig.

#### **7.2.5 Blood collection and analyses**

Methods of collecting blood and analyses have been described in Chapter 4.

#### **7.2.6 Calculations and statistical analyses**

The data for each pig on time spent eating and concentration of nutritionally-related blood metabolites were adjusted by the metabolic body weight (MBW) to give relative measurements. Metabolic body weight of each animal was calculated using the formula:

Metabolic body weight =  $\mathbf{BW}^{0.75}$

Where;

**BW** - body weight of a pig

A factorial experiment with a common control treatment was used to test for significance of fibre source and inclusion level on growth performance parameters, time spent eating and concentration of nutritionally-related metabolites using the General Linear Model procedure of SAS (SAS, 2008). Quadratic response surface model (SAS, 2008) was used to determine the relationship between growth performance parameters, time spent eating and concentration of nutritionally-related metabolites, and inclusion level of fibre. The STEPWISE procedure of SAS (2008) was used to determine best predictor variables for time spent on different behavioural activities. Variables entered into the model as predictor variables for time spent eating are HbG, TP, albumin, globulin, urea, uric acid levels and CK. Variables selected as best predictors of relative time spent eating to body weight of a pig were subjected to further analysis to determine the relationship existing with time spent eating using quadratic response surface model (SAS, 2008). The broken stick analysis model as described by Robbins *et al.* (2006) was conducted using NLIN procedure (SAS, 2008) to estimate the optimum concentrations of blood metabolites which reduce time spent eating.

## **7.3 Results**

### **7.3.1 Summary statistics**

Table 7.1 shows the summary statistics for growth performance parameters, feeding behaviour and concentration of blood metabolites of pig fed graded levels of fibre sources in diets. For performance parameters, effect of inclusion level of fibre in diets affected ADFI ( $P < 0.001$ ) and ADG ( $P < 0.01$ ).

**Table 7. 1: Summary statistics for growth performance parameters, feeding behaviour and concentration of blood metabolites of pig fed graded levels of fibre sources in diets**

	<b>F</b>	<b>I</b>	<b>F × I</b>
<b>Performance parameters</b>			
ADFI	NS	***	NS
ADG	NS	**	NS
FCR	NS	NS	NS
<b>Feeding behaviour</b>			
Time spent eating	NS	*	NS
<b>Blood metabolites</b>			
Total protein	*	*	*
Albumin	NS	*	*
Globulin	NS	NS	NS
Urea	NS	NS	NS
Uric acid	NS	**	**
CK	NS	***	**
HbG	*	*	NS

\*P < 0.05; \*\* P < 0.01; \*\*\* P < 0.001; NS - not significant (P > 0.05);

F – Effect of fibre source; I – Effect of inclusion level; F × I – interaction of fibre source and inclusion level

ADFI – Average daily feed intake ; ADG - Average daily gain; FCR – feed conversion ratio; CK – creatine kinase; HbG – glycated haemoglobin



Feed conversion ratio was not affected by inclusion level of fibre in diets of pigs ( $P > 0.05$ ). Effect of fibre source and fibre source  $\times$  inclusion level interaction did not affect all the performance parameters (ADFI, ADG and FCR) ( $P > 0.05$ ). Inclusion level of fibre affected time spent eating by finishing pigs ( $P < 0.05$ ).

For blood metabolite concentrations, effect of fibre source only affected concentration of TP and HbG in blood of pigs ( $P < 0.05$ ). Effect of inclusion level of fibres on concentration of TP, uric acid, albumin, CK and HbG in blood of pigs was significant ( $P < 0.05$ ). Only concentrations of globulin and HbG in blood of pigs were not significant ( $P > 0.05$ ) for effect of fibre source  $\times$  inclusion level interaction.

### **7.3.2 Effect of feeding fibrous diets on ADFI, ADG and FCR of finishing pigs**

The influence of PU, LH, MC and SH inclusion level on ADFI, ADG and FCR of finishing pigs is shown in Table 7.2. Average daily feed intake (ADFI) decreased linearly with inclusion level for all the fibre based diets (PU, LH MC and SH) ( $P < 0.05$ ). There was a linear decrease in ADG as level of PU and SH increased ( $P < 0.05$ ). Relationships between inclusion level of LH, MC and SH, and FCR were not significant. However, relationship between inclusion level of PU and FCR of pigs was significant ( $P < 0.05$ ).

1 **Table 7. 2: Effect of fibre source and inclusion level on average daily feed intake (ADFI) , average daily gain (ADG) and feed conversion**  
 2 **ratio (FCR)**

Performance parameter	Fibre source	Inclusion (g/ kg)						SEM	<i>P - value</i>	
		0	80	160	240	320	400		Linear	Quadratic
ADFI (kg /day)	PU	3.00	3.23	3.09	3.29	2.89	2.59	0.231	*	NS
	LH	3.60	2.92	3.66	3.20	2.26	2.38	0.231	**	NS
	MC	3.58	3.31	2.98	3.21	2.04	2.80	0.231	*	NS
	SH	3.41	3.26	3.43	3.34	2.67	2.61	0.231	*	NS
ADG (kg /day)	PU	1.01	1.06	0.90	0.95	0.77	0.53	0.137	*	NS
	LH	0.84	1.08	1.14	0.93	0.77	0.74	0.137	NS	NS
	MC	0.91	0.93	0.81	0.92	0.68	0.75	0.137	NS	NS
	SH	0.98	0.92	0.95	0.85	0.57	0.59	0.137	*	NS
FCR	PU	0.35	0.34	0.30	0.29	0.27	0.22	0.0808	*	NS
	LH	0.23	0.39	0.32	0.31	0.37	0.31	0.0808	NS	NS
	MC	0.26	0.30	0.27	0.29	0.35	0.27	0.0808	NS	NS
	SH	0.28	0.30	0.28	0.26	0.52	0.23	0.0808	NS	NS

3 \*P < 0.05; \*\* P < 0.01; NS - not significant (P > 0.05);

4 ADF - average daily gain; ADG – average daily gain; FCR – Feed conversion ratio

5 MC - maize cob; SH - sunflower hulls; LH - Lucerne hay; PU -dried citrus pulp

6

### **7.3.3 Effect of feeding fibrous diets on nutritionally related blood metabolites in finishing pigs**

Table 7.3 shows effect of fibre source and inclusion level on concentration of blood metabolites. There was a quadratic increase in TP concentration as level of PU and LH increased ( $P < 0.05$ ). A positive linear relationship was, however, observed between TP concentration and inclusion levels of MC ( $P < 0.05$ ). There was a linear decrease in CK concentration as levels of PU, LH and MC increased ( $P < 0.05$ ). Glycated haemoglobin concentration in blood of pigs showed positive linear responses with inclusion level of LH ( $P < 0.05$ ) and SH ( $P < 0.01$ ) in diets of pigs.

### **7.3.4 Relationship between nutritionally related blood metabolites and time spent eating**

Time spent eating increased with inclusion level of fibre ( $P < 0.05$ ; Figure 7.1). Using stepwise regression, TP was the most important blood metabolite selected to influence RTSE ( $P < 0.05$ ). A curvilinear response observed between RTSE and TP concentration. Optimum level of TP concentration in the blood which reduced time spent eating was at  $0.26 \pm 0.047$  (Figure 7.2).

## **7.4 Discussion**

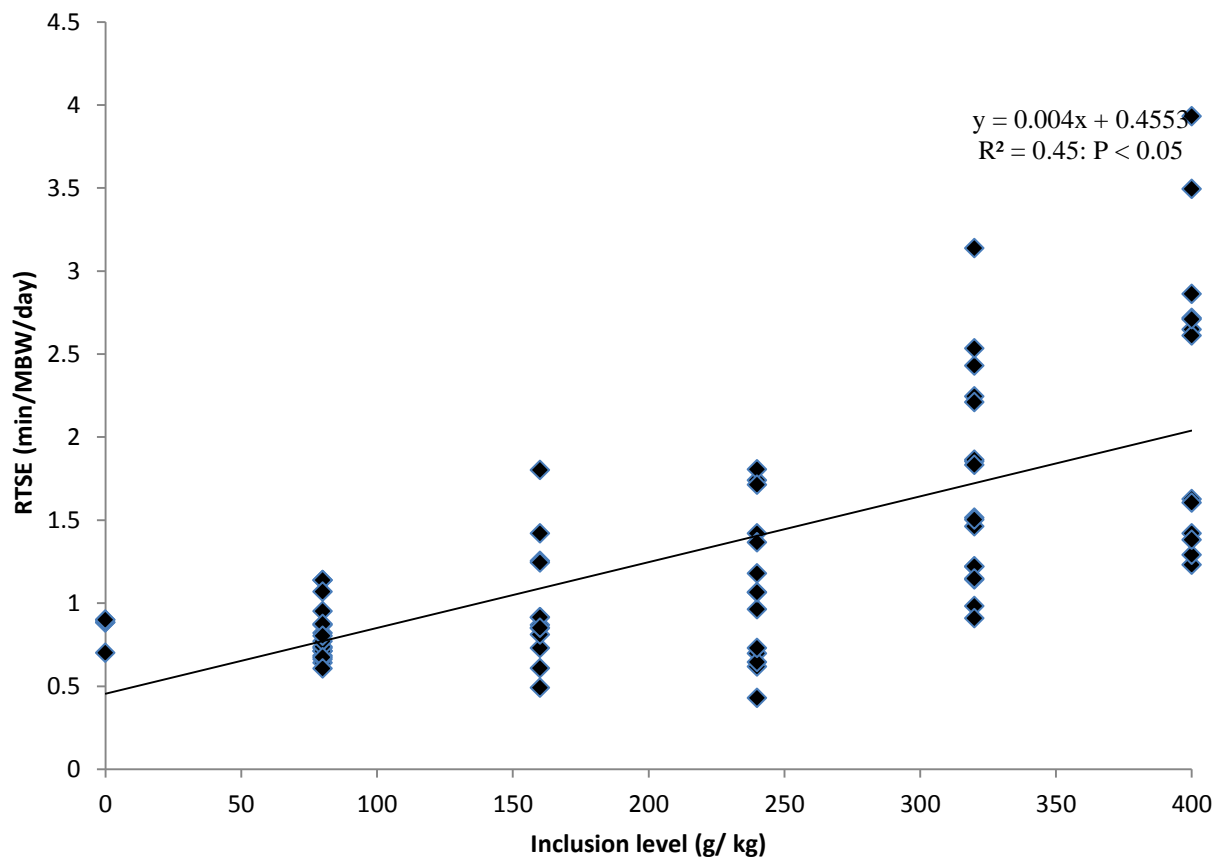
The observation that ADFI and ADG decreased with inclusion level of fibre was not expected. It was anticipated that increasing inclusion levels of fibre will result in pigs to eat more to compensate for the reduced nutrient density. Hence performance of the pigs is supposed to be the same. Decrease in weight gain of pigs fed fibrous diets was also observed in weaner pigs (Nyachoti *et al.*, 2006), gilts (Soren *et al.*, 2003) and finisher pigs. The reduction in ADG with inclusion level of fibre may be ascribed by pigs not getting enough nutrients for increasing or maintaining body weight.

