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MASTER THESIS

Effect of fiber inclusion in the rearing diets and energy concentration of the laying diets on productive performance and egg quality of brown egg-laying hens from 18 to 46 weeks of age

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تعلموا العلم، و تعلموا السكينة و الوقار

عمر بن الخطاب

****

Acquire knowledge, and learn tranquility and dignity

Omar ibn al-Khattab

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ABBREVIATION KEYS

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ºC: degree Celsius

%: percentage

*: $P<0.05$

**: $P<0.01$

***: $P<0.001$

AA: amino acid

ADFI: average daily feed intake

AMEn: nitrogen-corrected apparent metabolizable energy

Arg: arginine

BW: body weight

BWG: body weight gain

Ca: calcium

CF: crude fiber

cm: centimeter

cm^2: square centimeter

CP: crude protein

Cys: cysteine

d: day; día

DM: dry matter; materia seca

EE: ether extract

et al.: and others

FCR: feed conversion ratio

FEDNA: Fundación Española para el Desarrollo de la Nutrición Animal; Spanish Foundation for the Development of Animal Nutrition g: gram, gramo

GIT: gastro intestinal tract

GIT: gastrointestinal tract

GLM: general lineal model

GMD: geometric mean diameter

GSD: geometric standard deviation

h: hour; hora

HCl: hydrochloric acid

IC: índice de conversión

Ile: isoleucine

IU: international unit

kcal: kilocalorie

kg: kilogram

Lys: lysine

M: milliones

m: meter; metro

m^2: square meter

Met: methionine

mg: miligram

min: minute

mm: milimeter

N: nitrogen

n: number of replicates per treatment

NRC: National Research Council

NS: non significant difference ($P > 0.10$)

NSP: non starch polysaccharides

P: phosphorus

P: probability

ppm: parts per million

RL: relative length (cm/kg body weight)

RW: relative weight (g/kg body weight)

SAS: Statistical Analysis Systems

SCWL: Single Comb White Leghorn

SD: standard deviation

SEM: standard error of the mean

SI: small intestine

T: tons

Thr: threonine

Trp: tryptophan

Val: valine

vs.: versus

wk: week

µm: micrometer

CHAPTER 1:

ABSTRACT, RESUMEN, RÉSUMÉ

ABSTRACT

ABSTRACT

A common commercial practice used to enhance feed intake in young hens at the onset of the egg laying cycle consists in increasing the fiber content of the rearing diets. The inclusion of fiber in these diets might help in the adaptation of the gastrointestinal tract (**GIT**) of the bird for higher consumption during the laying phase, which may result in improved hen productivity. The experimental consisted in 12 treatments with 6 rearing diets (0-17wk) and 2 laying diets (18-46wk) in a factorial arrangement. Two of the rearing diets had similar nutrient content and were based on barley or corn. The other 4 diets formed a 2x2 factorial with 2 fiber sources (straw and sugar beet pulp) included at 2 or 4% at expense of in the corn diet. In the laying phase, half of the pullets of these 6 dietary treatments were fed diets with 2,650 or 2,750 kcal AME_n/kg but with similar nutrient content per unit of energy. No interactions between rearing and laying phase diets were found. Type of diet during the rearing did not affect laying hen productivity or the relative weight (% BW) of the GIT, the relative length (cm/kg BW) of the hens or of the small intestine, or the pH of the gizzard contents. Hens fed the high energy diet during the laying period ate less feed ($P<0.001$) and had better FCR ($P<0.01$) and higher BW gain ($P<0.05$) than hens fed the low energy diet. Egg quality was not affected by dietary treatment except for the dirty eggs that was higher ($P<0.001$) for the low energy diet. We conclude that neither the main cereal nor the inclusion of a fiber source in the rearing phase diets affected GIT development or laying hen production at 46 wk of age. Also, the use of high energy diets during the laying phase reduced ADFI and improved FCR but did not affect egg production or egg weight.

Key words: energy, pullet, straw, sugar beet pulp

RESUMEN

RESUMEN

Una práctica común para mejorar el consumo de alimento en gallinas jóvenes al inicio de la puesta consiste en aumentar el contenido de fibra de las dietas de recría. La inclusión de fibra podría favorecer la adaptación del tracto gastrointestinal (**TGI**) del ave para tener un mayor consumo durante la fase de puesta, lo que puede mejorar la productividad de las gallinas. El diseño experimental fue completamente al azar con un modelo factorial con 12 tratamientos con 6 dietas en la fase de recría (0-17semanas) y 2 dietas en la fase de puesta (17-46 semanas). Dos de las dietas de la fase de recría se basaron en cebada o maíz y las 4 dietas restantes se agruparon en un modelo factorial 2x2 con 2 fuentes de fibra (paja y pulpa de remolacha) incluidas a niveles de 2 ó 4% en la dieta a base de maíz. En la fase de puesta, la mitad de las pollitas de estos 6 tratamientos se alimentaron con dietas con 2.650 kcal AMEn / kg y la otra mitad con 2.750 kcal AMEn / kg, con el mismo contenido de nutrientes por unidad de energía. No se encontraron interacciones entre las dietas de fase de recría y las de fase de puesta. El tipo de dieta en la fase de recría no afectó a la productividad de las gallinas o al peso relativo (% peso vivo) del TGI, el pH del contenido de la molleja, o la longitud relativa de las gallinas (cm / kg de peso vivo) o del intestino delgado. Las gallinas alimentadas con la dieta alta en energía durante la fase de puesta comieron menos alimento ($P < 0{,}001$), pero presentaron mejor índice de conversión ($P < 0{,}01$) y una mayor ganancia de peso ($P < 0.05$) que las gallinas alimentadas con la dieta baja en energía. La calidad del huevo no se vio afectada por el tratamiento a excepción de la incidencia de huevos sucios, que fue mayor ($P < 0{,}001$) para la dieta baja en energía. En conclusión, ni el tipo de cereal ni la inclusión de fuentes de fibra en las dietas de recría, afectaron al desarrollo del TGI ni a la producción de las gallinas a 46 semanas de edad. Además, el uso de dietas de alta energía durante la fase de puesta reduce el consumo medio diario y mejora el índice de conversión, sin afectar a la producción de huevos o al peso del huevo.

Palabras clave: energía, paja, pollitas, pulpa de remolacha

RÉSUMÉ

RÉSUMÉ

Une des pratiques utilisées pour améliorer la prise alimentaire chez les jeunes poules au début du cycle de ponte consiste à augmenter la teneur en fibres de l'alimentation des poulettes. L'inclusion de la fibre dans ces régimes pourrait aider à l'adaptation de l'appareil digestif de la poule pour avoir une consommation plus élevée au cours de la période de ponte, ce qui peut améliorer sa productivité ultérieurement. Le dispositif expérimental était complètement aléatoire avec 12 traitements avec 6 régimes utilisés durant la phase d'élevage des poulettes (0-17semaines) et 2 régimes utilisés durant la phase d'élevage des poules pondeuses (17-46semaines) suivant un arrangement factoriel. Deux des régimes alimentaires des poulettes étaient basés sur l'orge ou du maïs , et les autres quatre régimes restants ont formé un factoriel 2x2 avec deux sources de fibres (la paille et la pulpe de betterave à sucre) incluses à des niveaux de 2 et 4% dans le régime alimentaire basé sur le maïs. Durant la période de ponte, la moitié des poulettes de ces 6 traitements alimentaires ont reçu une alimentation à 2.650 ou à 2.750 kcal EMAn / kg avec une même teneur en éléments nutritifs par unité de EMAn pour 2.650 ou 2.750 kcal AMEn / kg. Il n´y a pas d´interactions observées entre les régimes alimentaires utilisés durant la phase des poulettes et ceux qui ont été utilisés durant la phase de ponte. Le type d'alimentation distribué aux poulettes n'a pas affecté la productivité des poules pondeuses et même le poids relatif (% Poids vif) du tube digestif, le pH du contenu du gésier, ou la longueur des poules (cm / kg de poids vif) ou de l'intestin grêle. Les poules nourries avec le régime contenant un niveau élevé d´énergie durant la période de ponte ont présenté une baisse de consommation alimentaire (P <0,001), et par contre un meilleur indice de consommation (P <0,01) et un gain de poids plus élevé (P <0,05) que celui des poules nourries avec un régime alimentaire contenant un niveau faible d´énergie. La qualité des œufs n'a pas été affectée par les traitements alimentaires pratiqués, sauf qu´ il y ait une augmentation du pourcentage des œufs sales (P <0,001) affectée par la diète qui contient un niveau faible d´énergie. Nous concluons que ni le type de céréale utilisé, ni l'inclusion des sources de fibres dans le régime alimentaire des poulettes n'ont affecté le développement du tube digestif à 46 semaines d'âge ou même la production d´œufs. En outre, l'utilisation des régimes alimentaires avec un niveau élevé en énergie au cours de la période de ponte réduit la consommation alimentaire et améliore l´indice de consommation, mais il n'y a pas d'incidence que ce soit sur la production d'œufs ou sur le poids des œufs eux-mêmes.

Mots clés: énergie, la paille, la pulpe de betterave á sucre, poulette

CHAPTER 2:

LITERATURE REVIEW AND OBJECTIVES

1. General comments

The present Master Thesis has been carried out thanks to a CDTI research project (Ministerio de Ciencia e Innovacion, Madrid) with Camar Agroalimentaria. S. L, a leading egg production company placed in Toledo (Spain).

