



Various ratios of pectin to cellulose affect intestinal morphology, DNA quantitation, and performance of broiler chickens

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ABSTRACT

This study investigated the effect of various ratios of pectin to cellulose on intestinal morphology, DNA quantitation, and broiler chicken performance. Four hundred 1-day-old chickens were randomly divided into 4 experimental diets and 4 replications with 25 chickens each. Experimental diets consisted of a control (treatment 1) or diets containing different pectin:cellulose ratios: 2:1 (treatment 2), 1.5:1.5 (treatment 3), and 1:2 (treatment 4). Growth performance parameters were assayed on days 14, 21, and 42. Treatment 2 led to an increase in DNA content at day 14 ($P<0.05$), decreased small intestine weight at day 21, and increased small intestine weight at day 42 ($P<0.05$). The duodenal length increased by all experimental diets as compared to that of the control at day 21 but decreased at day 42 ($P<0.05$). The greatest jejunal length was observed with treatment 4 at 21 days of age ($P<0.05$). All treatments except treatment 3 induced a decrease in jejunal length at 42 days of age ($P<0.05$). The greatest and shortest cecum lengths were observed at 42 days of age with treatments 4 and 2, respectively ($P<0.05$). The smallest villous height, villous surface area, villous height:crypt depth ratio, and uppermost crypt depth were observed with treatment 2 at 14 days of age ($P<0.05$). Treatment 2 also resulted in the smallest villous height, villous surface area, and villous height:crypt depth ratio at day 21 ($P<0.05$). The greatest villous width was observed with treatment 1 ($P<0.05$). Overall, the growth performance parameters considerably declined by treatment 2.

In this study, we show that different ratios of non-starch polysaccharides (NSPs) can differentially affect the intestinal condition and performance. The effects of NSPs are governed by their solubility and fermentative ability. Indeed, the birds counteracted the effects of NSPs with changes in the intestinal length and weight, and with ageing. An increased ratio of cellulose to pectin partially hindered and moderated some of the effects of pectin.

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1. Introduction

The source of nutrients is abruptly changed as the yolk is replaced by exogenous feed after birds hatch (Uni et al., 1998). Thus, exogenous feed introduced during the critical phase of growth and development of the gastrointestinal tract (GIT) of broilers is of critical importance. The rapid development of the GIT and its related organs occurs when the ingested nutrients

are assimilated (Uni et al., 1998). The small intestine is anatomically completed at hatch (Lim and Low, 1977) but dramatically changes after receiving feed. If the nutrients are accessible, intestinal growth commences approximately 24 h after feed intake (Noy and Sklan, 1999); the weight of the small intestine starts increasing 48 h posthatch (Sklan and Noy, 2000) and can reach more than twice the initial size (Noy and Sklan, 1999). If feed is delayed, the small intestine weight, length, villous height, and crypt depth of different segments are reduced (Gonzales et al., 2003). Moreover, the morphology of the mucosa of different segments of the small intestine

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undergoes considerable changes with ageing, thereby increasing the efficiency of the intestinal functions (Uni et al., 1998; Geyra et al., 2001).

Dietary fibre is an important component of poultry diets. The inclusion of fibres increases the retention time of the digesta (from the crop to the gizzard) and stimulates gizzard function (Owusu-Asiedu et al., 2006). Consequently, the amount and type of fibres affect the development of the GIT and the growth performance (Owusu-Asiedu et al., 2006; Jiménez-Moreno et al., 2009). Insoluble fibres reduce the length of the GIT (González-Alvarado et al., 2007) as compared to soluble fibres in broiler diets (Viveros et al., 1994; Smits et al., 1997; Iji et al., 2001; González-Alvarado et al., 2007). In contrast, soluble fibres favour hypertrophy of the GIT and reduce nutrient digestibility and broiler performance (Lázaro et al., 2003; Owusu-Asiedu et al., 2006). Moreover, the relative weight of digestive organs such as the gizzard (Banfield et al., 2002; Jiménez-Moreno et al., 2009), the pancreas (Banfield et al., 2002), and the cecum (Smits et al., 1997; Banfield et al., 2002) is affected by soluble and insoluble non-starch polysaccharides (NSPs). Nevertheless, it is well documented that older broilers (Jorgensen et al., 1996) and turkeys have adapted to insoluble NSPs by increasing the GIT size, especially that of the small intestine and cecum (Sklan et al., 2003).