**Table 7. 3: Effect of fibre source and inclusion level on concentration of blood metabolites**

Metabolite	Fibre source	Inclusion level (g/kg)						SE	P-value	
		0	80	160	240	320	400		Linear	Quadratic
TP (g/dl)										
	PU	0.21	0.22	0.23	0.27	0.23	0.25	0.0153	NS	*
	LH	0.21	0.21	0.23	0.23	0.25	0.21	0.0153	NS	*
	MC	0.21	0.22	0.25	0.23	0.26	0.22	0.0153	*	NS
	SH	0.21	0.27	0.25	0.22	0.29	0.27	0.0153	NS	NS
Albumin (g/dl)										
	PU	0.10	0.11	0.12	0.14	0.12	0.12	0.0065	NS	NS
	LH	0.10	0.11	0.12	0.11	0.13	0.12	0.0065	*	NS
	MC	0.10	0.12	0.13	0.12	0.12	0.11	0.0065	NS	NS
	SH	0.10	0.14	0.13	0.10	0.12	0.13	0.0065	NS	NS
CK (mg/dl)										
	PU	51.5	49.9	46.3	47.6	39.6	31.1	6.61	*	NS
	LH	51.5	59.7	48.9	44.8	31.4	24.4	6.61	**	NS
	MC	51.5	43.8	53.7	27.7	18.2	16.8	6.61	**	NS
	SH	51.5	36	76.6	54.9	63.7	42.1	6.61	NS	NS
Uric acid (mg/dl)										
	PU	0.15	0.19	0.18	0.23	0.18	0.19	0.023	NS	NS
	LH	0.15	0.18	0.34	0.15	0.20	0.19	0.023	NS	NS
	MC	0.15	0.16	0.18	0.12	0.20	0.19	0.023	NS	NS
	SH	0.15	0.19	0.18	0.13	0.26	0.23	0.023	*	NS
HbG (%)										
	PU	0.003	0.002	0.007	0.006	0.005	0.006	0.0026	NS	NS
	LH	0.003	0.006	0.007	0.009	0.016	0.009	0.0026	*	NS
	MC	0.003	0.013	0.008	0.002	0.01	0.011	0.0026	NS	NS
	SH	0.003	0.007	0.009	0.011	0.013	0.013	0.0026	**	NS

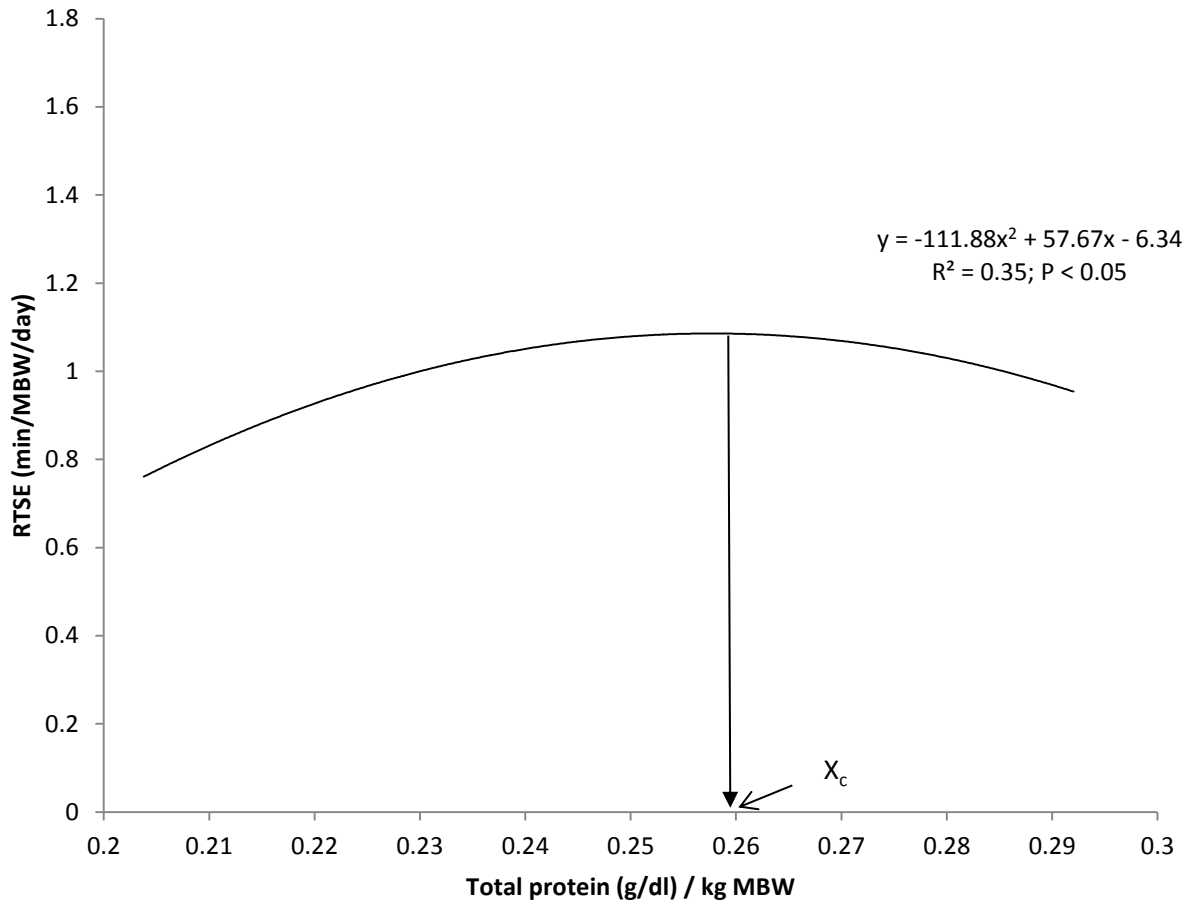
\*P < 0.05; \*\* P < 0.01; NS - not significant (P > 0.05); SE- standard error

MC - maize cob; SH - sunflower hulls; LH - Lucerne hay; PU - dried citrus pulp



RTSE - Relative time spent on eating to MBW of pig

**Figure 7. 1: Effect of inclusion level on relative time spent on eating to MBW of pig**



RTSE - Relative time spent on eating to MBW of pig; X<sub>c</sub> – threshold values

**Figure 7. 2: Relationship between time spent eating and concentration serum total proteins in finishing pigs**

Alternatively, decrease in weight gain can also be further explained by the reduction in feed intake with inclusion level of fibre as observed from pigs in the study. Increasing ADFI in the study by pigs might have been limited by gut capacity which induces early satiation (Van Wieren, 2000). The observation that ADFI decrease and at the same time HbG increase with inclusion level of fibre supports the glucostatic theory (Campfield *et al.*, 1995) where decline in glucose levels in the blood result in an increase in feed intake. Intake can only be reduced when glucose levels in the blood are high.

This has also been proved from pigs feeding various types of feeds (de Leeuw *et al.*, 2004; Forbes, 2009). Low ADFI at higher inclusion levels might also be attributed to the digestibility of feed. Whittemore (1993) has also reported voluntary feed intake to be limited by the digestibility of the diet offered to the pigs. High fibrous diets have a larger proportion of indigestible components resulting in more time spent chewing and less time eating.

There are several factors which influence nutrient digestibility of feedstuffs. The most important being feed composition, especially dietary fiber content (Le Goff *et al.*, 2002). Dietary fibre content varies depending on the fibre source used in the formulation of a diet; hence digestibility of nutrients such as proteins and soluble carbohydrates are likely to vary (Jorgensen *et al.*, 1996; Noblet and Le Goff, 2001). This can account for differences in HbG concentrations in blood of pigs subjected to diets with the different fibre sources (PU, LH, MC, and SH). High concentrations of HbG in blood of pigs fed high fibrous diets might be attributed to the nature of the diet presented. High-fibrous diets are eaten more slowly and are chewed more than low-fiber diets, which will lead to the delay in nutrient absorption resulting in high HbG concentration when blood was taken (Brouns *et al.*, 1997; Danielsen

and Vestergaard, 2001; Howarth *et al.*, 2001). Concentration of HbG will, however, reduce time spent eating when threshold value is reached regardless of nature of feed.

Creatine kinase can be used to measure the intensity of the body's response to stress (Kim *et al.*, 2004). Using growing pigs, CK levels increased with inclusion level of fibre. It was concluded that stress levels increased in growing pigs as inclusion level of fibre was increased. In the current study, an opposite trend was observed with finisher pigs. Decrease in CK levels with inclusion level might be ascribed to body weight of the pigs. Previous studies have shown animals with low body weights to produce low concentrations of serum CK compared to animals with high body weights. This might be the case in the current study as pigs receiving high fibrous diets had low body weights compared to pigs on low fibrous diets. Type of feed might also elicit different responses to stress. The observation that MC had low concentrations of CK whilst SH and high CK concentrations might be ascribed to the palatability as well as digestibility of the different fibre types, which can predispose the pigs to nutritional stress (Yeomans, 1998).

The observation that total proteins and albumin increased with inclusion level of fibre was not expected. Elevated levels of serum proteins might have emanated from protein catabolism (Zervas and Zijlstra, 2002). The observation that serum total proteins were the best predictor variable for time spent eating was expected. The broken-stick model indicated break point where time spent eating is reduced from concentration of  $0.26 \pm 0.047(\text{g/dl}) / \text{kg TP}$ . The break point value suggest that decisions when formulating fibrous diets should also take into considerations some of the nutritionally-related blood metabolites in pigs that influence time spent eating.



## 7.5 Conclusions

Graded levels of dried citrus pulp, Lucerne hay, maize cob and sunflower hulls influence average daily feed intake, average daily gain, time spent eating and nutritionally-related blood metabolites of the finishing pigs. Serum total protein was selected as the best predictor variable for predicting time spent eating. It produced a break point which determines satiety levels from relationship with time spent eating.

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## **Chapter 8: Prevalence of aggressive behaviours in grouped pigs fed fibrous diet**

*(Submitted to South African Journal of Animal Science)*

### **Abstract**

The objective of the study was to investigate the effect of feeding fibrous diets on growth performance occurrence of aggressive behaviours in growing pigs. Sixty healthy castrated pigs (Initial body weight:  $46.7 \pm 4.35$  kg) were used. A basal diet was diluted with maize cob to two levels (0 and 160g/kg DM). Behavioural activities were observed using video cameras for three weeks, 8 hours per day, starting at 0800h. Pigs subjected to control diet gained more weight compared to pigs receiving fibrous diet in week 1 (0.47 v/s 0.15 kg) and 2 (1.37 v/s 1.04) ( $P < 0.05$ ). Average daily gain was not affected by treatment diet in the third week. Pigs on high fibrous spent more time eating, lying down, standing, walking and fighting ( $P < 0.05$ ) compared to pigs on control diet. Week had an effect on time spent on different behavioural activities by pigs on different treatment diets ( $P < 0.05$ ). Time of day had an effect on time spent on different behavioural activities exhibited by all pigs on different treatment diet ( $P < 0.05$ ). Inactivity was greatest in 5th hour of the day (1200 -1300hrs) for all the pigs on different dietary treatments. Skin lesions appeared the most on neck and shoulder region followed by chest, stomach and hind leg region, and finally head region ( $P < 0.05$ ). Pigs on high fibre diet had more skin lesions in all body regions compared to pigs on control diet ( $P < 0.05$ ). It can be concluded that the high fibrous diet with maize cob did not reduce aggressive behaviours as expected. Aggressive behaviours emanated out of frustration when queuing on the feeder.

**Keywords:** Behaviour; Grouped growing pigs; Maize cob; Skin lesions

## 8.1 Introduction

Rearing of pigs in groups is more prominent under commercial farming practices. This offers benefits which include the use of labour-saving machinery and bedding systems, and practicing of all-in-all-out production which facilitates health management (Turner *et al.*, 2003). When pigs are in groups, they exhibit aggressive behaviours more often to establish ranks and also to compete for limited resources (feed and water). These behaviours cause injuries to pigs which lead to mortalities and also reduction in weight gain (Stookey and Gonyou, 1994). Fibrous feedstuffs are expected to reduce aggressive behaviours. The fibrous feedstuff increase the volume of feed eaten and/or time spent eating (Day *et al.*, 1996; Weber *et al.*, 2008), which is mainly attributed to the reduced density of nutrients. The pigs have to eat more of the less nutrient dense feed for them to meet requirements for growth. The fibres cause gut fill and also provide a substrate for chewing and rooting activities, thus, reducing the risk of the development and /or time spent on aggressive behaviours (Fraser, 1975; Lyons *et al.*, 1995; De Jong *et al.*, 1998; Van de Weerd *et al.*, 2005). Aggressive encounters frequently result in skin injuries, hence, apart from determining time spent on the behavioural activity, level of skin damage could also be used to reflect differences in observed levels of aggression of pigs fed fibrous diets.

Maize cob is the common fibre source and does not depress intake greatly at 160g/kg compared to other fibre sources like sunflower hulls and lucerne hay (Ndou *et al.*, 2013). The inclusion level of maize cob at 160 g/kg in a diet corresponds to 138 g/kg DM of ADF which constrained gut capacity (Ndou *et al.*, 2013). The objective of the study was, therefore, to determine the effect of feeding the fibrous diet on growth performance and aggressive behaviours of grouped growing pigs. It was hypothesized that the fibrous diet does not affect growth performance and it also reduces aggressive interactions.

## **8.2 Materials and methods**

### **8.2.1 Pigs and housing**

Care and use of the pigs were in compliance with internationally accepted standards for welfare and ethics in animals and was specifically approved by the University of KwaZulu-Natal Animal Ethics Research Committee (Reference number: 061/12/Animal). Sixty healthy castrated growing pigs (Initial body weight:  $23 \pm 3.1$  kg) were used in the study. The pigs were selected at random and put in six groups, 10 in each pen. Stocking density of pigs in each pen was  $0.627$  pig/  $m^2$ . The pens had concrete floors and brick walls. Feed was provided *ad libitum* from one tube feeder permanently fixed at the centre of each pen. Water was also provided *ad libitum* from low pressure nipple drinker located on the wall of the pen.

### **8.2.2 Diets and feeding**

A commercial feed (Grower, Meadow feeds Ltd, Pietermaritzburg, South Africa) with a low level of fibre was used as control diet (basal diet). Maize cobs were used as the fibre source. The cobs were ground to pass through a 3 mm screen and thereafter were included in a basal diet of pigs at a level of 160 g/kg. Three of the groups were subjected to a diet containing 160 g/kg maize cob meal (D2). The remaining three groups received a control diet (basal diet) (D1). Ingredients used in the formulation of basal diet were; yellow maize (500 g/kg), soya bean (158 g/kg), soybean oil cake (20.2 g/kg), wheat bran (163 g/kg), sunflower oil cake (85 g/kg), molasses syrup (25 g/kg) and additives (48.8 g/kg).