The objectives of the egg production industry to attain optimal performance and adequate economic output of a given flock of laying hens.Thus;

- ✓ The economic success of the egg industry depends on total egg mass produced by hen during the whole lay period. The total egg mass production per hen is influenced not only by the length of the laying period but also by the persistency of the egg production curve, the number of eggs produced, and the size of these eggs.

- ✓ The economic success of a laying hen operation requires a high peak of egg production of adequate size at the start of the laying cycle and a good persistency throughout the laying period. It is widely accepted that a high peak of production is positively related with egg mass production.

2. Literature review

2.1. Introduction

Spain, with around 37 M of industrial laying hens in 2012 is one of the leader egg producer countries in EU. This production corresponds to 12.3% of the total census of the EU-27 (European Commission, 2012). Total egg production in Spain is around 795,000 t per year (FAOSTAT, 2012a). Moreover, in 2011, Spain exported 92,000 t, mostly to other European countries, especially Germany (FAOSTAT, 2012b). Egg consumption in Spain is estimated to be over 220 eggs per person per year, equivalent to 14 kg of egg (FAOSTAT, 2012a). The use of non-enriched cages for egg production has been banned in the EU-27 in January 2012 and consequently, Spanish producers had to reconvert their facilities to meet the legislation which has caused a redaction of egg production. At present time, laying hens are housed primarily in enriched cages (93.5%) with a small percentage of hens housed on floor (4%) or under free range (2.6%) conditions (REGA, 2012).

The profitability of the egg industry depends of 3 keys factors; number of eggs per hen housed, egg size, and percentage of eggs that reach the table of the final consumer. One of the most critical points to improve egg rate and egg quality is proper management and feeding and nutrition practices of the pullets during the rearing phase. Adequate body weight (**BW**) at sexual maturity of pullets and uniformity are positively correlated with high peak production, persistency of egg production curve, and proportion of large eggs (Summers and Leeson, 1994). Adequate feed intake (**FI**) during the rearing phase will result in pullets with a well-developed gastrointestinal tract (**GIT**) that will allow the birds to meet their nutritional requirements, especially in the critical period of the onset of laying. However, the information available on nutrient requirements and the influence of dietary fiber on growth and development of the GIT is very limited in pullets. More information is needed to help the nutritionists to formulate diets that maximize FI and BW gain (BWG) and at the same time allows proper development of the GIT of the pullets and an adequate uniformity of the flock.

2.1. Utilization of fiber in poultry diets

The dietary fiber (**DF**) has typically been an undesirable component of diets. However, two main factors have increased recently the interest of dietary fiber inclusion in diets for chicks. The first one is the need for using non-traditional ingredients with larger concentrations of fiber and the second one is a growing public concern regarding the use of in-feed antimicrobials for livestock production. The concern is because of the potential transmission of antibiotic resistance from animals to humans (Witte et al., 2002). Therefore, the interest in dietary alternatives to promote or maintain health in chicks has grown.

Some authors have indicated that DF is the major source of energy to support microbial populations in the gastrointestinal tract (Bach Knudsen et al., 1991; Jørgensen et al., 1996). The chemical properties of DF, as well as its fermentation in the intestine, have an important role in keeping the balance between communities of commensals and pathogens (Montagne et al, 2003). Also, the intestinal microbiota has the capacity to modulate the host's immune response (Bauer et al., 2006). Thus, the source and concentration of DF in the diets of chicks represent an opportunity to manipulate the susceptibility and capacity to recover from enteric diseases. On other hand, DF might help to protect the mucosa of the GIT, reducing the incidence of ulcers and colitis, as well as chronic inflammation of the digestive mucosa (Mateos et al., 2012).

2.2.1. Definition of dietary fiber

The DF was originally defined as ""the skeletal remains of plant cells in the diet, which are resistant to hydrolysis by the digestive enzymes of man" (Trowell, 1974). This excluded the polysaccharides added to the diet such as food additives (e.g. plant gums, modified cellulose) and the definition was later expanded to include "all polysaccharides and lignin, which are not digested by the endogenous secretions of the human digestive tract (Trowell et al., 1976). Despite extensive research over the last quarter of the 20th century, the definition of DF has been continuously debated and no universal agreement has yet been reached (Cummings et al., 1997; DeVries et al., 1999). However, currently most researchers are using either a physiological or a chemical definition. According to the physiological definition, DF is "the dietary components resistant to degradation by mammalian enzymes", while chemically it is "the sum of non-starch polysaccharides (**NSP**) and lignin" (Theander et al., 1994).

The DF is predominantly found in plant cell walls and consists of a complex mixture of NSP that are associated with a number of other, non-carbohydrate components (McDougall et al., 1996). It is now known that DF exhibits a range of physical properties that act in concert with the chemical properties to determine the physiological effects of the feed. In the analysis of DF, it is common to characterize DF in accordance with its solubility. Soluble NSP (**s-NSP**) tend to impair digestion and absorption of nutrients in the fore-gut, whereas the negative effects of insoluble NSP (**i-NSP**) are less pronounced. The i-NSP primarily act in the large intestine where, increase faecal bulk, dilute colonic contents and decrease transit time.

2.2.2. Physicochemical properties of fiber

The DF exhibits a range of physical properties that act in concert with the chemical properties to determine the physiological effects in animals. The physicochemical properties that are more commonly studied include: particle size and bulk volume, hydration properties and the viscosity, lipid adsorption capacity, surface area and porosity characteristics, and ion exchange capacity (Guillon and Champ, 2000, 2002).

2.2.2.1. Particle size and bulk volume

Particle influences a number of events occurring in the GIT, such as transit time, fermentation capability, and bulk of the fecal contents (Guillon and Champ, 2000). Particle size depends on the nature of cell walls present in the ingredients and the degree of processing. Auffret et al. (1994) indicated that grinding not only reduces particle size but also modifies the structure of the fiber. Particle size of the fiber component may vary during transit throughout the GIT as a result of chewing, grinding in the gizzard and bacterial degradation in the large intestine. In addition, some components involved in the cohesiveness of the fiber matrix may be solubilized during the digestive process. Consequently, particle size of the fiber before ingestion is not necessarily relevant to assess the potential action and benefits of fiber on transit time. The analysis of particle size distribution is typically carried out by dry sieving through a series of sieves with decreasing mesh size as described by American Society of Agriculture Engineers (1995). The measurement of particle size in wet form may be more relevant when comparing the bulk volume of fiber in the GIT than the measurements in dry form. In any case, the method and system utilized for the determination of particle size of fiber sources should be always indicated.

2.2.2.2. Hydration properties and the viscosity

The hydration properties are characterised by the swelling capacity, solubility, water holding capacity (**WHC**) and water binding capacity (**WBC**). The first part of the solubilisation process of polymers is swelling, in which incoming water spreads the macromolecules until they are fully extended and dispersed. Afterwards, they are solubilised (Thibault et al.,1992). The DF swells to a variable extent in water. For example, isolated pectins swell quite well, but when there are less hydrophilic substances in the mesh it swells worse. Solubilisation is not possible in the case of polysaccharides that adopt regular, ordered structures (e.g. cellulose or linear arabinoxylans), where the linear structure increases the strength of the non-covalent bonds, which stabilise the ordered conformation. Under these conditions only swelling can occur (Thibault et al., 1992). The WHC and WBC are also used to describe hydration properties and have been used interchangeably in the literature since both reflect the ability of a fiber source to incorporate water within its matrix. The WBC is determined by the physico-chemical structure of the molecules, and by the pH and electrolyte concentration of the surrounding fluid. Thus, during passage throughout the gut, DF may swell to a variable extent.

The majority of polysaccharides gives viscous solutions when dissolved in water (Morris, 1992). The viscosity is primarily dependent on the molecular weight of the polymer and the concentration. Large molecules increase the viscosity of diluted solutions and their ability to do this depends primarily on the volume they occupy.

2.2.2.3. Lipid adsorption capacity

The ability of DF sources to retain oil can be important in nutrition where the ability to absorb or bind bile acids and increase their excretion is associated with plasma cholesterol reduction (Schneeman, 1999). The DF sources rich in uronic acids and phenolic compounds may have a hypo cholesterolaemic action.

2.2.2.4. Surface area characteristics and porosity

The available surface area and porosity of DF influence its availability to microbial degradation in the colon. The porosity of a fiber matrix (gross porosity > 1 μm; or microporosity < 1 μm) is defined by macromolecular interaction and combination with cell walls and also by the cohesiveness of the tissues (Guillon and Champ, 2000).

2.2.2.5. Ion-exchange capacity

There is a suspicion that fibrous fraction impairs mineral absorption because of polysaccharides such as the pectins, and associated substances, such as phytates in cereal fibers. In fact, charged polysaccharides do not have nutritionally effect on mineral and trace element absorption while associated substances may have a detrimental effect.

2.2.3. Effects of dietary fiber on the functionality of the gastrointestinal tract of poultry

Poultry do not produce the enzymes required to digest fiber present in the feed. Grains constitute the dominating ingredients of poultry feeds and the birds appear rather well suited for such diets as the capacity to digest e.g. wheat starch is typically very high (Svihus and Hetland, 2001; Hetland et al., 2003). Therefore, it might be surprising that poultry should not be classified as granivores, and consequently best described as omnivores (Klasing, 2005). Even if fiber cannot be digested by broiler chickens and laying hens, it is not inert. A brief look at some characteristic segments of the digestive tract of poultry may justify such an assumption.

The anterior segment of the digestive tract is characterized by an enlargement of the oesophagus, called the crop. The crop is used as a food storage organ and filling of the crop inhibits food intake (Richardson, 1970). It may be that the fiber fraction of the feed is important to the sensation of satiety, as the quality of ingested fiber affects the rate of entry and time of retention in the crop (Vergara et al., 1989).