Little scientific literature exists on the effect of the physicochemical properties of the NSP source on the morphology of the intestine and the development of the GIT. Moreover, the existing results are equivocal. The inclusion of water-soluble pectin into diets of chicks (Langhout et al., 1999) and pigs (Hedemann et al., 2006) markedly affects the morphology of the small intestinal wall. The addition of viscous NSPs to chicken diets increases jejunal crypt depth but reduces ileum villous height and DNA concentration at 14 days of age (Iji et al., 2001). Pig diets containing insoluble fibres improve gut morphology by increasing villi length (Hedemann et al., 2006). The exact mechanisms of GIT adaptation to dietary fibres are not clearly understood yet. Importantly, digesta viscosity has been implicated in this response (Smits et al., 1997; Banfield et al., 2002). Non-absorptive fibres stimulate mucus development (Johnson and Gee, 1986), suggesting that, beyond viscosity, the fermentative ability is fundamental for GIT adaptation (Gee, 1996).

Soluble NSPs increase digesta viscosity and affect the viscosity of the aqueous fraction in the small intestine, thereby impairing its performance (Smits and Annonson, 1996). Insoluble NSPs make up the bulk of the total dietary fibres but have little or no effect on nutrient utilization in monogastric animals (Hetland et al., 2004). These products can shorten the retention time of the digesta and may cause low nutrient digestibility.

Although the effects of soluble and insoluble NSPs separately present in broiler diets have been occasionally investigated, little is known about the physiological effects of particular ratios of NSPs (soluble and insoluble) on the intestinal characteristics. Since broiler diets contain approximately 3% of fibres (Bach Knudsen, 1997), we considered 3% the lowest amount that can induce a significant effect in broiler diets. In this study, we used cellulose and pectin as substrate models of NSPs and evaluated the influence of various ratios of cellulose to pectin (in a total of 3% of the diet

composition) on intestinal development, morphology, DNA quantitation, and broiler performance. Such observations could help understanding the exact mechanisms of the interactions between soluble and insoluble NSPs in diets. In addition, it should be possible to extrapolate these results to any diet in which NSPs are present.

2. Materials and methods

2.1. Birds and diets

A total of 400 chickens were randomly divided into 4 treatments and 4 replications with 25 chickens each. The diet used as control (treatment 1) was formulated to meet the nutrient requirements of chickens (National Research Council, 1994). The experiment diets were formulated to assure the minimum amount of fibres (especially NSPs) and only the effect of NSPs was investigated. These treatments consisted of the control diet plus pectin (highly methylated) (HP, USA) and cellulose (UPM-Kymmene, Finland) in different ratios of 2:1 (treatment 2), 1.5:1.5 (treatment 3), and 1:2 (treatment 4) (Table 1). Feed rations were offered *ad libitum* at 1–21 or 22–42 days of age. Light was provided 24 h a day and was gradually reduced until 23 h a day. The temperature was also gradually reduced by 3 °C per week from the initial 32 °C. The weight gain (WG), feed intake (FI), and feed conversion ratio (FCR) were measured.

2.2. Intestine assay

At 14, 21, and 42 days of age, 2 birds per replicate were randomly selected and slaughtered. Immediately after dressing, the GIT was removed. The digestive tracts from the gizzard to the bile duct and from the bile duct to the Meckel's diverticulum were dissected and designated duodenum and jejunum, respectively. The tract between the Meckel's diverticulum and the ileocaecal junction was designated ileum. The caeca were also dissected. The length and weight of all mentioned segments were recorded.

2.3. Light microscopy

At 14, 21, and 42 days of age, middle sections of the jejunum (3–4 cm) of 2 birds per replicate were cut and histological indices were measured according to the method reported by Iji et al. (2001). Formalin-fixed jejunum tissue samples were dehydrated, cleared, and impregnated with paraffin. The processed tissue was then embedded in paraffin wax and cut into 6-µm sections with a LEICA RM 2145 microtome. To avoid wrinkles, sections were floated on warm water (55–60 °C) prior to mounting on 10% poly-L-lysine-coated slides. The slides were stained with haematoxylin and eosin.

Histological indices were determined by use of a computer-aided light microscopic image analyser (Motic Images, 2000 1.2, Scion Image). The villous height (from the top of the villous to the crypt opening) and crypt depth (from the base of the crypt to the crypt opening) were measured, and the villous surface area (the area of the villous per length unit per bird) and villous height: crypt depth ratio were calculated. The mean

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