### **8.2.3 Analyses of physicochemical properties of diets**

Methods of analyses have been described in chapter 3. Chemical and physical properties of control and fibrous diet are described in Table 8.1.

**Table 8. 1: Chemical and physical properties of control and fibrous diet**

<b>Treatment diets</b>	<b>Control</b>	<b>Fibrous diet</b>
DM(g/kg)	90.14	90.91
<sup>1</sup> calc DE(g/kg DM)	10.75	10.56
CP(g/kg DM)	247.8	207.9
EE(g/kg DM)	3.73	5.45
ASH(g/kg DM)	5.52	5.04
NDF(g/kg DM)	430.93	470.3
ADF(g/kg DM)	68.39	185.59
BD(gDM/ml)	1.83	1.65
WHC(gwater/g DM)	4.6	5.71

DM – Dry matter; CP – Crude protein; EE – Ether extract; calc DE – calculated digestible energy; NDF – neutral detergent fibre; ADF – acid detergent fibre; BD – bulk density; WHC – water holding capacity.

<sup>1</sup> calc DE = 949 + (0.789 × GE) - (43 × % Ash) - (41 × % NDF) (Noblet and Perez,1993).

#### **8.2.4 Observations**

Pigs were allowed to adapt to their new environment, the feed offered and to using the feeders for two weeks. The period would also allow them to establish ranks so that the effect of diet on aggressive behaviour could be clearly defined. Each pig was weighed at the beginning of the experiment and, thereafter, weekly.

Injuries emanating from aggression behaviours were assessed on individual pigs in each group. This was done at the beginning of the experiment and, thereafter, weekly on fresh lesions when the pigs were being weighed. To facilitate counting, the body was divided into head, neck and shoulder regions, and the remaining parts of the body. Recordings were done only on fresh lesions, which were judged subjectively by colour and the estimated age of scabbing (Turner *et al.*, 2006). The scoring method used followed that of Stukenborg *et al.* (2012). To keep the scores consistent throughout the experiment, one person assessed the injury scores. Behavioural activities of pigs in each pen were recorded from video cameras mounted on the ceiling for eight hours, starting at 0800h and ending at 1600h. Feed was introduced before recording of behavioural activities started. All pigs were numbered on the back with paint, which aided in the accurate identification during observation. Behavioural activities observed in the study are shown in Table 8.2. All occurrence sampling method, which involve continuous recording all behavioural activities was used. Time spent on different behavioural activities was observed for a period of three weeks, once a week. Time spent on the behavioural activities was scored by the same person throughout the whole experiment which lasted for three weeks. For determining number of fights made by pigs in each group, a fight was recorded when it took longer than 2s and a new fight started if there was an intervening period of more than 8s (Puppe, 1998).



**Table 8. 2: Description of behavioural activities**

<b>Behavioural activity</b>	<b>Description</b>
<b>Feeding behaviour</b>	
Eating	Consumption of feed material from the feed
Drinking	Manipulating the water bowl
<b>Postures</b>	
Standing	Standing without eating or manipulating the water bowl
Lying down	Sprawl down to rest
Walking	Moving from one position to another
<b>Abnormal behaviours</b>	
Fighting	Repeated biting and pushing
Other	Object-biting, nose-rubbing, mounting

### **8.2.5 Statistical Analyses**

For the analysis of the behavioural data, PROC CAPABILITY procedures of SAS (SAS 2008) were used to check for normality of data for time spent on different behavioural activities exhibited by pigs. The variables showed deviations from normality, hence, were logarithmically transformed. Data for behavioural activities, skin lesion scores and number of fights were analysed using the mixed model procedures for repeated measures (SAS, 2003). The DUNNETT t-test was used to compare means for treatment with the control at  $P < 0.05$ . For the comparison of behaviour of grouped pigs subjected to the treatment diets, the pen was considered as the experimental unit and the model included the effects of treatment diet, week, time of day and possible interactions as independent variable.

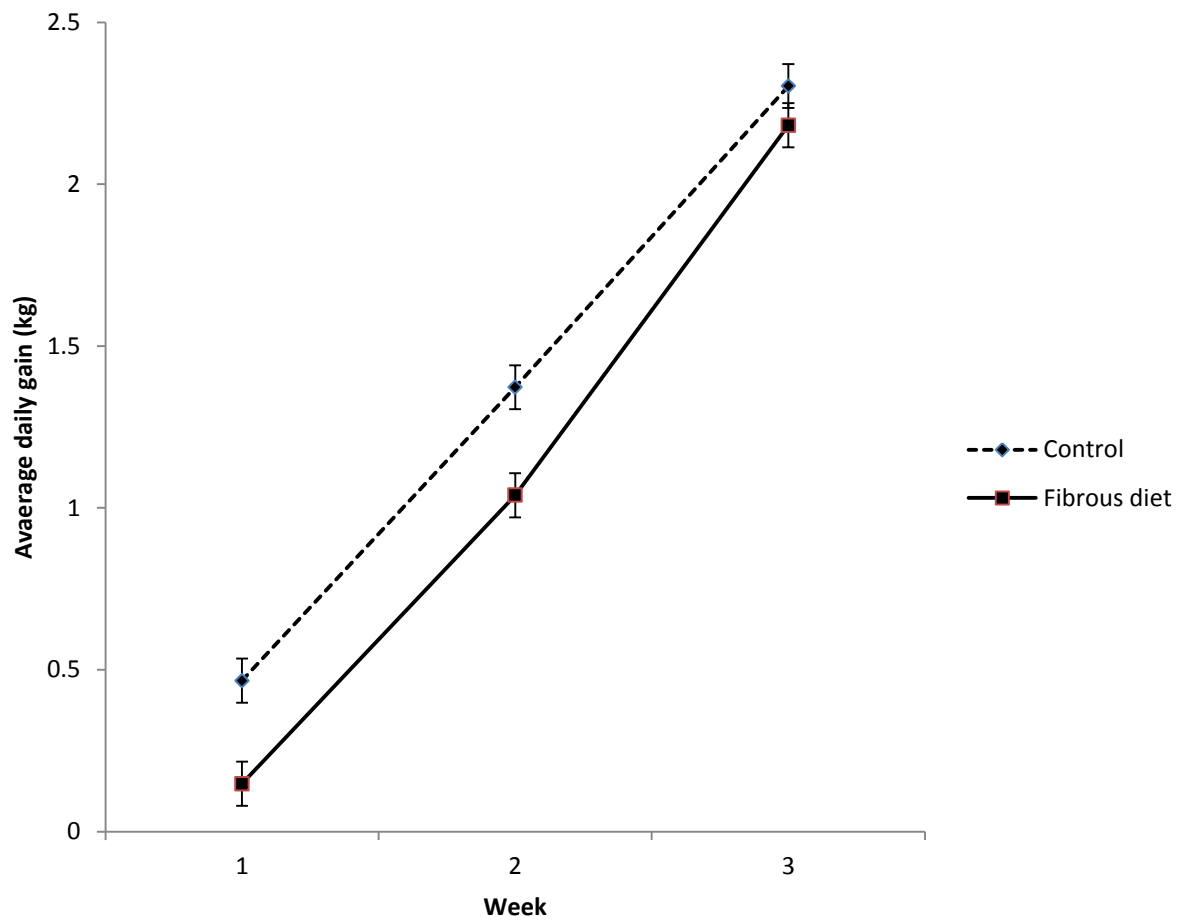
## **8.3 Results**

### **8.3.1 Effect of feeding fibrous diets on average daily gain of grouped pigs**

Average daily gain for all the pigs subjected to the treatment diets increased during the three week period as illustrated in Figure 8.1. Pigs subjected to control diet gained more weight compared to pigs receiving fibrous diet in week 1 and 2 ( $P < 0.05$ ). In week 3, there were no differences in weight gain for all the pigs subjected to the different dietary treatments ( $P < 0.05$ ).

### **8.3.2 Effect of inclusion level of maize cob on time spent on different behavioural activities by grouped pigs**

Effect of diet on time spent on different behavioural activities was significant ( $P < 0.05$ ; Table 8.3). Pigs subjected to fibrous diet spent more time eating, lying down, standing, walking and fighting. However, no differences were observed for time spent on other behavioural activities.



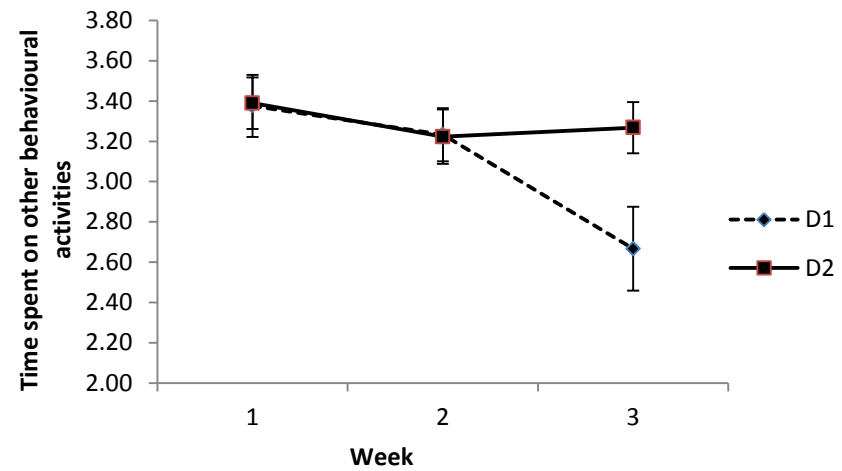
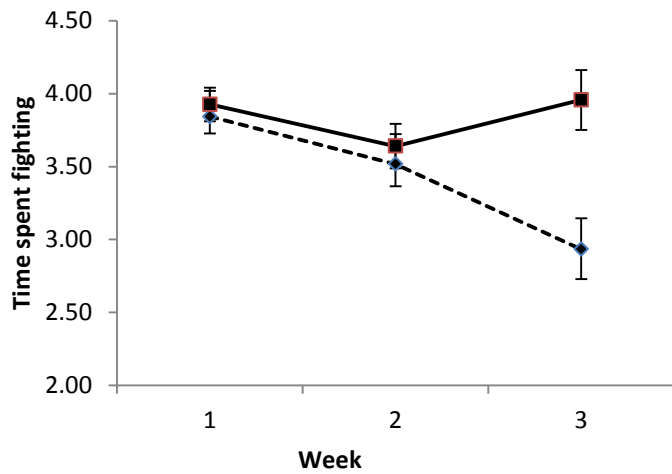
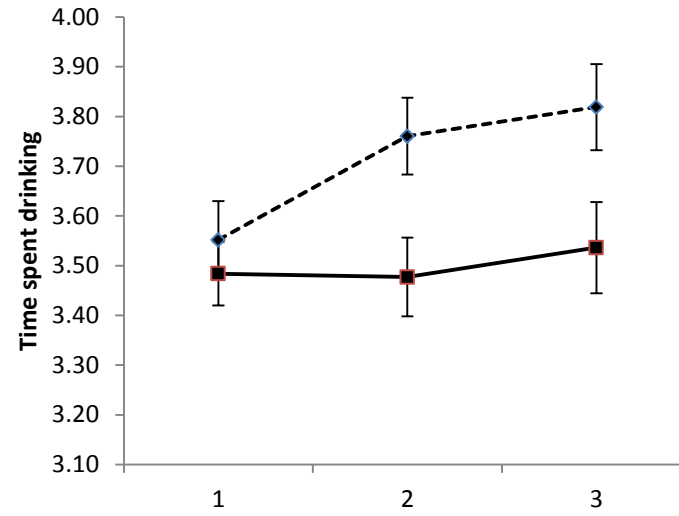
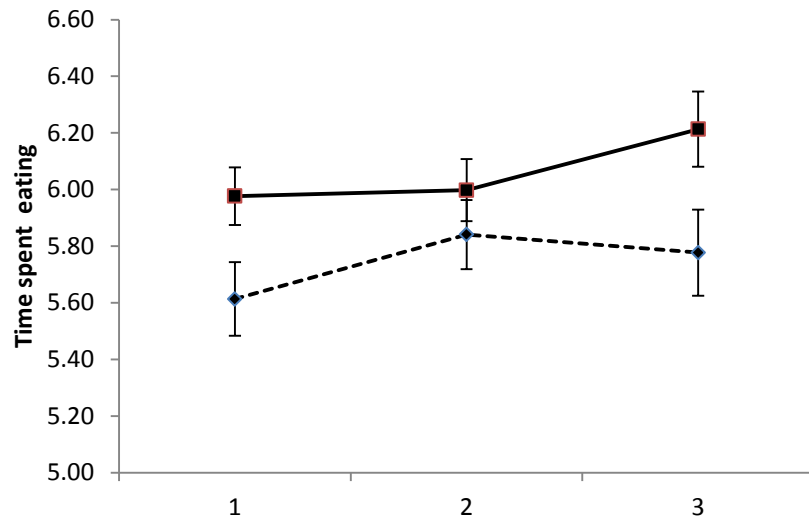
**Figure 8. 1: Average daily gain for pigs on different treatment diets**