Ingested items are either retained in the crop, or passed down directly to the next segment of the digestive tract. The anterior compartment of this segment is called the glandular stomach, or proventriculus, and constitutes the site for hydrochloric acid and pepsinogen secretion. The posterior compartment is named the ventriculus, or more commonly, the gizzard. In the gizzard, the particle size of ingesta is reduced by means of grinding, while the chyme is mixed with the secretions of the proventriculus. To function properly, however, the gizzard requires mechanical stimulation. Here, the excitatory effect of fibrous materials on the gizzard is well documented (Rogel et al., 1987; Hetland et al., 2005; Steenfeldt et al., 2007).

Once ingested items have been ground to a critical size, the particles are moved into the small intestine. The size of the fowl intestine has been adapted to flight and is therefore comparatively short. To compensate for this reduction in digestive capacity, poultry reflux digesta between various locations of the digestive tract (Sklan et al., 1978; Duke, 1988). Although data are quite limited, ingested fiber was shown to influence peristalsis in man (Cherbut et al., 1994). In fact, fiber is believed to influence the gastroduodenal reflux of digesta in poultry (Hetland et al., 2003).

Nutrients are digested and absorbed along the small intestine; but as previously indicated, poultry cannot degrade the fiber fraction of the feed. Instead, completely or partly undigested fractions of water soluble digesta particles, including fiber, are moved by means of anti-peristalsis into the caeca. Coarser fractions of digesta are prevented from entering the caeca by a filter-like meshwork of villi stretching into the lumen (Björnhag, 1989). The caeca harbour a large number of bacteria with the capacity to use the energy present in the fiber. Some metabolic end products from this fermentation, such as short chain fatty acids, can finally be absorbed and utilized by the bird.

In summary, it seems that fiber exerts different effects on the digestive tract of the fowl. However, to assess the significance of the fiber fraction of poultry feeds, it is necessary to understand the nature of the different fiber fractions where they come from and what their features are.

2.2.4. Effect of fiber quality in poultry

Today, following the development of more sophisticated methods of fiber determination, it is known that different fiber fractions have different properties and should consequently be viewed differently. The solubility is an important feature of fibers as it largely determines their effect on production performance (Choct, 2002). More specifically this effect is primarily mediated by the actions of the NSP in the digestive tract. In some cases these interactions are manifested on a gross anatomical level, sometimes they occur on a cellular or even molecular level. In general, it seems reasonable to suggest that s-NSP tend to act detrimentally on digestion whereas the negative effects of i-NSP are less pronounced, and in some cases, even beneficial to the bird. The underlying factors of such a difference will now be scrutinized.

2.2.4.1. The soluble fraction of dietary fiber

Chemically, the WBC of fiber reflects the frequency of bonds between the carbohydrate units, which determines how much water can be trapped in the intermolecular spaces (Carré, 2002). The WBC of the feed may have both positive and negative aspects for birds. In systems where restricted feeding is practised, such as in broiler chicken parent stocks, a high WBC of the feed has been suggested to distend the digestive tract, thereby possibly increasing the sensation of satiety (Hocking et al., 2004). On the other hand, WBC of fiber increases the water consumption in birds (Langhout et al., 1999). This fact can implicate an elevation of moisture of litter increasing the risk of a lower hygiene and a higher foot-pad inflammation incidence (Wang et al., 1998). In fiber with low frequency of intermolecular bonds, such as in pectin substances, less water is entrapped in cavities between the molecules. Nevertheless, more water is directly associated with the many saccharide units due to their hydrophilic nature (Carré, 2002). Thus, fiber characterized by long-stretched chains of sugar units can associate with large amounts of water, resulting in increased viscosity. It is important to take into account that the physicochemical characteristics of the feed may not always be static. For example, it is likely that changes in the degree of polymerisation of the fiber explain the increased feed viscosity often seen in response to high pelleting temperatures (Cowieson et al., 2005). The viscosity-inducing properties of s-NSP are believed to be a key factor in their detrimental effect on feed utilization (Smits and Annison, 1996; Langhout et al., 1999). In humans, water-binding fiber reduces ingestion, the rate of glucose absorption by altering the motility pattern of the small intestine (Cherbut et al., 1994). Similarly, high viscosities are believed to hinder the contact of digesta with the intestinal epithelium of poultry and digestive enzymes impairing feed utilization. This effect is particularly evident in fat digestion, due to emulsification requires rigorous mixing of digesta (Bedford, 2002). Besides, the magnitude of the negative effects of s-NSP on feed utilization is mediated by the bacteria present in the digestive tract of the bird (Langhout et al., 2000; Maisonnier et al., 2003). Many of the more than 640 bacterial species residing in the gut have the capacity to use fiber as substrates for growth, but they also compete with the host for other nutrients such as nitrogenous compounds, fat and minerals (Apajalahti et al., 2004). Although some bacteria are believed to benefit the host, many species do not, and the competition for nutrients implies that any factors hindering an efficient feed digestion and utilization for birds are likely to promote the microflora. For this reason, reduction in bird performance due to high levels of s-NSP in the feed is often mirrored in quantitative and qualitative changes of the bacterial community (Langhout et al., 1999). The negative impact of s-NSP is not only manifested in disadvantageous allocations of nutrients to bacterial growth in the gut lumen of birds, but can also be expressed as damage to the intestinal mucosa. Serving as the interface between the constituents of digesta and the underlying mucosa, the epithelial cells must facilitate an efficient absorption of digestion derivates while preventing microbes from damaging or infiltrating the tissue. The intestinal mucosa is ordered in finger-like protrusions into the lumen, called villi, in order to increase the absorptive area. At the villus tip epithelial cells are continuously sloughed off and replaced by migrating cells formed in the crypts of Lieberkühn at the villus base. It is assumed that the length of villi reflects their absorptive capacity, and some studies have demonstrated that s-NSP have a deleterious effect on villus height (Viveros et al., 1994; Teirlynck et al., 2009), although results are not consistent (Rolls et al., 1978; Langhout et al., 1999; Iji et al., 2001). Other authors have found that s-NSP increase the depth of the crypts, which possibly would indicate an increased rate of cell renewal, and indirectly, more damage of the villus tip (Wu et al., 2004). In general, divergences in the literature indicate that the effects of s-NSP on mucosal morphology are not clear-cut. Today, the detrimental effects of s-NSP in the feed are largely alleviated by the routine use of fiber-degrading enzymes in the feed. The efficacy of many mono- and multicomponent enzyme products is well documented, but their mode of action has been a matter of debate. In essence, the question has resolved around whether the benefits of the enzymes should be attributed to their capacity to reduce the viscosity of the digesta, or their capacity to release nutrients encapsulated in the fiber matrix of the grain cell walls, or both (Bedford, 2002).

2.2.4.2. The insoluble fraction of dietary fiber

As previously mentioned i-NSP share some physical features with s-NSP, but the negative effects exerted by i-NSP are much less severe. For example, i-NSP may display some water-holding capacity, but their viscosity-inducing properties are relatively low (Smits and Annison, 1996). Although i-NSP are less rapidly fermented by the microflora, they are not inert, and may indeed interact with the constituents of the digesta. In theory, these effects may partially be mediated by the intestinal microflora as i-NSP tend to favour certain bacterial species (Baurhoo et al., 2007), some of which display a capacity to deconjugate bile salts (Guban et al., 2006). Further, depending on the pH, some NSP may associate with cations (Smits and Annison, 1996). Although certain part of fiber may improve mineral retention in broiler chickens (Ortiz et al., 2009), the effects of fiber on different minerals are ambiguous (van der Aar et al., 1983). Overall, there is a scarcity of in vivo experiments on the effects of i-NSP on the uptake of nutritionally important minerals for poultry like calcium. In contrast, the suggestion that i-NSP may stimulate the digestion of macronutrients and benefit bird welfare has gained increasing attention in the scientific community. The majority of hypothesized explanations for these observations relate to the effects of i-NSP on gastrointestinal function. Whereas water-soluble particles dissolve in the intestinal fluids and rapidly pass the anterior digestive tract, i-NSP are retained for a longer period of time in the crop and gizzard (Ferrando et al., 1987; Vergara et al., 1989). Increased retention time of digesta in the anterior digestive tract may alleviate problems with over-consumption of feed in broilers (Svihus et al., 2010), probably as feed intake is partially controlled by distension-sensitive receptors in this section of the digestive tract (Duke, 1988). Numerous studies have demonstrated that i-NSP stimulate gizzard development (Riddell, 1976; Hetland et al., 2003, 2005; Jiménez-Moreno et al., 2010) and reduce the occurrence of spontaneous gizzard erosion and ulceration (Kaldhusdal et al., 2012). The notion that gizzard development is important to digestion derives from several experiments. For example, gizzard size is correlated with the efficiency of mechanical degradation of raw potato starch granules (Rogel et al., 1987), and broiler chickens selected for high utilization of a wheat-based diet displayed increased gizzard weights (de Verdal et al., 2010). The effects of i-NSP on gizzard function are not restricted to the control of feed intake and the digesta particle size, but probably also involve the regulation of digesta flow throughout the digestive tract. Ingestion of i-NSP increases bile acid content and amylase activity in digesta, hypothetically via an increased gastroduodenal reflux (Hetland et al., 2003). An increased frequency of small intestinal reflux has been suggested to facilitate the digestion of nutrients (Duke, 1997). In addition to the aforementioned effects of i-NSP on gizzard grinding capacity, the elevated contents of bile acids in digesta reported by Hetland et al. (2003) may help explaining the increased digestibilities of fat in young broiler chickens fed i-NSP from soybean hulls and oats (Jiménez-Moreno et al., 2009). When considering the effects of i-NSP on nutrient utilization, the importance of particle size should be emphasized. Since digesta must pass through the gastroduodenal junction of the gizzard, larger particles are retained for a longer time and consequently, stimulate gizzard function more than fine particles do (Jiménez-Moreno et al., 2010).