**Table 8. 3: Effect of feeding different diets on time spent (*sec/hr*) (Mean  $\pm$  standard error) on different behavioural activities**

	Treatment diets	
	Control diet	Fibrous diet
<b>Feeding behaviour</b>		
Eating	5.74 $\pm$ 0.0785 <sup>a</sup>	6.06 $\pm$ 0.0666 <sup>b</sup>
Drinking	3.71 $\pm$ 0.0468 <sup>b</sup>	3.50 $\pm$ 0.0456 <sup>a</sup>
<b>Postures</b>		
Lying Down	7.05 $\pm$ 0.0396 <sup>a</sup>	6.65 $\pm$ 0.0392 <sup>b</sup>
Standing	4.35 $\pm$ 0.0698 <sup>a</sup>	5.25 $\pm$ 0.0592 <sup>b</sup>
Walking	3.82 $\pm$ 0.0540 <sup>a</sup>	4.18 $\pm$ 0.0497 <sup>b</sup>
<b>Abnormal behaviours</b>		
Fighting	3.43 $\pm$ 0.123 <sup>a</sup>	3.84 $\pm$ 0.104 <sup>b</sup>
Other	3.09 $\pm$ 0.0966 <sup>a</sup>	3.29 $\pm$ 0.0797 <sup>a</sup>

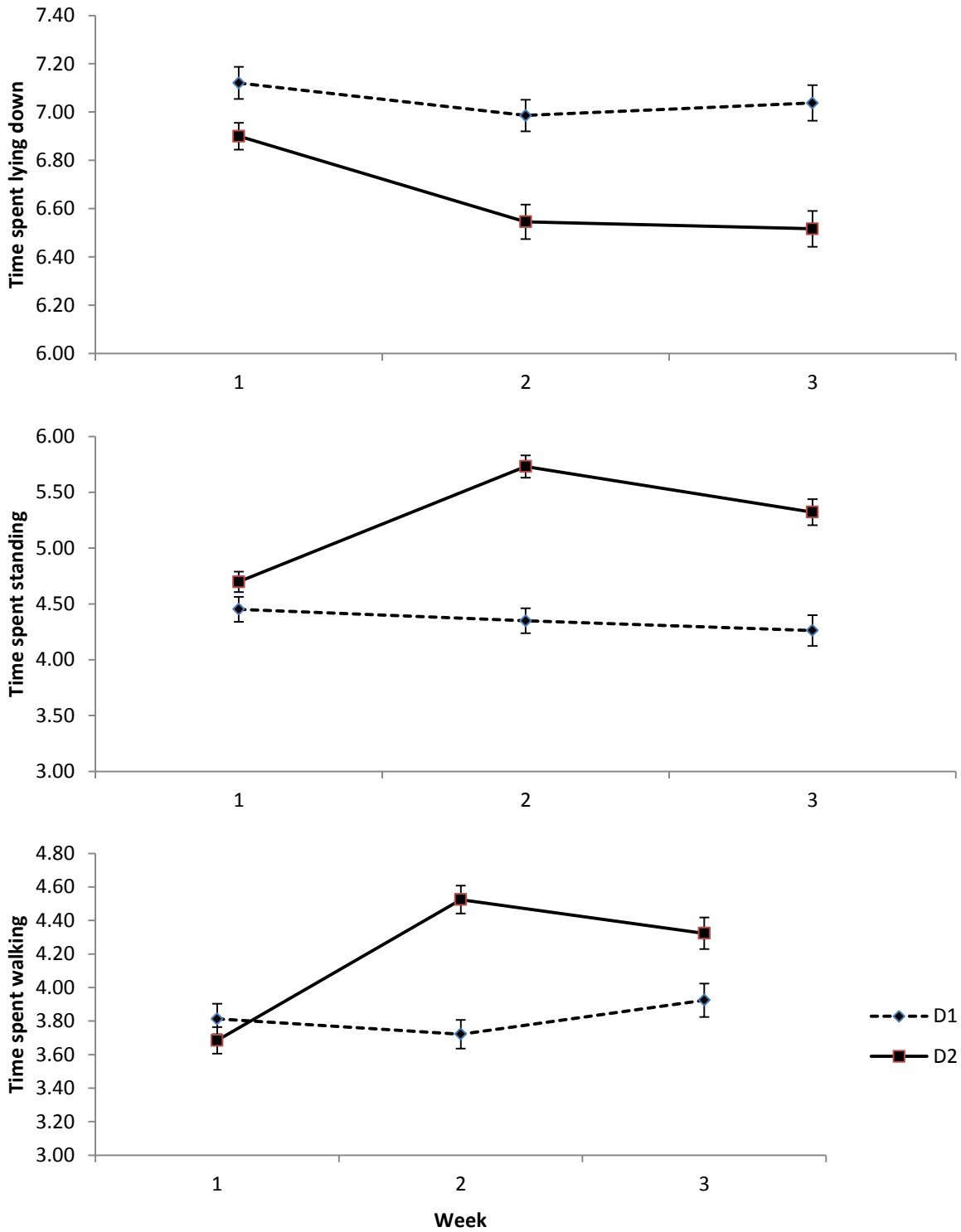
### **8.3.3 Interaction effect of week and inclusion level of maize cob on time spent on different behavioural activities**

Interaction of week and treatment on time spent on different behavioural activities was also significant ( $P < 0.05$ ). Generally, time spent eating and drinking increased as the pigs grew (as body weight increased) on the different dietary treatments (Figure 8.2). Pigs receiving fibrous diet spent more time eating compared to pigs subjected to control diet. Conversely, pigs receiving control diet spent more time drinking compared to pigs subjected to fibrous diet. A similar trend was observed for interaction of week and time spent lying down, fighting and other behavioural activities for all pigs (Figures 8.2 and 8.3). Time spent eating increased in the first two weeks for all the pigs. However, in the third week time spent eating by pigs receiving fibrous diet increased compared to pigs on a control diet. Time spent drinking increased for the pigs receiving different dietary treatments as the weeks progressed. Time spent standing and walking in week 1 and week 2 increased then later decreased in the third week for pigs subjected to control diet.



D1- control diet; D2-diet with maize cob

0 **Figure 8. 2: Effect of week and treatment on time spent ( $\log(sec)$ ) on feeding and abnormal behaviours of pigs**



D1- control diet; D2-diet with maize cob

**Figure 8. 3: Effect of week and treatment on time spent (*log (sec)*) on different postures exhibited by pigs**

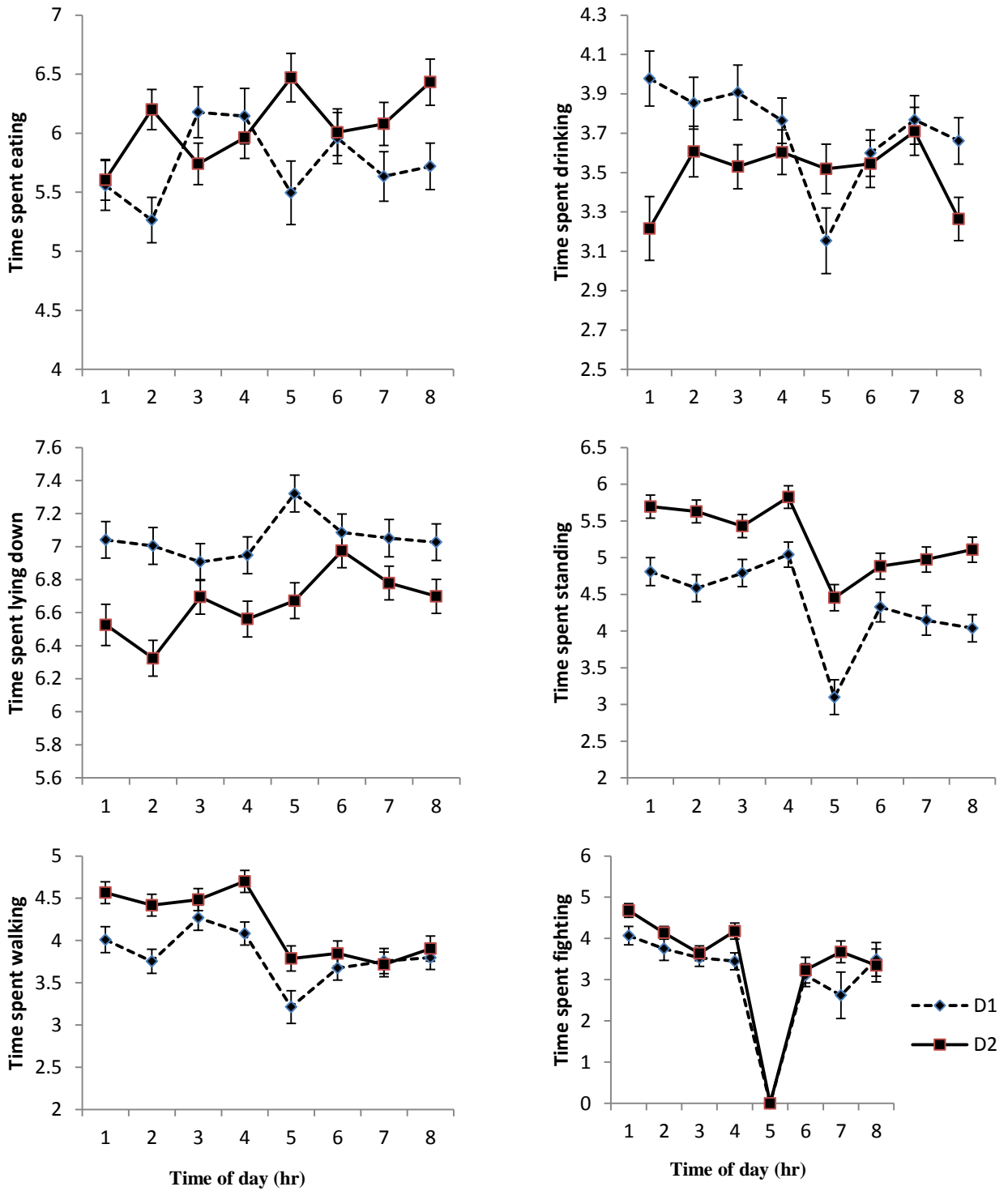
### **8.3.4 Effect of time of day and inclusion level of maize cob on time spent on different behavioural activities**

Figure 8.4 shows the effect of time of day and inclusion level of maize cob on time spent on different behavioural activities. There was a similar pattern for time spent lying down, standing, walking and fighting regardless of diet offered to the pigs. Peaks for time spent lying down for pigs receiving control and fibrous diet were in the 5<sup>th</sup> (1200 -1300h) and 6<sup>th</sup> (1300 -1400h) hour of the day, respectively. A sharp decrease in time spent standing, walking and fighting was observed in the 5<sup>th</sup> (1200 -1300h) hour of the day for all the pigs. In general, aggressive behaviours decreased as the day progressed.