2.3. Energy level in poultry diet

Hens eat to satisfy their energy requirements and therefore an increase in the energy content of the diet should decrease FI proportionally (Hill et al., 1956).

2.3.1. Energy content of laying hen diets

2.3.1.1. Effects on productive performance

Bouvarel et al. (2010) reviewed a serie of experiments conducted with laying hens during the last 20 years. They reported that as an average, a 10% of increase in AMEn content of the diet reduced FI by only 5.5%. Changes in energy concentration of the diet have resulted in contrasting results with respect to productive performance (Harms et al., 2000). In laying hens, Grobas et al. (1999c) have reported that increasing the AMEn content of the diet from 2,680 to 2,810 kcal/kg a (4.8% of increase) decreased feed intake by the same proportion a (5.0% of decrease) but egg production and egg mass were not affected. Similarly, Peguri et al. (1991) reported a 5% of decrease in FI but similar egg production when AMEn of the diet was increased from 2,700 to 2,910 kcal/kg a (8% of increase). In contrast, Joly and Bougon (1997) reported a 1.3% of increase in egg production and 4.5% of increase in egg mass as the energy content of the diet increased from 2,200 to 2,700 kcal AMEn/kg in brown laying hens diets from 19 to 68 wk of age.

Most of published trials which study the effect of energy level of diet reported an improvement in egg weight (**EW**) with increasing levels of energy (De Groote, 1972; Walker et al., 1991). The hens tend to maintain its energy intake modifying the FI (Leeson et al., 1973; Newcombe and Summers, 1985), over consuming energy in high energy diets (De Groote, 1972; Walker et al., 1991). Thus, the excess of nutrients improve the EW (De Groote, 1972; Walker y col., 1991). According to these authors, EW improved from 0.10 to 0.20% per each 100 kcal. Bouvarel et al. (2010) analyzed data from 11 experiments conducted for the last 20 years and reported that EW increased 0.96 g per each 100 kcal of increase in dietary AMEn. The reasons for the discrepancies among authors in relation to the effects of energy level on EW are not apparent but might be related with the level of fat and the linoleic acid (**LNL**) content of diets.

The effects of energy level on egg production are variable. Thus, while Mathlouthi et al. (2002) reported with Single Comb White Leghorn (**SCWL**) hens that egg production increased as the AMEn of diet increased from 2,650 to 2,750 kcal/kg, Grobas et al., (1999c) with brown hens fed diets varying from 2,680 to 2,810 kcal AMEn/kg, Harms et al. (2000) with brown and SCWL hens fed diets varying in AMEn from 2,500 to 3,100 kcal/kg , and Jalal et al.(2006, 2007) in SCWL hens fed diets varying from 2,800 to 2,900 kcal AMEn/kg did not detect any significant difference in egg production. In commercial flocks, increasing the energy content of diet at the onset of laying period is quite common, especially when the pullets have not an homogeneous BW or when the pullets have a low BW at the beginning of laying period. Thus, some authors have reported that in places with hot climate like Spain, the increase of energy concentration of the diet can improve the performance especially in light hens (Kling and Hawes, 1990; Daghir, 1995).

2.3.1.2. Effects on egg quality

The reasons for the discrepancies among authors in respect to the variation in egg quality results with AMEn of diet are not apparent, but might be related to the different use of basal diets and fats. Grobas et al. (1999a) reported that the increase in energy concentration of the diet did not affect the percentage of dirty, broken, or shell less throughout the laying period. Zimmermann and Andrews (1987) and Junqueira et al.

(2006) reported that the increase in energy concentration of the diet did not affect the haugh units (**HU**). However, Wu et al. (2005) reported a decrease in HU when the AMEn of the diets was increased from 2,720 to 2,960 kcal/kg. On the other hand, Gunawardana et al. (2008) reported higher yolk pigmentation in SCWL hens fed a diet with 5.0% added fat than in hens fed a control diet without any added fat. Also, Lázaro et al. (2003) reported higher yolk pigmentation in SCWL hens fed high AMEn diets. These results can be related to a better absorption of Xanthophylls, the main pigment source responsible for egg yolk color, which are soluble in fat. Also, when fat is used to increase the energy concentration of the diet, the proportion of shell in the egg could be affected due to fat can modify calcium absorption, basically in broilers. However, this effect is not clear in pullets or laying hens. Junqueira et al. (2006) reported a linear decrease in egg shell proportion as the AMEn increased from 2,850 to 3,050 kcal/kg in brown egg-laying hens from 76 to 84 wk of age. However, Gunawardana et al (2008) did not find any effect of energy content of the diet on egg shell proportion in SCWL fed diets varying in AMEn content from 2,750 to 3,050 kcal/kg.

2.3.2. Fat content of laying hen diets

2.3.2.1. Effects on productive performance

Fats are used in poultry feeds to increase the energy content of diets. Fat inclusion usually results in higher energy intake, BW gain and EW (Grobas et al., 2001; Bouvarel et al., 2010), probably due a better palatability with less dust formation (ISA Brown, 2011). Also, supplemental fat has been shown to reduce rate of feed passage, facilitating the contact between digesta and enzymes, improving digestibility and utilization of other nutrients such as the lipid and carbohydrate fractions of dietary ingredients (Mateos and Sell, 1980). Whitehead et al. (1993) studied the effect of supplemented fat on EW and concluded that eggs produced with maize oil were heavier than those produced with others sources of fat, such as fish oil (long chain polyunsaturated fatty acids (**FA**), coconut oil (shorter chain saturated FA) or tallow (medium to long chain length saturated FA). Probably, readily absorbable unsaturated FA of corn oil improves the EW. Grobas et al. (2001) studied the effect of 4 different sources of supplemented fat on EW and reported that eggs were heavier when hens were fed diets supplemented with soy oil than when supplemented with linseed oil, olive oil, or tallow. Atteh and Leeson (1983, 1984,1985) studied the effect of FA profile on performance and mineral metabolism of laying hens and broilers, and reported that fat and some minerals like calcium can interfere together, leading to the formation of insoluble soaps responsible of the decrease in absorption of both, FA and minerals. Furthermore, they reported that soap formation was higher with saturated (palmitic and stearic acids) that with unsaturated FA and an increase in the Ca content of the diet increased the soap formation.

Many studies have shown that a reduction in supplemental fat (**SFAT**) decreases egg size (Keshavarz and Nakajima, 1995; Grobas et al., 1999a,b; Sohail et al., 2003). Grobas et al. (2001) reported that SFAT improved EW and egg mass output in both, SCWL hens and brown laying hens throughout the production cycle. Grobas et al. (1999b) compared isonutritive diets for brown laying hens differing in fat content (0 and 4 %) from 22 to 65 wk of age and observed that SFAT improved productive performance and egg size, although FCR was not affected. In this research, the improvement in egg rate observed occurred from 38 to 61 wk of age, whereas the beneficial effects on EW were more noticeable from 22 to 57 wk of age. Whitehead (1981) showed that supplementation of diets with 0.4 or 3 % of fat significantly increased EW. Whitehead et al. (1993) compared 5 inclusion levels of fat (0, 1, 2, 4, and

6 %) and concluded that, with the exception of fish oil, which hindered productive performance when included at 2 %, maize oil, tallow, and coconut oil perform well till 4 % of inclusion. Furthermore, Grobas et al. (1999b) showed that supplementation of the diet with 4 % increased EW as compared with a non-supplemented diet. However, these authors showed that further increases from 5 to 10% of fat supplementation did not have any positive effect on EW (Grobas et al., 2001).

2.3.2.2. Effects on egg quality

Added fat increased, both yolk and albumen weights, but in some studies the improvement was proportionally greater for the albumen than for the yolk (Grobas et al., 1999b). Whitehead (1995) hypothesized that the beneficial effect of SFAT on albumen weight was due to the influence of certain unsaturated FA on the production of oestrogens which are the main responsible for albumen secretion Regarding egg quality traits, Grobas et al. (1999a) observed that the increase in EW with SFAT was accompanied by a similar increase (3.5%) in yolk and albumen weights. The mechanism by which SFAT increases egg size is uncertain. Whitehead et al. (1991) suggested that SFAT increased yolk weight by stimulating lipid deposition, and albumen weight by stimulating oestrogen secretion, which controls protein synthesis in the oviduct. Parsons et al. (1993) reported that a reduction in SFAT from 6 to 2% of the diet reduced the proportion of large and above eggs in SCWL. Same results have been reported by Bohnsack et al. (2002) with similar type of diets. Haugh units were not affected by SFAT (Grobas et al., 2001; Usayran et al., 2001). Previous research has shown that SFAT exerts a favorable effect on EW beyond that attributable to an increase in LNL content of the diet (Sell et al., 1987; Grobas et al., 1999a).