### **8.3.5 Effect of inclusion level of maize cob on lesion scores and number of fights in grouped pigs**

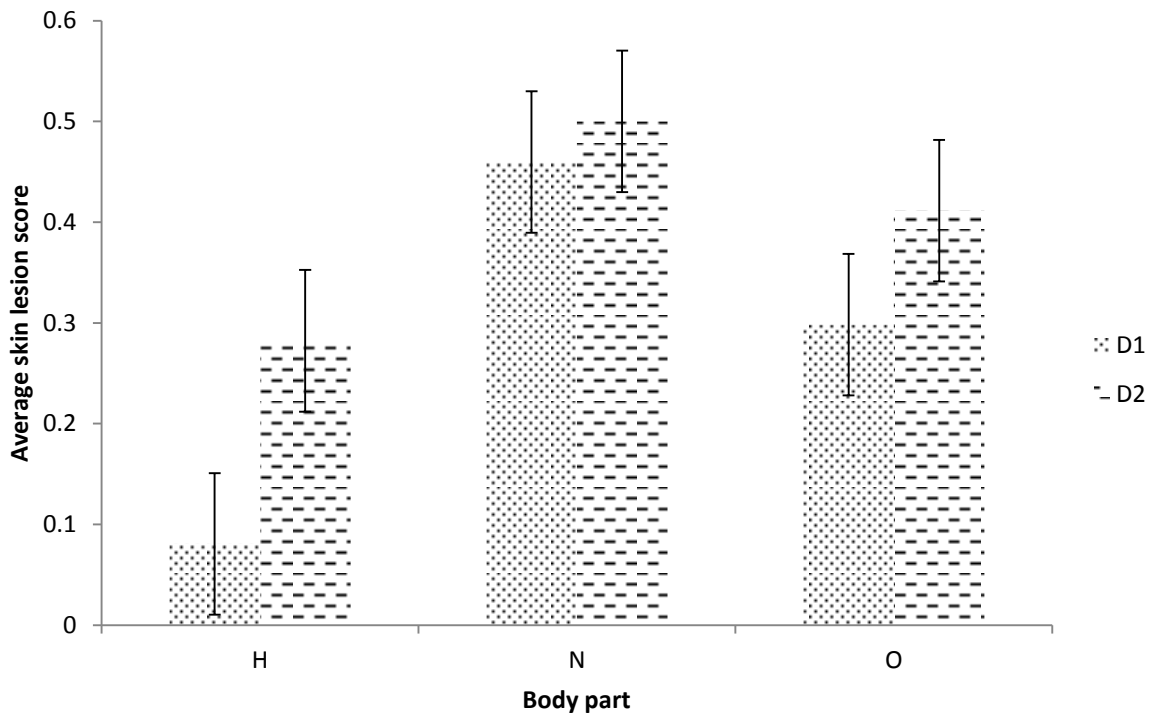
Greatest number of lesions occurred on neck and shoulder region followed by chest, stomach and hind leg region, and finally head region ( $P < 0.05$ ; Figure 8.5). Pigs subjected to fibrous diet had more lesions compared to pigs receiving control diet ( $P < 0.05$ ). Week had no effect on the number of lesions on pigs receiving the two diets. Figure 8.6 shows the mean number fights for pigs subjected to the two diets. Higher incidence of fighting was observed in pigs subjected to fibrous compared to control diet ( $P < 0.05$ ).





D1- control diet; D2-diet with maize cob

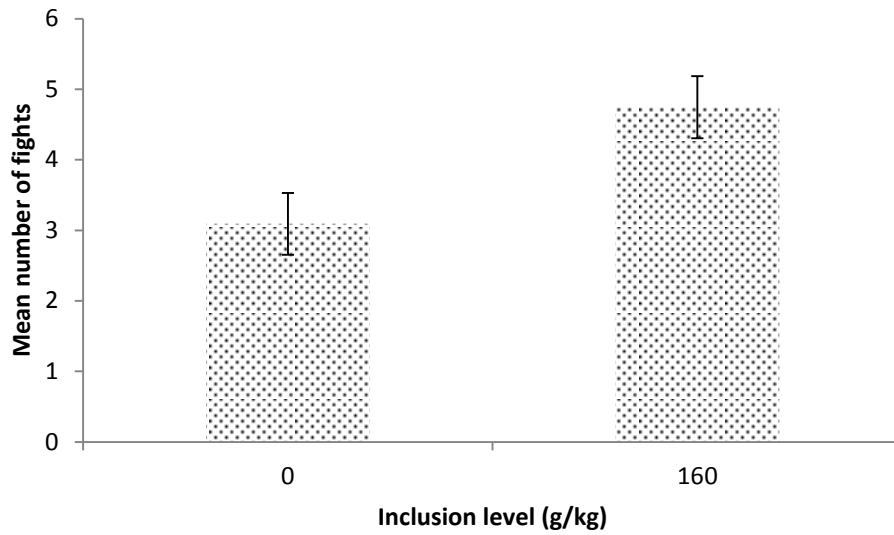
**Figure 8. 4: Effect of time of day and treatment on time spent (log (sec)) on different behavioural activities**



D1- control diet; D2-diet with maize cob

H- head region; N-neck and shoulder region; O-rest of the body

**Figure 8. 5: Mean skin lesion scores for pigs on different treatment diets**



D1- control diet; D2-diet with maize cob

**Figure 8. 6: Mean number of fights for pigs on different diets**

## 8.4 Discussion

In the study, maize cob was used because it is the most abundant and readily available fibre source, and has a relatively low water holding capacity which does not depress feed intake greatly (Ndou *et al.*, 2013). Inclusion at 160 g/kg DM maize cob in a diet increased the bulkiness of the feed. It was, therefore, assumed that any effects of the fibrous diet on feeding behaviour were a result of the physical properties of diet. The observation that weight gain was greater for pigs receiving diet with maize cob compared to control diet in the week 3 might be an indication that the pigs are beginning to tolerate the fibrous diet provided. This is in agreement with study conducted by Konstanze *et al.* (2012).

The study showed severity of skin damage on pigs to reflect levels of aggression in pigs subjected to the treatment diets. This is evident from the observations made were pigs on fibrous diet spent more time fighting compared to pigs on control diet. Simultaneously, more lesions were found on pigs receiving fibrous diet compared to pigs on control diet.

The observation that lesion scores were greater in diets with fibrous diet compared to control diet was not expected. Inclusion of fibrous diet was expected to reduce incidence of fighting since most of the pigs will be devoting most their time eating. The high feeding motivation frequently resulted in aggression, particularly around feeding stations. The observation that more lesions occur on the neck and shoulder region was expected. This is mainly because when pigs are fighting they position themselves so that their heads align on both of their shoulders. Slamming of their heads into each other's shoulders will result in more cuts or lesions on the neck and shoulder regions. Fighting will persist until one pig admits defeat and runs away. Lesions on the head during the study period occurred when pigs were displacing each other from the feeder and nipple drinkers.

The observation that more time was spent eating by pigs on a fibrous diet compared to control diet is in agreement with a previous studies (Whittaker *et al.* 1999; Whittemore *et al.*, 2002; Rijnen *et al.*, 2003). More time spent eating is a result of unsatisfied feeding motivation, as a consequence of dilution effect of maize cob to the basal diet. The pigs will be eating more to get nutrients to satisfy their nutritional requirements for growth.

As observed in the current study, more time spent eating by a pig receiving a fibrous diet at the feeder resulted in more time spent queuing by another pig at the feeder. This might account for more time spent standing by pig receiving fibrous diets as observed in the current study. More time spent walking emanated when pigs were walking around sniffing the floor. Such findings are in agreement with previous studies which refer the action as substrate-directed behaviour (de Leeuw *et al.*, 2005). Substrate-directed behaviour occurs when there is an increase in feeding motivation and limited access to the feed. Increase in time spent lying down came entirely from a decrease in time spent eating.

Previous studies have shown time spent eating to be positively correlated to time spent drinking (Barber *et al.*, 1989; Dybkjær *et al.*, 2006). Pigs receiving fibrous diets are expected to spend more time drinking compared to pigs subjected to low fibrous diets. This was not the case in the study as pigs on low fibrous diet spent more time drinking. Possible explanation to this might lie in the method used in the study for assessing drinking behaviour (Meiszberg *et al.*, 2009). Drinking behaviour in the study was defined as mouth in contact with the nipple drinker and does not include the amount of water ingested. More time spent drinking by pig on control diet might have resulted from boredom and, therefore, will be playing with the nipple drinker.

With regard to diurnal behavioural patterns, decrease in the activities; walking, standing, drinking and fighting in the 5<sup>th</sup> hour (1200 -1300h) might be attributed to the pigs spending more time lying down mainly because it was the hottest hour of the day. More time spent lying down could be explained as a sign of enhanced satiety for all the pigs subjected to the different treatment diets (Robert *et al.*, 1993). Pigs showed an almost similar diurnal pattern for the behavioural activities regardless of treatment diet. There may be a physiological mechanism induced by photoperiod influencing behavioural patterns exhibited by all pigs subjected to the different treatment diets (Whittemore *et al.*, 2002). In the morning when fresh feed is put in feeders, there will be a lot of activity among the pigs as they will be anticipating a share of the feed. They, therefore, queue at the feeder. Aggression emanates out of frustration as they will be waiting for a longer period for another pig to finish eating (Whittaker *et al.*, 1999).

## **8.5 Conclusion**

It can be concluded that weight gain of the experimental pigs was not affected by the treatment diets. Furthermore, high fibrous diets did not reduce aggressive behaviours as expected. Aggressive behaviours emanated out of frustration when queuing at the feeder for feed.

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## **Chapter 9: General Discussion, Conclusions and Recommendations**

### **9.1 General discussion**

The use of fibrous diets is receiving increasing interests among feed compounders to produce a cheaper feed and at the same time to reduce competition for grain between humans and livestock species. These fibres have been included in diets of pigs at different inclusion levels. However, different responses with regards to behaviour and also optimum inclusion levels for feeding the different fibre sources have been drawn. A definite answer is not yet in hand on such different responses from feeding different fibre sources. Since the different fibre sources have different physicochemical properties, it is possible that a single conclusion can be drawn from these physicochemical properties. The hypotheses tested for the study were that physicochemical properties of fibrous diets do not influence time spent on different behavioural activities, gut architecture and performance, and blood metabolites are not indicators of nutritional status and cannot be used to predict time spent eating in growing and finishing pigs.

The hypothesis tested for Chapter 3 was that physicochemical properties of different fibre sources are not predictors of time spent on different behavioural activities by growing pigs. Different fibre sources were used to give a wide range of physicochemical properties for predicting time spent on different behavioural activities. Physicochemical properties of fibre sources influenced time spent on different behavioural activities. An increase in indigestible components in diets (ADF and NDF) resulted in pigs to spend more time eating. It was expected in the study that feed bulk cause distension of the stomach walls, hence the reduction in time spent eating by growing pigs. By this, optimum inclusion level of fibre from physicochemical properties is estimated using the broken stick method. This was not the case as time spent eating showed a linear increase as indigestible components of diets

increased. Hence, optimum inclusion level could not be estimated. It was concluded that the linear response was a result of the chewing activity which made it impossible to estimate gut capacity from eating behaviour (time spent eating). More time spent eating resulted from more time spent chewing the fibrous diets. Decrease in time spent lying down came entirely from an increase in time spent eating.

Feed characteristics and nutritionally-related blood metabolites usually work in synergy to control time pigs spent eating. Hence, in Chapter 4 hypothesis tested was that blood metabolites do not influence time spent eating and drinking, and nutritional status of growing pigs. The blood metabolites provided positive relationships between time spent eating and drinking activities. Although the experimental pigs showed increased time spent eating, they did not get the required nutrients from the feed, resulting in the body to waste tissue to increase protein and uric acid concentrations in blood. Threshold values were obtained from the relationships. Hence, the metabolites can be used to predict when the pig will start or stop eating. The metabolites also provided valuable information on nutritional status of pigs. Elevated concentrations of protein and uric acid in blood at higher fibre inclusion level suggest that protein catabolism was eminent. This resulted in loss of weight at higher fibre inclusion level. Feed compounders will get an idea if pigs are getting feed with the right nutritional requirements.

The hypothesis tested in Chapter 5 was that physicochemical properties of fibrous diet do not predict intestinal villi height and apparent villi surface area of growing pigs. Bulk density and neutral detergent fibre influenced VH and AVSA. Villi height and AVSA increased as NDF increased and also when BD decreased. The bulk properties of fibrous diets (NDF and BD) might have triggered a natural increase in the absorption and utilisation of nutrients by

increasing VH and AVSA as nutrients became more limiting when NDF increased and also as BD decreased. The findings show that the pigs were adapting to the fibrous diet by increasing villi height and apparent villi surface area.

In Chapter 6, it was hypothesized that the relationship between physicochemical properties of fibrous diet and time spent on different behavioural activities by growing pigs and finishing pigs was similar. Both growing and finishing pigs showed linear increase in time spent eating as bulkiness of feed was increased. Continual increase in time spent eating in finishing pigs could be attributed to bite sizes instead of time spent chewing. This is mainly because adult pigs have much more developed masseter muscles and are expected to chew better than growing pigs. Duration of the visit on the feeder was influenced by water holding capacity. This supported the much adopted perception that water holding capacity of a feed causes swelling of feed material which results in distension of the stomach walls. Satiety is induced, thus, reducing duration of visit on the feeder.

There is also a need to know how the nutritionally-related blood metabolites might also influence time spent on different behavioural activities by finishing pigs. Chapter 4 highlighted the metabolites that influence time spent eating and drinking. In Chapter 7, it was hypothesized that the metabolites do not predict time spent eating in finishing pigs. The serum proteins provided a relationship with time spent eating. Elevated levels of serum total proteins are also owed to by mass wasting of muscle tissues. The much proposed hypothesis that glucose concentrations determine voluntary intake is supported in the chapter using finishing pigs. Transient decline in glucose levels resulted in increases in time spent eating. The metabolites can also be used to predict when a pig will start or stop eating.