3. Objectives

The general aim of this Master Thesis was to study the influence of nutritional factors during the rearing and laying phases that might affect the productivity and egg quality of commercial brown egg-laying hens. With this objective, six experimental diets for the rearing phase varying in the main cereal (Barley or corn) and with different level of inclusion of two fiber sources straw and sugar beet pulp, and two diets with different level of energy for the laying phase diets to study:

-The effects of diets composition during the rearing and laying phases in eggs quality during the laying phase

-The effects of diets composition during the rearing and laying phases of the development of the GIT of the hens at 48 wk of age

- The effects of inclusion of different sources of fiber in the rearing diets and its effect on productive performance during the laying cycle.

-Potential interaction between the characteristics of the rearing phase diet and the laying hen diet on performance of the hens during the laying cycle.

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CHAPTER 3: THE EXPERIMENT

EFFECT OF FIBER INCLUSION IN THE REARING DIETS AND ENERGY CONCENTRATION OF THE LAYING DIETS ON PRODUCTIVE PERFORMANCE AND EGG QUALITY OF BROWN EGG-LAYING HENS FROM 18 TO 46 WEEKS OF AGE.

1. Introduction

Egg production and egg quality depends on multiple factors, including nutrition during the rearing and laying phases. Grains are the main ingredients of poultry feeds and birds are well equipped to utilize diets with high starch contents (Hetland et al., 2003). Most research conducted with poultry have shown similar productive performance in hens fed diets based on corn or barley, provided that the diets were supplemented with an enzyme complex with adequate β-glucanase and xylanase activity (Lázaro et al. 2003; Safaa et al, 2009; Pérez Bonilla et al, 2011). Fiber is an important component of poultry diets but fiber cannot be digested by the bird, although it might affect gastrointestinal tract (**GIT**) development and gizzard function (González-Alvarado et al., 2010). Traditionally, dietary fiber has been considered as a diluent of the nutrient content of the feed with negative impact on palatability and nutrient digestibility (Mateos et al., 2002, 2012). The increased availability of vegetable sources with high fiber content, such as DDGS from the ethanol industry, has increased the interest to study the effects of dietary fiber on poultry performance. Recent studies have shown that the inclusion of adequate amounts of certain types of fiber in the feed improves the adaptation of the GIT to existing production systems and might reduce digestive disorders in poultry, improving bird performance (Jiménez-Moreno et al., 2009, 2013 a, b)

It is a common commercial practice to increase the fiber content of the feeds during the last part of the rearing phase to enhance the ability of the pullet to increase voluntary feed intake at the start of the laying period. However, the scientific information available on the benefit of this practice is very limited. The hypothesis of this study was that changes in the development of the GIT during the rearing phase, a consequence of including fibrous ingredients in the diet, could improve subsequent hen production and that the benefits could be more pronounced in hens fed during the laying phase low energy diets than in hens fed high energy diets.

2. Materials and Methods

2. 1. Husbandry, Diets, and Experimental Design

All experimental procedures were approved by the Animal Ethics Committee of the Universidad Politécnica de Madrid and were in compliance with the Spanish guidelines for the care and use of animals in research (Boletín Oficial del Estado, 2013).

In total, 480 Lohmann Brown egg-laying pullets were fed during the 3 periods (1 to 5 wk, 5 to 10 wk, and 10 to 17 wk of age) of the rearing phase one of 6 diets that consisted in 2 isonutritve diets based on barley or corn, and 4 extra diets that included 2 or 4% of either cereal straw or sugar beet pulp at the expense of the corn diet. The ingredient composition and nutritive value of the diets and the management of the pullets during the rearing phase have been reported elsewhere (Guzman et al., 2013). At 17 wk of age, the pullets were moved from the rearing house to the laying hen facility where they were housed in groups of 10 in enriched cages (0,64 m × 1,2 m; Facco S.A., Padova, Italy) equipped with an open trough feeder and 2 nipple drinkers. At arrival to the laying hen facility, hens received a common diet that included an antibiotic (Tylosin, 200 ppm/Elanco valquimica, Madrid, Spain) for 7 d and then, their respective experimental laying diets to 46 wk of age. Half of the pullets of each previous rearing group received a low (LE; 2,650 kcal AME_n/kg) or a high (HE; 2,750 kcal AME_n/kg) energy diet. The two laying diets had similar nutrient content per unit of energy and therefore, the AME_n: indispensable AA ratio did not vary between the 2 diets (Table 1). Diets were formulated according to Fundacion Española Desarrollo Nutrición Animal (2010) and met or exceeded the nutrient requirements for brown-egg laying hens according to Fundacion Española Desarrollo Nutrición Animal (2008). No commingling was practiced and therefore, all the hens within each laying cage belonged to the same pullet replicate. Pullets and hens were vaccinated and managed according to commercial practices (Lohmann, 2012). The light program during the laying phase consisted of 16 h of continuous light per day throughout the experiment. The temperatures inside the barn were recorded daily an varied from 28 ±3ºC in June (first period of the experiment) to 19 ± 3ºC in January (last period of the experiment).

The experimental design was completely randomized with 12 treatments arranged as a 6x2 factorial with 6 diets used during the rearing phase and 2 diets used during the laying phase as main effects. Each of the 12 treatments was replicated 4 times and the experimental unit during the laying period was an enriched cage with 10 hens.

2. 2. Laboratory Analysis

The nutritive values of the diets used during the rearing phase have been reported elsewhere (Guzmán et al., 2013). Representative samples of the laying phase diets were ground in a laboratory mill (Model Z-I, Retsch Stuttgart, Germany) equipped with a 0.5-mm screen and analyzed for moisture by oven-drying (method 930.01), total ash using a muffle furnace (method 942.05), and nitrogen by combustion (method 990.03) using a LECO analyzer (Model FP-528, LECO, St. Joseph, MI), as described by AOAC International (2000). Ether extract was determined by Soxhlet analysis after 3N HCl acid hydrolysis (Boletín Oficial del Estado, 1995) and gross energy in an isoperibol bomb calorimeter (Model 356, Parr Instrument Company, Moline, IL). The amino acids (**AA**) content of the diets were determined by chromatography (Hewlett-Packard 1100, Waldbronn, Germany) as described by De Coca-Sinova et al (2008). The geometric mean diameter (**GMD**) of the diets was determined in triplicate in 100 g samples using a Retsch shaker (Retsch, Stuttgart, Germany) equipped with 8 sieves ranging in mesh from 5,000 to 40 µm according to the methodology outlined by the ASAE (1995). The determined analyses and the GMD of the diets are shown in (Table 2).

2. 3. Productive Performance and Egg Quality

Feed intake and egg production were recorded by replicate for each of the seven 28 d periods and for the entire experimental period (18 to 46 wk of age). All the eggs produced the last 2 days of each week were weighed by replicate and the average value of the 4 wk was used to estimate average egg weight for each of the 7 experimental periods. In addition, hens were weighed by replicate at the start of the experiment and at the end of each of the 7 experimental periods. Mortality was recorded and weighed as produced. From these data, egg production, average daily feed intake (**ADFI**), egg mass, feed conversion ratio (**FCR**) per kilogram of eggs and per dozen of eggs, and BW gain were calculated by period and cumulatively. In addition, the number of dirty, broken, and shell-less eggs was recorded daily by replicate. An egg was considered as dirty when a spot of any kind or size was detected on the shell (Lázaro et al., 2003). Egg quality, including Haugh units, yolk color, shell strength, shell thickness, and shell color were measured in 6 eggs collected randomly from each replicate the last day of each of the 7 experimental periods. Haugh units and yolk color (Roche color fan) were measured using a Multitester equipment (QCM System, Technical Services and Supplies, Dunnington,York, UK) as indicated by Pérez Bonilla et al., (2012). Shell color was measured using a Minolta colorimeter (Chroma Meter Model CR-200, Minolta Corp., Ramsey, NJ) and the Hunter color values, L* (lightness), a* (green to red), and b* (blue to yellow) values were recorded (Hunter and Harold, 1987). Egg shell strength was evaluated applying increased pressure to the broad pole of the egg using a press meter, and expressed in kg/cm^2. Shell thickness was measured at the two pole ends and at the middle section of the egg using a digital micrometer (QCM System) and the average of the 3 measurements of each of the 6 eggs was used for further analyses.

2. 4. Digestive Traits

At 46 wk of age, after the corresponding productive performance control, 2 birds per replicate were randomly selected, weighed individually, and euthanized by CO_2 inhalation. The digestive tract (from the end of the crop to the cloaca, including the digestive contents, spleen, liver, and pancreas) was removed aseptically and weighed. Then, the liver, and the full proventriculus and full gizzard were excised and weighed. Data on organ weight were expressed relative (%) to BW. Then, the gizzard was emptied from any digesta, cleaned, dried with desiccant paper, and weighed again. The fresh digesta content of the gizzard was measured as the difference between the full and the empty organ weight and expressed relative to the weight (%) of the full organ. In addition, the pH of the gizzard digesta was measured in these hens using a digital pH meter fitted with a fine tip glass electrode (model 507, Crison Instruments S.A., Barcelona, Spain) as indicated by Jiménez-Moreno et al. (2009) (Table5). No attempt was made to measure the fresh digesta content of the proventriculus. The length of the hens, measured from the tip of the beak to the end of the longest phalanx in extended birds, and the length of the duodenum (from gizzard to pancreo-biliary ducts), jejunum (from pancreo-biliary ducts to Meckel's diverticulum), ileum (from Meckel's diverticulum to ileo-cecal junction), and the two ceca (from the ostium to the tip of the right and left ceca) were measured on a glass surface using a flexible tape with a precision of 1 mm (Table 6). Tarsus length and tarsus diameter (measured in the middle point of the bone) were also determined using a digital caliper. All these measurements were expressed in cm/kg BW. All traits were measured in duplicate and the average value of the two measurements was used for further statistical analysis.