Under commercial farming practices pigs are normally kept in groups. In groups when fed these fibrous diets, they are likely to exhibit behavioural activities that might also compromise their performance. The hypothesis tested in Chapter 8 was that feed bulkiness does not influence growth performance and aggressive behaviours of the grouped pigs fed graded level of maize cob. We were expecting aggressive behaviours to be reduced in pigs on fibrous diet. This was not the case in the study as more aggressive behaviours were observed in the pigs fed fibrous diets. High fibrous diets increased motivation to eat in pigs. This increased competition to feed in pens where feeders were limited resulting in pigs to fight. Growth performance of pigs receiving low and high fibrous diets was, however, not compromised.

## **9.2 Conclusions**

Different physicochemical properties of fibrous diets influence time spent on different behavioural activities. In weaner pigs, time spent eating was influenced by ADF and bulk density feed. In finishing pigs, ADF and NDF influenced time spent eating. The physicochemical properties of fibrous feeds provide relationships with time spent eating. Apart from determining time spent on different behavioural activities, the physicochemical properties of fibrous feeds also influence villi height and apparent villi surface area in small intestines of the pigs. Neutral detergent fibre and bulk density influenced villi height and apparent villi surface area. Nutritionally-related blood metabolites provide relationships with time spent eating. They can be used to determine satiety levels in both weaner and finishing pigs. Feeding fibrous diets did not reduce aggressive behaviours in pigs.

### **9.3 Recommendations and further research**

Physicochemical measurements of bulk provide relationships describing changes in time spent eating and mucosal architecture. When formulating diets, feed compounders should consider the physicochemical properties of ingredients (NDF, ADF, WHC and BD) to ensure that pigs receive the much needed nutrients to achieve their optimum growth performance and gut health. Performance of pigs in the study was thwarted at higher inclusion levels of fibres in diets of pigs. There is, therefore, a need for further research to improve performance of pigs fed diets with higher fibre inclusion levels.

Aspects that require further research include:

1. Determining effect of physicochemical properties of fibrous diet on feeding behaviour and growth performance of different pig genotypes. This will help in identifying pig genotypes that are better able to utilise these fibrous materials.
2. Assessing blood metabolites which indicate the actual level of stress in pig genotypes subjected to fibrous diets.
3. Determining the effects of pelleting fibrous diets with different physicochemical properties on growth performance and feeding behaviour of pigs
4. Determining the effects of exogenous feed additives on growth performance and feeding behaviour of pigs fed fibrous diets varying in physicochemical properties.
5. Determining the effects of feeding fibrous diets with different physicochemical on intestinal enzyme function

**Appendix 1: Published article in Livestock Science**



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## Influence of physicochemical properties of fibrous diets on behavioural reactions of individually housed pigs



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### ABSTRACT

The objective of the study was to predict time spent on different behavioural activities of individually housed growing pigs from physicochemical properties of feeds. Maize cob, maize stover, sunflower hulls, veld grass, sawdust and lucerne were used to provide a wide range of physicochemical properties. The fibre sources were included at 0, 80, 160, 240, 320, 400 g/kg inclusion levels in pig diets. Time spent eating, drinking, lying down, sitting/standing and other activities was observed using video cameras. Pigs spent most of their time lying down (71.4%) followed by time spent eating (23%), drinking (3.2%) and sitting/standing (2.4%). Digestible energy (DE), bulk density (BD), acid detergent fibre (ADF) and water holding capacity (WHC) were the most important variables for predicting time spent on different behavioural activities ( $P < 0.001$ ). Bulk density and ADF produced linear responses with time spent eating and drinking ( $P < 0.001$ ). There was a quadratic response between time spent lying down and ADF content of feed ( $P < 0.001$ ). Water holding capacity was the most important physicochemical property of feeds for predicting number of visits made by the pig to the feeder. Total time spent on each visit by a pig per day was best predicted by ADF. In conclusion, physicochemical properties of diets alter behaviour of penned growing pigs.

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### 1. Introduction

The search for cheap alternative feed ingredients for pigs (*Sus scrofa*) is receiving attention mainly because of the increased global demand for grains for both human and livestock consumption (McCalla, 2009). Alternative feed ingredients that can be used to feed pigs are crop by-products: maize cob, maize stover, sunflower husks and, also grasses and sawdust. These fibre sources are readily available, cheap and can be consumed by pigs (Ferguson et al., 2003; Kyriazakis and Emmans, 1995; Ndindana et al., 2002; Owen and Ridgman, 1967, 1968). Fibres have been reported as diluents in previous studies (Len et al., 2007;

Whittemore et al., 2002). Their use, depending on the inclusion level in a diet, improves welfare of pigs by reducing stereotypic behaviours, chronic colitis, dysentery and constipation (Day et al., 1996; Filer et al., 1986; Metzler and Mosenthin, 2008; Thomson, 2009).

As fibre content of diets increase, the dilution of the resultant diets alters the time that pigs spend on various behavioural activities. For example, pigs fed on fibrous diets have an increased motivation to eat (D'Eath et al., 2009). Ramonet et al. (1999) reported that increasing fibre level in sow diets increases time spent eating. High-fibre diets also increased time spent eating in non-pregnant nulliparous gilts (Robert et al., 1997). Behavioural reactions of growing pigs subjected to the fibrous diets, however, remain unclear. Apart from assessing feed intake and growth performance, behavioural responses of the pigs need to be considered in the determination of appropriate fibre inclusion levels.

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Physicochemical characteristics of fibrous feeds might play a key role in influencing behaviour of pigs. These include water holding capacity (WHC), bulk density (BD), acid detergent fibre (ADF), neutral detergent fibre (NDF) and crude fibre (CF). The physicochemical characteristics vary across fibre sources (Ndou, 2012). Fibres with a high WHC, for example, will absorb more water, swell and occupy more space in the stomach. Consecutive stimulation of stretch receptors due to distension of the gut will lead to satiety (De Leeuw, 2004; Lepionka et al., 1997). Satiety is the state of being satisfactorily full and unable to take on more feed. This affects feeding behaviour and, consequently, performance of growing pigs.

Although physicochemical properties have been demonstrated to predict feed intake (Kyriazakis and Emmans, 1995; Ndou et al., 2013), their effect on time spent on different behavioural activities in growing pigs is still unclear. For example, Whittemore et al. (2002) conducted a study with few fibrous sources with a narrow range of physicochemical properties. Use of more fibrous feedstuff at varying inclusion levels provides a wider variation of physicochemical properties to generate more accurate predictions of behavioural activities exhibited by growing pigs. For close monitoring of pigs and to avoid the effects of association which can affect normal behavioural activities when in groups, it is necessary to house pigs individually. Time spent on different behavioural activities might be a first step in providing an insight on how specific physicochemical characteristics of different fibre sources affect satiety levels in pigs. The objective of the study was, therefore, to predict the influence of physicochemical properties of feeds on time spent on different behavioural activities in growing pigs; the hypothesis being that there is no relationship between the physicochemical properties and time spent on different behavioural activities.

## 2. Materials and methods

### 2.1. Pigs and housing

A total of 124 clinically-healthy, castrated, male growing pigs were used. The care and use of the animals was performed according to the Certificate of Authorization to Experiment on Living Animals provided by the University of KwaZulu-Natal (Reference number: 061/12/Animal). Initial body weight for the pigs used in the experiment was  $22.4 \pm 3.12$  kg. Each pig was used as an experimental unit. They were put in individual pens measuring  $1.5 \text{ m} \times 1 \text{ m}$  and had a slatted floor. The space in the pens allowed the pigs to turn around and have limited exercise. The pens contained a plastic tube feeder (Big Dutchman Lean Machine®) and a low-pressure nipple drinker providing clean water ad-libitum. The individual pens were not in total isolation because a pig had visual contact with the pig in adjacent pens and could hear and communicate with each other. Pigs were put in individual pens for five weeks including the adaptation period of one week. The ambient temperature and relative humidity were recorded automatically throughout the trial using HOBO data loggers (Onset Computer Corporation, Pocasset, MA, USA). The average temperature and humidity were  $21.15 \pm 2.74$  °C and  $42.64 \pm 1.56\%$ , respectively.

### 2.2. Diets

A premium commercial feed (Express Weaner, Meadow feeds Ltd.) with a low level of crude fibre (CF) was used as the basal feed. Six fibre sources, namely maize cob, maize stover, sunflower hulls, grass hay, lucerne and saw dust were selected to dilute the basal diet. Maize cobs used in the experiment were remains left after removal of kernels from the cob. Maize stover, on the other hand, consisted of the leaves and stalks of maize (*Zea mays*) plants left after removal of the cob. Sunflower hulls were the by-products from sunflower oil seeds after removal of kernels whilst sawdust or wood dust was the by-product of cutting, grinding, drilling; it is composed of fine particles of wood. Grass hay and lucerne were plants that have been cut, dried, and stored for use as animal fodder. All the fibre sources were used in the experiment to increase the range of physicochemical properties. Maize cob, maize stover, sunflower hulls, grass hay and lucerne hay were ground to pass through a 3 mm screen. The fibre sources were then included at different inclusion levels of 0, 80, 160, 240, 320 and 400 g/kg in diet of pigs. It was assumed that the pigs would increase their feed intake proportionately to the degree of dilution of the basal feed with the fibre source. Hence, all pigs would get nutrients that met requirements for growth. Each of the 31 diets (i.e. one control diet and 30 fibre-based diets) was fed to four randomly selected pigs in their own pens.

### 2.3. Analyses of physicochemical properties of diets

Tables 1–3 show composition of ingredients and physicochemical properties of diets used in the experiment. Ash was determined by combusting the sample in a furnace for 4 h at 550 °C according to method 942.05 of AOAC (2005). The dry matter (DM) was determined in an oven at 65 °C for 48 h according to the method 2001.12 of AOAC (2005). Crude protein (CP) was determined by using a Truspec N analyser (Leco, MI, USA) based on Dumas Combustion method 990.03 (AOAC, 2005). Crude fibre (CF) was determined according to the method 2001.12 of AOAC (2005). Neutral detergent fibre (NDF) and acid detergent fibre (ADF) were analysed using filter bags by means of a fibre analyser (Ankom 220; Ankom Technology Corp., Macedon, NY, USA). Gross energy (GE) was determined using a bomb calorimeter. Digestible energy (DE) of feeds

**Table 1**  
Composition of ingredients in a basal diet (B).

Ingredients	Composition (%)
Yellow maize whole	42.6
Full fat soya	17.6
Whole wheat	10.0
Wheat bran middlings	10.0
Soyabean oil cake	8.4
Sunflower oil cake	7.5
Fish meal	2.0
Vitamin mineral mix	1.9

Table 2

Nutritional composition of diets with different inclusion levels of Lucerne, Sawdust, Sunflower Hulls, Grass hay, Maize cob and Maize stover.

Inclusion level (g/kg)	Fibre source	Nutritional composition				
		DM (g/kg)	calc DE (MJ/kg) <sup>a</sup>	CP ; (g/kg DM)	EE (g/kg DM)	Ash (g/kg DM)
0	B	989.3	13.9	195.7	52.9	63.1
80	LC	989.3	13.4	192.6	49.2	63.1
	SD	989.2	12.3	117.4	51.5	53.7
	SH	989.3	13.5	166.9	54.1	56.6
	GH	989.5	13.1	186.4	51.7	60.4
	MC	989.3	13.2	185.7	51.2	59
	MS	990.1	13.8	178.6	50.1	62.3
160	LC	990	12.6	187.6	45.2	65.1
	SD	989.7	11.2	164.5	47.7	48.3
	SH	989.4	12.8	145.7	55.4	53.8
	GH	990	12.3	145.3	49.3	61.9
	MC	990.4	12.1	168.1	45.9	54.9
	MS	990	13.0	169.8	44.9	64.2
240	LC	990	11.8	174.3	41.5	67.2
	SD	989.7	10.2	145.8	45.55	44.1
	SH	989.6	12.2	134.6	55.7	51.6
	GH	990.5	11.2	135.5	40.7	63.9
	MC	990.5	11.0	152.8	45.2	53.2
	MS	989.9	12.0	147.8	39.3	65.3
320	LC	989.9	10.9	168.5	39.2	69.2
	SD	989.7	9.2	131.3	42.35	41.6
	SH	989.7	11.6	132.1	56.6	48.5
	GH	990.5	10.2	120.8	36.6	64.9
	MC	990.2	10.3	139.4	41.3	52.1
	MS	989.8	11.0	143	37.1	66.4
400	LC	989.9	10.2	165.9	33.3	71.3
	SD	989.8	8.1	117.5	36.7	39.2
	SH	989.8	11.3	123.5	57.3	44.9
	GH	990.8	9.2	112.9	32.4	65.9
	MC	991.1	9.3	116.4	39.9	46.7
	MS	989.8	10.1	141.3	31.9	66.5

B – Basal diet; LC – Lucerne; SD – Sawdust; SH – Sunflower Hulls; GH – Grass hay; MC – Maize cob; MS – Maize stover.

DM – Dry matter; GE – Gross energy; CP – Crude protein; EE – Ether extract; calc DE – calculated digestible energy.

<sup>a</sup> calc DE = 949 + (0.789 × GE) – (43 × %Ash) – (41 × %NDF) (Noblet and Perez, 1993).

was calculated as:  $DE = 949 + (0.789 \times GE) - (43 \times \% \text{ ash}) - (41 \times \% \text{ NDF})$  (Noblet and Perez, 1993). Bulk density of the feeds was measured according to the water displacement method described by Peterson and Baumgardt (1971). The method is based on the Archimedes Principle of determining the volume of a known mass of feed ingredient. Water holding capacity (WHC) was determined by centrifugation (Robertson and Eastwood, 1981). With the method, diets with inclusion levels of different fibres were soaked for 48 h in distilled water (1 g/500 ml) and samples containing 0.25–0.5 g dry fibre were then transferred to tared 25 ml polypropylene centrifuge tubes which contained distilled water (15 ml). After 1 h the contents of the tubes were centrifuged at 6000g for 15 min, excess water decanted and the tubes inverted and left to drain for 30 min. The fresh weight of the fibre was determined before freeze drying to determine the dry weight and hence WHC as g

Table 3

Composition of physical properties of diets with different inclusion levels of lucerne, sawdust, sunflower hulls, grass hay, maize cob and maize stover.

Inclusion level (g/kg)	Fibre source	Physical properties				
		CF (g/kg DM)	NDF (g/kg DM)	ADF (g/kg DM)	Density (g DM/ml)	WHC (g water/g DM)
0	B	26.1	192.3	88.4	1.45	3.76
80	LC	46.1	218.1	101.2	1.54	3.65
	SD	77.4	257.1	128.2	1.54	3.41
	SH	55.6	223.1	94.5	1.49	3.16
	GH	40.1	230.3	121.8	1.51	3.76
	MC	36.4	230.6	111.5	1.52	3.17
	MS	48.6	192.3	110.7	1.49	4.15
160	LC	67.6	261.1	126.4	1.46	4.26
	SD	129.7	318	177.7	1.49	3.95
	SH	86.3	269.8	110.7	1.44	3.48
	GH	55.1	284.2	166.7	1.5	4.28
	MC	47.9	294.4	147.4	1.47	3.57
	MS	72.2	228.1	142.9	1.31	5.24
240	LC	87.9	301.9	150.7	1.39	5.04
	SD	182.1	378.8	227.4	1.43	4.27
	SH	116.9	316.9	126.2	1.33	3.76
	GH	70.2	337.6	214.3	1.47	5.14
	MC	59.3	355.3	181.2	1.42	4.08
	MS	95.8	279.8	175.2	1.3	6.48
320	LC	108.9	343.9	175.5	1.32	5.56
	SD	234.4	439.8	277.3	1.4	4.79
	SH	147.6	363.3	142.1	1.31	4.06
	GH	85.2	391.2	259.4	1.31	6.45
	MC	70.8	401.3	218.4	1.25	4.41
	MS	119.4	332.3	207.4	1.12	7.49
400	LC	129.9	385.7	200.2	1.27	6.52
	SD	286.8	500.8	326.6	1.31	5.21
	SH	178.2	409.6	157.8	1.21	4.38
	GH	100.3	444.1	304	1.2	7.13
	MC	82.2	457.4	250.5	1.22	4.75
	MS	143	383.2	239.7	1.1	8.31

B – Basal diet; LC – Lucerne; SD – Sawdust; SH – Sunflower hulls; GH – Grass hay; MC – Maize cob; MS – Maize stover.

CF – Crude fibre; NDF – Neutral detergent fibre; ADF – Acid detergent fibre; WHC – water holding capacity.

water/g fibre. Three replicates for each sample were analysed.

#### 2.4. Recording of pig behaviour

Pigs were allocated at random and put on to their trial diets. They were allowed to adapt to the feed offered and to using the feeders for seven days, based on previous studies on dietary fibre (Ferguson et al., 1991; Kyriazakis and Emmans, 1991). Thereafter, behavioural activities were observed for each pig for 16 separate days, 8 h per day using overhead video cameras. Behavioural activities for each pig were continuously recorded for 8 h per day. Video cameras were mounted in such a way that each camera focused on six pigs in separate pens. The use of video cameras was to prevent disturbances from observers to the pigs whilst they are exhibiting their day-to-day normal



**Table 4**  
Ethogram of behaviours recorded for pigs in individual pens.

Behaviour	Description of behaviour
Eating	Time spent with the head in the feeder or very close to feeder (includes nosing feeder).
Drinking	Time spent with mouth at the nipple drinker.
Standing/sitting	Standing idle with body equally supported by all four legs/standing on fore-legs, hind quarter on the floor.
Lying down	Lying with shoulder or sternum in contact with floor.
Other	When the focal animal is not involved in any of the listed behaviours (nonpostural and feeding behaviours).

behavioural activities. In addition, number of visits to the feeder and duration of each visit on the feeder were recorded. Number of visits in the study is defined as movement of pig to the feeder to eat. The behavioural activities recorded were the time spent on eating, drinking, standing/sitting, lying down and other activities (object biting and licking). Description of the behavioural activities is shown in Table 4. Videos recorded to monitor behavioural activities of pigs were scored by the same person throughout for the whole experiment which lasted for four weeks. Proportion of time spent on different behavioural activities was calculated as time spent on a particular behaviour exhibited by the pig over the total time for all behavioural activities for the whole day.

Body weights of the pigs were recorded once a week for four weeks. Feed intake of the different types of fibres was recorded after every week for four weeks. Weights of feed refusals and spillages were subtracted from the total weight of the feed that was put into the feeder to determine feed intake.

Body weight usually affects behavioural activities exhibited by the pig. Time spent on each behavioural activity by a pig was, therefore, expressed as relative time spent to body weight of a pig. The formula used was

$$\frac{t}{B}$$

where  $t$  is the time spent on a particular behavioural activity by each pig and  $B$  is the body weight of each pig.

### 2.5. Statistical analyses

The PROC CAPABILITY procedure of SAS (2008) was used to check for normality of the data for time spent on each behavioural activity. Thereafter, logarithmic transformation was used to normalise the data for time spent on different behavioural activities since it was not normally distributed. A  $6 \times 6$  factorial analysis was used to test for significance of main effects and interactions on time spent on each behavioural activity using the General Linear Model procedure of SAS (2008) software. The model used was as follows:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha \times \beta)_{ij} + e_{ijk};$$

where,

$Y_{ijk}$  is the response variable (for time spent on different behavioural activities);  $\mu$  the overall mean response;  $\alpha_i$  the effect of fibre source ( $i$  the maize cobs (MC), grass hay (GH), lucerne hay (LH), maize stover (MS), sawdust (SD) and sunflower hulls (SH));  $\beta_j$  the effect of inclusion level ( $j=0, 80, 160, 240, 320$  and  $400$  g/kg);  $(\alpha \times \beta)_{ij}$  the interaction of effect of fibre source and effect of inclusion level;  $e_{ijk}$  the residual error.

The STEPWISE procedure of SAS (2008) was used to determine best predictor variables for time spent on different behavioural activities, average daily feed intake (ADFI), average daily gain (ADG), number of visits to the feeder and duration of each visit by to pig on the feeder. Variables incorporated in the model as predictor variables for time spent on different behavioural activities were DM, DE, CP, EE, ash, CF, NDF, ADF, BD and WHC. Relationships between time spent on different behavioural activities, ADFI, average daily gain, number of visits to the feeder and duration of each visit by to pig on the feeder, and properties of feeds were determined using the quadratic response surface model (SAS 2008).

### 3. Results

Effect of inclusion level and interaction of fibre source and inclusion level was significant ( $P < 0.05$ ). However, effect of fibre source was not significant. Time spent on different behavioural activities varied with inclusion level of fibre per day ( $P < 0.05$ ). Pigs spent most of their time lying down followed by eating, drinking and sitting/standing for all the fibre sources per day (Fig. 1). Time spent eating was observed to increase steadily with inclusion level of fibre. Conversely, time spent lying down decreased with inclusion level of fibre. Other behaviours which did not involve feeding activities (eating and drinking) and postural activities (lying down, sitting and standing) were infrequent in pigs subjected to treatment diets. The behavioural activities were, therefore, not reported in the current study.

Physicochemical properties of the feed were used to predict ADFI, ADG and time spent by pigs on different behavioural activities. Acid detergent fibre was the most important predictor variable for determining ADFI and ADG. Fig. 2 shows the relationships between ADF and ADFI, and ADF and ADG. As ADF content in feed increased, a significant increase ( $P < 0.05$ ) in ADFI was noted. A different trend was, however, noted for ADG. A significant decrease ( $P < 0.05$ ) in ADG was noted as ADF content of diets increased to a level where it becomes constant.

Table 5 shows regression equations predicting time spent on different behavioural activities of pigs from physicochemical properties of diets. Linear responses were noted for time spent eating and drinking from, DE, BD and ADF ( $P < 0.001$ ). A linear response was observed for time sitting/standing from BD as best predictor variable ( $P < 0.05$ ). A quadratic response, on the other hand, was observed for time spent lying down from ADF as best predictor variables ( $P < 0.001$ ).

Water holding capacity was the most important variable for predicting the number of visits made by the pig to the feeder per day ( $P < 0.05$ ). Fig. 3 shows the relationship

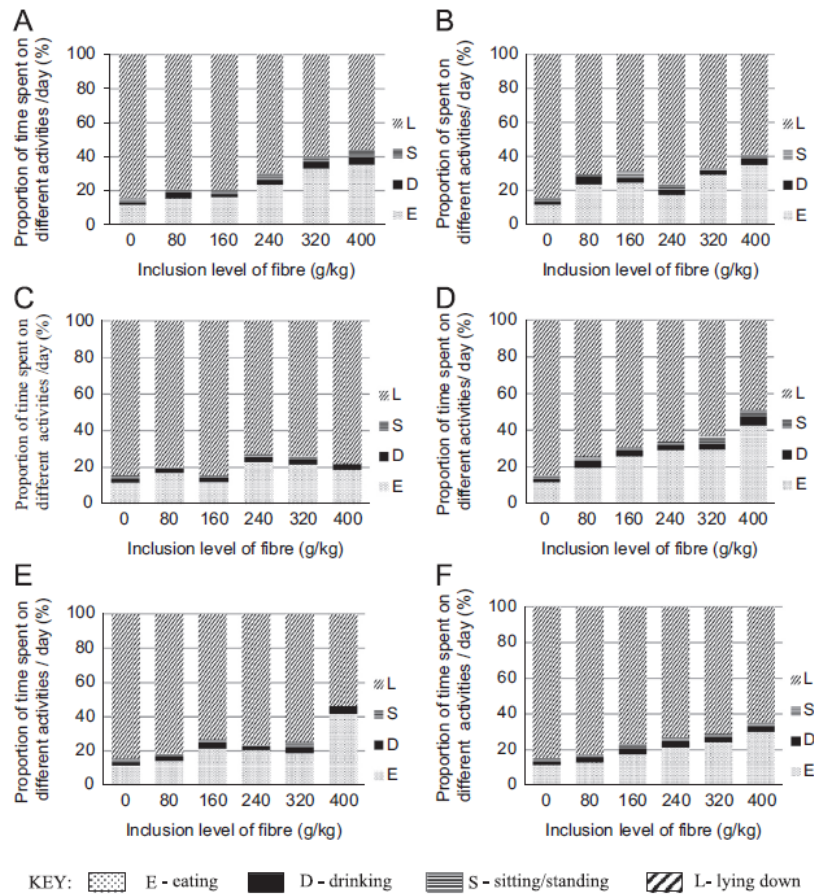


Fig. 1. Proportion of time spent on different behavioural activities by pigs fed (A) maize cob; (B) grass hay; (C) lucerne; (D) maize stover; (E) sawdust; (F) sunflower hulls; at different inclusion levels.

between mean number of visits to the feeder per day and water holding capacity. A significant decrease ( $P < 0.001$ ) in number of visits to the feeder by a pig per day was observed. A maximum of nine visits per day were made when water holding capacity was low in a diet. The visits were reduced to minimum of three visits per day at higher water holding capacity. For total time spent on each visit by a pig per day, ADF was the most important predictor variable ( $P < 0.05$ ). Duration of each visit on the feeder per day increased as the content in diets increased ( $P < 0.001$ ) (Fig. 4).

Other physicochemical properties that were not used to predict ADFI, ADG, time spent by the pigs on different behavioural activities, number of visits to the feeder and duration of each visit at the feeder were not statistically significant ( $P > 0.05$ ).