2. 5. Statistical Analysis

Data were analyzed as a completely randomized design with 12 treatments arranged as a 6x2 factorial with 6 rearing phase diets and 2 laying phase diets. Main effects and their interaction were analysed by ANOVA using the GLM procedure of SAS Institute (1990). Four of the rearing diets formed a 2x2 factorial (2 sources of fibre at 2 levels of inclusion) and therefore, the main effects of source and level of fiber of the rearing diets on laying hen performance were also studied. When the model was significant, treatment means were separated using the Tukey test. Differences between treatment means were considered significant at P <0.05. In addition, the linearity of straw and SBP inclusion in the diet and the following contrasts (0 *vs*. 2 and 4 % and 2 *vs*. 4% fiber inclusion) were also studied. Results in tables are presented as means and differences were considered significant at P < 0.05.

3. Results

3. 1. Productive Performance and Egg Quality Traits

No interactions between type of diet fed during the rearing phase and the laying phase were detected and therefore, only main effects are presented. Neither the main cereal nor fiber inclusion in the diet affect any of the productive performance variables studied during the laying phase with the exception of egg production that was reduced (P<0.05) when diets containing SBP were used (Table 3). An increase in the energy content of the laying diet reduced ADFI (114.7 vs. 111.3 g/d; P<0001), improved FCR per kilogram (2.02 vs. 1.97; P < 0.01) and per dozen (1.49 vs. 1.46 ; P < 0.01) of eggs and increased BW gain (375 vs. 339 g; P<0.05). However, egg production, egg weight, and egg mass were not affected by energy content of the laying diets. Data on the effects of fiber source, in the rearing phase diets and energy concentration of the laying phase diets on hen production by period are shown in Figures 1 to 3. It can be observed that most of the negative effects of the inclusion of SBP in the rearing diet as compared with the inclusion of the straw were detected at the end of the laying phase (Figure 1).

Egg quality was not affected by fiber inclusion in the rearing phase diets (Table 4). Similarly, energy content of the laying phase diets did not affect egg quality, except for the percentage of dirty eggs that was higher (2.5 vs. 1.7 %; P<0.001) in eggs from hens fed the LE diets than in eggs from hens fed the HE diet.

3. 2. Digestive Traits

At 46 wk of age, the relative weight of the digestive organs (GIT, liver, proventriculus, and gizzard) and the weight, digestive content, and pH of the gizzard were not affected by dietary treatment (Table 5). Similarly, the length of the hens and of the tarsus and the length of the small intestine and cecum were not affected by dietary treatment (Table 6).

4. Discussion

4. 1. Productive Performance and Egg Quality

The inclusion of straw in the pullet phase diet had not effect on the posterior productive performance of the hens during the laying phase. However, SBP inclusion reduced egg production without affecting any other variable. Consequently, as compared with hens fed the control die, energy efficiency per kilogram of eggs was improved by in hen fed straw but hindered in hen fed SBP. The data suggest that, opposite to the general believe, the inclusion of a fiber source during the last period of the rearing phase does not affect voluntary feed intake at the onset of egg production. It seems that pullets fed high fiber diet adapt quickly the size of the GIT to changes in diets composition, supporting the suggestion of Mateos et al (2012) that birds respond quickly to changes in fiber content of the diet by modifying the length and weight of the different organs of the GIT. In fact, Ramzi et al., (2013) reported no differences in the relative weight of the GIT and the gizzard or in the relative length or the small intestine in 17 wk old pullets that were fed mush continuously or crumbles followed by mush from 0 to 5 wk or 0 to 10 wk of age. Many factors such as type (viscous vs. non-viscous) and level of fiber, and age of the birds, as well as the composition of the basal diet, likely influence the response of the GIT and the epithelial mucosa to dietery fiber (Iji et al., 2001; Jimenez-Moreno et al., 2013b). The inclusion of SBP in the rearing diets hindered egg production during the laying phase, an effect that was consistent with the reduction in feed intake observed. Pettresson and Razdan al (1993) also reported that the high water holding capacity (**WHC**) of the pectins present in the SBP could increase digesta bulk and reduce feed intake in broilers. In broilers, González-Alvarado et al.(2010) reported a reduction in FI in broilers from 1 to 42 d of age when the oat hulls (**OH**) of the diets was substituted by SBP. In this research, FI during the laying phase was 1.5 % lower in hens fed SBP during the rearing period than in hens that were fed the control diet although the difference was not significant.

In the current research, a 4.0 % increase in the energy content of the laying diet (from 2,650 to 2,750 kcal AMEn/kg) decreased FI by only 3.05%. Hens fed the LE diets try to compensate for the lower energy by increasing FI proportionally, in agreement with previous research (Hill et al., 1956; Jackson and Waldorup., 1988; Bouvarel et al.. 2010). Pérez-Bonilla et al. (2012) reported that an 11% increase in the energy content of the diet (from 2,650 to 2,950 kcal AMEn/kg) decreased FI by only 4%.

Egg production was not affected by the energy concentration of the diet, in agreement with data of Grobas et al (1999c) in brown egg-laying hens fed isonutritive diets with 2,680 or 2,810 kcal AMEn/kg. In contrast, Mathlouthi et al. (2002) reported in SCWL hens that egg production increased as the AMEn of the diet increased from 2,650 to 2,750 kcal/kg. Also, Pérez-Bonilla et al. (2012) observed that egg production increased in brown egg-laying hens as the AMEn concentration of the diet increased from 2,650 to 2,850 kcal/kg in Lohman brown hens.

Egg weight was not affected by energy concentration of the diet, consistent with data of (Grobas et al., 1999b; Ciftci et al.,2003; Pérez-Bonilla et al.,2012). However, Harms et al. (2000) and Wu et al. (2005, 2007) reported that egg weight increased linearly with increases in dietary energy. Similar results en egg weight have been reported by Bray (1967) and DeGroote (1972). The reasons for the inconsistencies among authors in respect to the effects of energy content of the diet on egg weight are not known, but might be related to the level of linoleic acid (**LNL**) and supplemental fat of the diets (Safaa et al., 2008). When the energy content of the diets increases, the level of supplemental fat and of LNL increases which in turn might result in an increase in egg weight (Grobas et al., 1999a,c). Therefore, egg weight response to increases in energy content of the diet will depend also on changes in LNL and fat content of the diets (Pérez-Bonilla et al 2011). In the current research, the two diets had a LNL content above requirements (Shannon and Whitehead., 1974) and the fat content was similar. Therefore, no difference in egg weight was expected.

Egg mass did not change when the AMEn of the diet increased from 2,650 to 2,750 kcal/kg, consistent with data of Wu et al. (2005) who did not detect any effect on egg mass with increases in the AME of the diet from 2,720 to 2,960 kcal/kg in SCWL hens from 21 to 36 wk of age. Pérez Bonilla (2012) reported that egg mass increased as the AMEn of the diet increased from 2,650 to 2,750 kcal/kg but that a further increase to 2,850 or 2,950 kcal/kg did not result in any further improvement.

Feed conversion ratio improved as the energy content of the diet increased, in agreement with most published reports (Grobas et al., 1999b; Wu et al., 2005). In contrast, Keshavarz (1998) reported no differences in feed efficiency in SCWL hens from 18 to 66 wk of age fed diets with 2,820 or 3,040 kcal AMEn/kg. Hens eat to satisfy their energy requirements and there FCR should be improve with increases in energy concentration of diets

BWG increased 0.20 g/hen per day per each100 kcal increase in AMEn concentration of the diet, a value similar to that reported by Grobas et al. (1999c) in brown laying-hens from 22 to 65 wk of age fed diets with 2,680 or 2,810 kcal AMEn/kg, and higher of than 0.11 g/d reported by Pérez-Bonilla et al. (2012) in brown egg-laying hens from 24 to 59 wk of age fed diets with 2,650 to 2,950 kcal AMEn/kg. The value, however, is lower than 0.45 g/d reported by Harms et al. (2000) in SCWL from 36 to 44 wk of age fed diets with 2,520 to 3,080 kcal AMEn/kg. Modern brown egg-laying hens might respond to increases in energy content of the diets with moderate increases in BW gain. On the other hand, hens fed the HE diet had 3.0% lower FI but 0.7% higher energy intake than hens fed the LE diet. The 0.7% increases in energy intake observed when the HE diet was used resulted in an increase of 9.6 % of the BWG of the hens rather than in an increase in egg mass production. This finding is consistent with data of Pérez-Bonilla et al. (2012) who reported that hens fed a diet with 2,950 kcal AMEn/kg had 29.0 % BWG extra over hens fed a diet with 2,650 kcal AMEn/kg.

Energy concentration of the diet did not affect the percentage of broken or shell less eggs of the hens, consistent with data of Pérez-Bonilla et al. (2012) and Grobas et al. (1999a). However, the percentage of dirty eggs was reduced with increases in energy concentration of the diet. The ingredient composition and the amount of Neutral detergent fiber (**NDF**) was similar for both diets, and therefore, We do not have any explanation for this finding. HU was not affected by AMEn concentration of the diets, results that agree with data of Junqueira et al. (2006). In contrast, Pérez-Bonilla et al. (2012) reported a linear decrease in albumen quality with increases in energy concentration of the diet. On the other hand, Wu et al. (2005) reported a decrease in HU when the AMEn of the diets was increased from 2,720 to 2,960 kcal/kg. The authors suggested that the decrease in HU observed was possibly due to the lower AA intake of the hens fed the high energy diets. However, in the current research, the amino acids (**AA**) intake was similar for the 2 groups of hens, and therefore, no differences in albumen quality should be expected.