#### 4. Discussion

A preliminary study was conducted for the feeding behaviour trial where video cameras were used for 24 h. Most of the pigs in the study were lying down after 1600 h and activities started to increase after 0800 h the next

morning when they were offered feed. Observations were, therefore, made between 0800 h and 1600 h. The observation that pigs are inactive during the night hours is also in agreement with the study conducted by Whittemore et al. (2002).

The observation that time spent eating increased with inclusion level of fibre corresponds with the previous work in gilts (Rushen et al., 1999), dry sows (Brouns et al., 1997) and pregnant gilts (McGlone and Fullwood, 2001), which has shown that foods high in bulk made the pigs spend more time eating. Fibres are known as diluents in diets of pigs. Inclusion of fibres, as a result, reduces energy density of a diet (Rijnen et al., 2003; Robert et al., 1997) resulting in the pig to compensate for the reduced density at higher inclusion levels by eating more to meet the nutrient requirements for growth. When energy density is low, pigs are expected to eat more. Conversely, at high energy density of feed, the time spent eating is reduced. Gut capacity due to feed bulk was supposed to be revealed from the decrease in time spent eating as fibre level was increased. The observed linear increase in time spent eating might also be attributed to more time being channelled to mastication as the diet became coarser with

increasing fibre inclusion level (Wenk, 2001). Increased chewing may be necessary to breakdown coarse feed and properly mix feed with saliva to facilitate swallowing. Time spent masticating in the current study was, however, not measured and warrants further investigations.

Acid detergent fibre accounted for the effects on ADFI and ADG. Acid detergent fibre is an indigestible feed component in diets consumed by the pigs. The indigestible component increases transit time of digesta in the gut causing early satiety resulting from gastric signals in response to the elongation of the stomach wall. Due to distension of the gut and indigestible feed material (ADF), the pig will not acquire enough nutrients which meet requirements for growth from eating a diluted feed. This can account for the decrease in average daily feed intake and, hence, low weight gain at higher fibre inclusion

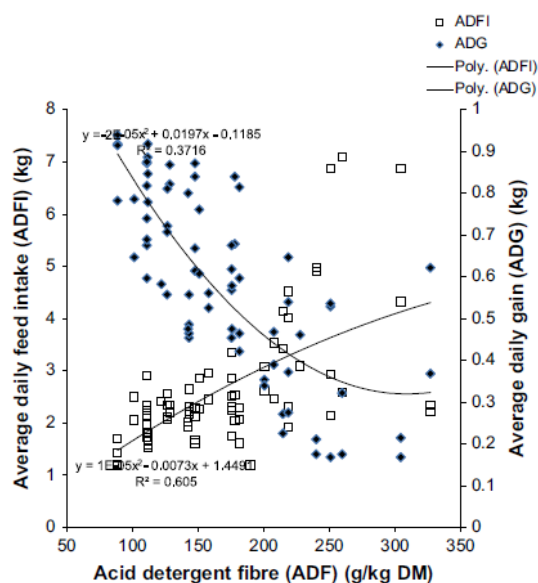


Fig. 2. Relationship between average daily gain (ADG) and acid detergent fibre (ADF) of feed, and average daily feed intake (ADFI) and Acid detergent fibre (ADF).

Table 5

Relationships of best predictor variables (bulk properties) of relative time spent on different behavioural activities to body weight to pigs.

Behavioural activity	Regression	Equation	R <sup>2</sup>	P-value
Eating	Linear	$E = -20.54(28.20)BD + 26.71(18.86)$	0.45	< 0.001
	Linear	$E = 0.031(0.015)ADF - 1.33(1.37)$	0.48	< 0.001
	Linear	$E = -3.16(1.491)DE + 27.47(8.312)$	0.4134	< 0.001
Drinking	Linear	$D = -1.22(1.43)BD + 1.37(0.958)$	0.41	< 0.001
	Linear	$D = 0.0012(0.000722)ADF - 0.091(0.0662)$	0.49	< 0.001
Lying down	Quadratic	$L = -0.00011(0.000047)ADF^2 + 0.055(0.019)ADF + 4.60(1.70)$	0.15	< 0.001
Standing/Sitting	Linear	$S = -8.98(6.53)BD + 7.104(4.37)$	0.11	0.0010

E – time spent eating; D – time spent drinking; L – time spent lying down; S – time spent sitting and standing. ADF – Acid detergent fibre; BD – bulk density.

levels. The low weight gain at higher inclusion levels could also be attributed to protein in feed that is made available for use by the pig. Nitrogen not available for use by the pig

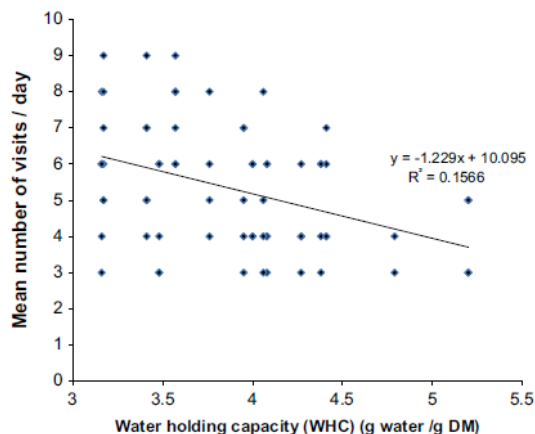


Fig. 3. Relationship between mean number of visits to the feeder and water holding capacity (WHC).

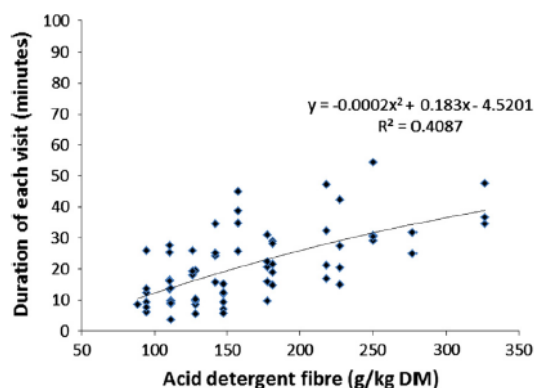


Fig. 4. Relationship between duration of visit on the feeder and acid detergent fibre.



would be bound by fibre (22%) resulting in low weight gains at high inclusion level of fibre (Bindelle et al., 2005). Low ADG at higher inclusion level of fibre indicates the negative animal welfare due to qualitative feed restriction.

The observed increase in time spent drinking with inclusion level of fibre was expected. There is a positive relationship between feed intake and quantity of water consumed by growing pigs (Jackson, 2007). Ratio of water to feed intake is estimated to be 3:1 (3 l of water/kg feed). Also, an adaptive mechanism that can be employed by a pig to digest fibrous feed material is to produce prodigious quantities of saliva (Dryden, 2008). Saliva facilitates in breaking down fibres and contains urea, which can be used by the microbes in the lower gut of a pig. A large proportion of saliva constitutes water and, to produce large quantities of saliva, the pig will need to increase water consumption. Saliva production was, however, not measured in the current study.

In the current study with growing pigs, decrease in time spent lying down came entirely from an increase in time spent eating. Lying down might be a sign of satiety in growing pigs receiving low fibrous diets. They would have acquired enough nutrients to satisfy their appetite. As pigs grow, their ability to utilise fibrous feeds is enhanced (Bindelle et al., 2005), making sows better able to utilise dietary fibre than growing pigs. Time spent lying down compared to other activities (eating, drinking and sitting/standing) may be a result of unavailability of space for the pig to manoeuvre freely. In the present study, the design of the individual pens did not permit such free movements. Pigs, as a result, spent more time lying down compared to other activities.

Time spent sitting/standing as observed in the study is an indication of pigs showing levels of discomfort by changing different positions and will, therefore, be trying to cope up with stress (Pearce and Paterson, 1993). The pigs also exhibit the activity to reduce the risk of shoulder lesions caused by hard floors in pens (Rolandsdotter et al., 2009). The observation that time spent sitting/standing increased with a decrease in BD of feed might also explain levels of alertness to sound and movement pigs exhibit as they will be anticipating a much more palatable feed.

Growing pigs adhere to a feeding pattern that provides them with sufficient feed which can meet their nutrient requirements for growth (Nielsen et al., 1996). This might be the case in the current study where pigs receiving high fibrous diets had few visits of long duration. The long visits at the feeder are motivated by hunger as the pig will be trying to get nutrients which meet requirement for growth.

Considerable number of visits of short duration in low fibrous diets compared to high fibrous diets, on the other hand, might be attributed to property diets. Low fibrous diets are easily fermented and absorbed for use by the pig. This stabilises glucose levels and enhance satiety on the short term. Long durations per visit per day at high ADF content of diets may be due to the texture of the feed, which results in a pig spending more time masticating. All this creates more time for the ingestion process and less time for other activities.

## 5. Conclusions

Time spent on different behavioural activities of growing pigs was significantly influenced by the inclusion levels of fibre in their diet. Pigs spent more time eating and less time lying down at higher inclusion level of fibres. Bulk density, DE, ADF and WHC were the most important bulk properties for predicting time spent on different behavioural activities. Inclusion of fibre in diets, therefore, alters behaviour of individually housed growing pigs and could have an impact on their welfare. Body weight of a pig influences time spent on different behavioural activities of growing pigs. Further investigations are, however, needed to determine time spent on different behavioural activities in grouped growing pigs subjected to diets with varying inclusion levels of fibre and also to predict time spent on different behavioural activities from the different types of non-starch polysaccharides.

## Conflict of interest statement

There are no conflicts of interest to declare.

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## Appendix 2: Ethical Approval of Research Project on Animals



UNIVERSITY OF  
KWAZULU-NATAL

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YAKWAZULU-NATALI

**Research Office  
Animal Ethics Research Committee**

Govan Mbeki Centre, Westville Campus,  
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Telephone 27 (031) 260-2273/35 Fax (031) 260-2384  
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14 March 2012

Reference: 061/12/Animal

Mr AG Bakare  
School of Agricultural, Earth and  
Environmental Sciences  
University of KwaZulu-Natal  
Pietermaritzburg Campus

Dear Mr Bakare

**Ethical Approval of Research Project on Animals**

I have pleasure in informing you that on recommendation of the review panel, the Animal Ethics Sub-committee of the University Research and Ethics Committee has granted ethical approval for 2012 on the following project:

***"Behaviour and gut health of pigs fed fibres."***

Yours sincerely

**Prof. Theresa HT Coetzer (Chair)  
ANIMAL RESEARCH ETHICS COMMITTEE**

Cc Registrar, Prof. J Meyerowitz  
Research Office, Mr N Moodley  
Supervisor, Prof. M Chimonyo  
Dean & HOS, Prof. A Modi

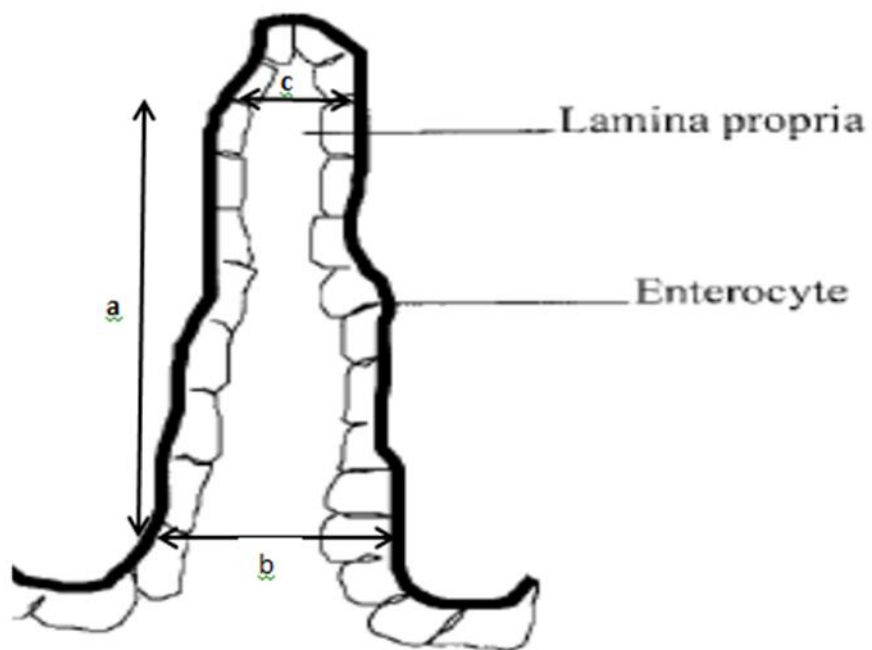


**Founding Campuses:**

- █ Edgewood
- █ Howard College
- █ Medical School
- █ Pietermaritzburg
- █ Westville



### Appendix 3: Measurement of apparent villi surface area (AVSA)



Outline of villus showing measurements for calculation of AVSA (a) villi height (b) villus basal width (c) villus apical width

Apparent villus surface area was estimated as:

$$AVSA = \frac{(b + c)}{2} \times a$$