Yolk pigmentation did not change with increases in energy concentration of the diet. In contrast, Pérez-Bonilla et al. (2012) observed that yolk pigmentation increased linearly with increases in energy concentration of the diet. Xanthophylls are the main pigment source responsible for egg yolk color (Bouvarel et al., 2010). Mateos and Sell (1980) reported that the inclusion of fat in the diet reduced rate of feed passage facilitating the contact between nutrients and digestive enzymes. Consequently, supplemental fat might improve the utilization of other dietary components of the diet.

Shell strength and shell thickness were not affected by energy concentration of the diet, data that agree with results of Safaa et al. (2008) who reported similar egg shell quality in late phase of production in hens fed diets with 1.1 or 3.0% supplemental fat.

4. 2. Digestive Traits

Dietary fiber affects the anatomy and the development of the GIT (Mateos et al., 2012), increases the retention time of digesta in the proximal part of the GIT, and increases gizzard size and function (Hetland et al., 2005; Jimenez-Moreno et al., 2013b) and the length of the digestive organs (Van der Klis and van Voorst, 1993; Iji et al., 2001). In the current research, digestive tract traits, including relative weight or length of the GIT at 46 wk of age did not change when the AMEn of diet increased from 2,650 to 2,750 kcal/kg. The authors have not found any report in the literature comparing the effects of energy concentration of the diet on the size and the length of the different organs of the GIT to compare with the results of current experiment. Probably, a change in the energy level of the diet of 100 kcal AMEn/kg is not an important factor that could affect GIT traits. Frikha et al. (2009, 2011) and González-Alvarado et al. (2007, 2008) have shown that other factors such as feed form, main cereal of the diet of egg-laying pullets, and inclusion of fiber in the diet might affect GIT development in poultry.

5. Conclusion

Type of cereal or the inclusion of 2 to 4% of a fiber source in the rearing diets resulted in similar feed intake and hen performance during the laying phase. No interactions between the characteristics of the rearing diets and the energy content of the laying diets were detected for any laying trait. Therefore, the effects on an increase in the level of fiber in the rearing diets on the development and function of the gastrointestinal tract disappear with time and have no effects on subsequent hen performance. An increase in the energy content of the laying diets reduces feed intake, improves feed efficiency, and increases BW gain but did not affect any of the other production variables studied.

Table 1. Ingredient composition and calculated analysis (% as fed bases, unless otherwise indicated) of the experimental diets.

	AMEn (kcal/kg)	
	Low[3]	**High**[4]
Ingredient		
Wheat	40.0	40.0
Corn	20.0	17.0
Soybean meal, 47% CP	17.1	18.0
Sunflower meal, 34% CP	10.3	11.0
Sunflower oil soapstocks	1.45	3.39
Dicalcium phosphate	1.08	1.18
Calcium carbonate	8.24	8.50
Sodium chloride	0.28	0.30
DL-methionine, 99%	0.11	0.13
Sepiolite	0.94	-
Vitamin and mineral premix[1]	0.50	0.50
Calculated analysis[2]		
Dry matter	89.4	89.8
EMAn, Kcal/kg	2,650	2,750
Crude protein	17.6	18.2
Linoleic acid	1.7	2.8
Crude fiber	4.4	4.6
Neutral detergent fiber	11.1	11.4
Digestible amino acids		
Arg	1.05	1.09
Ile	0.63	0.65
Lys	0.68	0.71
Met	0.37	0.40
Met+cys	0.63	0.66
Thr	0.53	0.55
Trp	0.18	0.19
Val	0.72	0.75
Total ash	12.4	12.9
Ca	3.59	3.72
Total phosphorus	0.61	0.64
Digestible phosphorus	0.29	0.31

[1] Provided the following (per kilogram of diet): vitamin A (trans-retinyl acetate), 6,000 IU; vitamin D3 (cholecalciferol), 1,200 IU; vitamin E (all-rac-tocopherol-acetate), 5 mg; vitamin K3 (bisulphate menadione complex), 1.5 mg; riboflavin, 3.5 mg; betaine, 67.5 mg; thiamin (thiamine-mononitrate), 1 mg; vitamin B12 (cyanocobalamin), 15 µg; Se (Na2SeO3), 0.1 mg; I (KI), 1.9 mg; Cu (CuSO4・5H2O), 4 mg; Fe (FeCO3), 18 mg; Mn (MnO), 66 mg; and Zn (ZnO),

[2] According to Fundación Española Desarrollo Nutrición Animal (2010).

[3] Low energy: 2,650 kcal AMEn/kg

[4] High energy: 2,750 kcal AMEn/kg

Table 2. Determined analysis[1]and particle size distribution[2] of the experimental laying phase diets (% as fed basis, unless otherwise indicated)

	AMEn (kcalAMEn/kg)	
	Low[3]	High[4]
Composition		
Gross energy, kcal/kg	3,848	3,872
Dry matter	94.2	93.8
Ether extract	5.0	5.2
CP	17.5	18.5
Arg	1.16	1.18
Ile	0.72	0.73
Lys	0.80	0.81
Met	0.41	0.43
Met+cys	0.72	0.74
Thr	0.63	0.64
Trp	0.22	0.22
Val	0.83	0.85
Total ash	13.7	14.2
Particle size		
>2,500	11.80	12.77
1,250	36.31	36.93
630	29.21	28.99
315	17.91	15.11
160	4.77	6.18
<80	0.01	0.03
GMD[5] ± GSD[6]	1,112± 2.06	1,132± 2.10

[1] Analyzed in triplicate

[2] Sieve diameter, µm.The percentage of particles smaller than 40 µm and bigger than 2500 µmwere negligible for all diets.

[3] Low energy: 2,650 kcal AMEn/kg

[4] High energy: 2,750 kcal AMEn/kg

[5] Geometric mean diameter

[6] GSD = Log normal SD.

Table 3. Influence of source and level of fiber of the rearing phase diets and AME$_n$ of the laying phase diets on productive performance of the hens (18 to 46wk of age)

Rearing phase			Laying phase	Egg production (%)	Egg weight (g)	Egg mass (g/d)	Feed intake (g/d)	FCR[3] (kg/kg)	FCR (kg/dozen)	BW gain (g)
Cereal	Fiber source	Inclusion level, %	AME, kcal/kg							
Barley		0	2,650	94.5	60.7	57.4	114.9	2.04	1.48	335
			2,750	94.0	61.3	57.7	111.4	1.97	1.44	354
Corn		0	2,650	94.0	62.4	58.7	115.8	2.01	1.50	371
			2,750	93.7	62.0	58.2	111.8	1.96	1.45	381
	Straw	2	2,650	95.1	61.0	58.1	114.5	2.00	1.47	322
			2,750	93.4	62.3	58.3	111.6	1.95	1.46	378
		4	2,650	93.8	62.1	58.4	114.8	2.00	1.49	327
			2,750	94.1	60.9	57.4	112.6	2.01	1.46	392
	SBP[1]	2	2,650	90.7	62.7	56.9	113.4	2.04	1.53	337
			2,750	92.7	62.7	58.2	110.5	1.94	1.45	336
		4	2,650	94.0	61.1	57.5	114.8	2.03	1.49	342
			2,750	90.1	62.1	56.1	109.6	2.00	1.49	407
Main effects										
Rearing phase diets										
Fiber source										
	Straw			94.1^{a}	61.6	58.0	113.4	1.99	1.47	355
	SBP			91.9^{b}	62.2	57.2	112.1	2.00	1.49	356
Inclusion level, %										
	0			93.9	62.2	58.4	113.8	1.98	1.48	376
	2			93.0	62.2	57.9	112.5	1.98	1.48	343
	4			93.0	61.5	57.3	113.0	2.01	1.48	367
Laying phase diets, AMEn/kg										
	2,650 kcal			93.7	61.7	57.8	114.7^{a}	2.02	1.49^{a}	339^{b}
	2,750 kcal			93.0	61.9	57.6	111.3^{b}	1.97	1.46^{b}	375^{a}
SEM[2]				1.20	0.56	0.83	1.29	0.027	0.021	25.3
							Probability			
General model[4]				0.160	0.118	0.653	0.024	0.102	0.239	0.302
Main effects										
Fiber source during rearing phase				0.030	0.223	0.186	0.240	0.660	0.201	0.964
Fiber level during rearing phase				0.734	0.266	0.320	0.619	0.270	0.943	0.259
Energy level during laying phase				0.369	0.512	0.713	<.0001	0.002	0.008	0.016
Contrast										
Straw ,0 vs 2 and 4%				0.814	0.223	0.565	0.757	0.683	0.644	0.353
Straw, 2 vs 4%				0.832	0.756	0.706	0.666	0.344	0.553	0.725
SBP, 0 vs 2 and 4%				0.068	0.922	0.077	0.206	0.433	0.505	0.374
SBP, 2 vs 4%				0.719	0.255	0.673	0.843	0.936	0.612	0.435
Straw linear				0.921	0.220	0.505	0.951	0.361	0.913	0.513
SBP linear				0.142	0.289	0.053	0.228	0.184	0.584	0.964

[1] Sugar Beet palp

[2] Standard error of the mean (4 replicates of 10 hens each per treatment)

[3] Feed conversion ratio

[4] All of the interactions studied between level and source of fiber, and energy level were not significant (p>0.10)

[a, b] Means with different superscripts are significantly different

Table 4. Influence of source and level of fiber of the rearing phase diets and AME_n of the laying phase on egg quality from 18 to 46 wk of age

Rearing phase			Laying phase								Shell color		
Cereal	**Fiber source**	**Inclusion level, %**	**AME, kcal/kg**	**Dirty eggs %**	**Broken eggs %**	**Shell-less eggs %**	**Haugh Units**	**RCF[1]**	**Shell strength kg/cm^2**	**Shell thickness mm**	**L[2]**	**a[3]**	**b[4]**
Barley		0	2,650	2.2	0.65	0.10	91.4	10.2	5.2	0.381	62.1	15.2	32.4
			2,750	1.6	0.67	0.34	89.9	10.2	5.0	0.390	62.0	15.1	32.5
Corn		0	2,650	2.7	0.64	0.32	90.2	10.3	5.1	0.375	62.4	15.0	32.3
			2,750	1.8	0.66	0.22	91.3	10.2	5.0	0.381	61.7	15.2	31.9
	Straw	2	2,650	2.4	0.47	0.24	91.1	10.2	5.0	0.384	60.4	16.3	33.0
			2,750	1.6	0.89	0.20	91.0	10.1	5.1	0.385	61.9	15.1	32.1
		4	2,650	2.8	0.44	0.18	90.9	10.3	5.2	0.384	61.3	15.4	32.2
			2,750	1.7	0.73	0.20	90.7	10.1	5.2	0.378	61.9	15.1	32.3
	SBP	2	2,650	2.2	0.83	0.33	91.3	10.4	4.9	0.380	60.9	15.7	32.4
			2,750	2.0	0.33	0.13	90.7	10.2	5.2	0.384	61.6	15.3	32.0
		4	2,650	2.4	0.32	0.19	90.3	10.3	5.3	0.384	61.4	15.4	32.4
			2,750	1.3	0.43	0.28	90.7	10.3	5.2	0.383	61.0	15.8	32.2
Main effects													
Rearing phase diets													
Fiber source													
	Straw			2.1	0.63	0.21	90.9	10.2	5.1	0.383	61.4	15.5	32.4
	SBP			2.0	0.48	0.24	90.8	10.3	5.1	0.383	61.2	15.5	32.2
Inclusion level, %													
	0			2.3	0.65	0.27	90.8	10.2	5.1	0.378	62.05	15.1	32.1
	2			2.0	0.63	0.23	91.0	10.2	5.0	0.383	61.2	15.6	32.4
	4			2.0	0.48	0.21	90.7	10.2	5.2	0.382	61.4	15.4	32.3
Laying phase diets, AMEn/kg													
	2,650 kcal			2.5^{a}	0.56	0.23	90.9	10.3	5.1	0.381	61.4	15.5	32.5
	2,750 kcal			1.7^{b}	0.62	0.23	90.7	10.2	5.1	0.383	61.7	15.3	32.2
SEM[5]				0.40	0.142	0.080	0.70	0.14	0.20	0.005	0.53	0.36	0.34
								Probability					
General model[6]				0.188	0.108	0.494	0.927	0.952	0.940	0.761	0.361	0.392	0.706
Main effect													
Fiber source during rearing phase				0.596	0.217	0.597	0.744	0.232	0.891	0.986	0.712	0.743	0.442
Fiber level during rearing phase				0.855	0.328	0.692	0.717	0.996	0.365	0.361	0.197	0.327	0.593
Energy level during laying phase				<0.001	0.508	0.996	0.692	0.334	0.916	0.454	0.402	0.295	0.137
Contrast													
Straw, 0 vs 2 and 4%				0.767	0.895	0.372	0.788	0.644	0.680	0.234	0.142	0.267	0.227
Straw, 2 vs 4%				0.564	0.532	0.726	0.733	0.901	0.446	0.522	0.387	0.252	0.380
SBP, 0 vs 2 and 4%				0.471	0.211	0.623	0.994	0.572	0.752	0.240	0.081	0.172	0.593
SBP, 2 vs 4%				0.537	0.186	0.950	0.468	0.978	0.314	0.704	0.882	0.834	0.802
Straw linear				0.970	0.646	0.326	0.952	0.746	0.478	0.486	0.394	0.691	0.545
SBP linear				0.310	0.064	0.681	0.727	0.649	0.455	0.241	0.112	0.191	0.561

[1] Roche color fan.

[2] L* value means lighter color; a higher [3] a* value means a redder color; a higher [4] b* value means a more yellow color

[5] Standard error of the mean (4 replicates of 10 hens each per treatment)

[6] All of the interactions studied between level and source of fiber, and energy level were not significant (p>0.10)

Table 5. Influence of source and level of fiber of the rearing phase diets and AME_n of the laying phase diets on relative weight (% BW) of the digestive organs and on gizzard content (% organ weight) of hens at46 wk of age

Rearing phase			Laying phase					Gizzard			
Cereal	**Fiber source**	**Inclusion level, %**	**AME, kcal/kg**	**BW (g)**	**GIT[1] (%)**	**Liver (%)**	**Proventriculus[2] (%)**	**RW[2] (%)**	**Weight[2] (g)**	**Content (%)**	**pH**
Barley		0	2,650	1910	12.7	2.65	0.55	2.73	52.15	35.04	3.87
			2,750	1799	12.0	2.70	0.56	2.60	46.87	33.46	3.87
Corn		0	2,650	1941	12.0	2.49	0.51	2.74	53.01	35.48	4.10
			2,750	1802	12.3	2.78	0.51	2.58	46.73	33.83	4.05
	Straw	2	2,650	1843	12.1	2.63	0.53	2.79	51.53	32.97	3.97
			2,750	1776	12.9	2.83	0.58	2.82	50.05	32.16	4.04
		4	2,650	1904	12.6	2.56	0.60	2.98	56.33	32.40	4.12
			2,750	1851	12.6	2.57	0.53	2.72	50.30	30.52	3.87
	SBP	2	2,650	1819	13.0	2.74	0.54	2.70	49.02	31.75	4.21
			2,750	1862	12.5	2.57	0.56	2.69	50.35	30.01	4.13
		4	2,650	1747	12.8	2.58	0.56	2.74	47.76	31.89	4.00
			2,750	1874	12.7	2.73	0.62	2.83	52.88	33.03	4.11
Main effects											
Rearing phase diets											
Fiber source											
	Straw			1843	12.5	2.65	0.56	2.83	52.05	32.01	4.00
	SBP			1826	12.7	2.66	0.57	2.74	50.00	31.67	4.11
Inclusion level, %											
	0			1872	12.1	2.64	0.51	2.66	49.87	34.65	4.08
	2			1825	12.6	2.69	0.55	2.75	50.24	31.73	4.09
	4			1844	12.6	2.61	0.58	2.82	51.82	31.96	4.02
Laying phase diets, AMEn/kg											
	2,650 kcal			1861	12.5	2.61	0.55	2.78	51.63	33.25	4.04
	2,750 kcal			1827	12.5	2.70	0.56	2.71	49.53	32.17	4.01
SEM[3]				65.3	0.36	0.097	0.051	0.138	3.150	1.870	0.140
						Probability					
General model[4]				0.646	0.605	0.368	0.942	0.824	0.639	0.676	0.760
Main effect											
Fiber source during rearing phase				0.717	0.420	0.911	0.721	0.344	0.356	0.791	0.247
Fiber level during rearing phase				0.727	0.196	0.503	0.288	0.344	0.683	0.261	0.793
Energy level during laying phase				0.368	0.898	0.118	0.731	0.345	0.239	0.304	0.667
Contrast											
Straw, 0 vs. 2and4%				0.625	0.199	0.922	0.849	0.144	0.432	0.153	0.516
Straw, 2 vs. 4%				0.304	0.883	0.116	0.259	0.731	0.430	0.536	0.963
SBP, 0 vs. 2and4%				0.426	0.057	0.850	0.160	0.481	0.962	0.101	0.760
SBP, 2 vs. 45				0.653	0.993	0.966	0.465	0.518	0.841	0.380	0.390
Straw linear				0.925	0.242	0.460	0.308	0.168	0.281	0.144	0.576
SBP linear				0.358	0.104	0.882	0.135	0.370	0.887	0.344	0.873

[1] Gastrointestinal tract full

[2] All of digestive organ expressed are full

[3] Standard error of the mean (4 replicates of 10 hens each per treatment)

[4] All of the interactions studied between level and source of fiber, and energy level were not significant (p>0.10)

Table 6. Influence of source and level of fiber of the rearing phase diets and AME_n of the laying phase diet on the relative length (L, cm/kg BW) of the hen and of the tarsus, small intestine (SI) and cecum of hens at 46 wks of age

Rearing phase			Laying phase	Relative length, cm/kg BW[3]							
Cereal	**Fiber source**	**Inclusion level, %**	**AME, kcal/kg**	**Hen**	**Tarsus**	**Tarsus width**	**Duodenum**	**Jejunum**	**Ileum**	**SI[1]**	**Cecum**
Barley		0	2,650	35.0	5.3	0.67	14.1	41.4	37.7	86.1	12.5
			2,750	36.8	5.6	0.72	13.9	41.5	37.6	86.1	11.3
Corn		0	2,650	33.6	5.3	0.67	14.0	40.2	36.3	85.2	12.1
			2,750	36.0	5.5	0.71	13.8	42.1	38.6	87.5	11.8
	Straw	2	2,650	36.0	5.5	0.69	13.1	40.4	37.2	84.1	13.3
			2,750	36.8	5.6	0.73	14.0	43.5	38.5	89.0	12.4
		4	2,650	35.2	5.4	0.68	13.7	41.4	38.8	87.1	12.2
			2,750	35.4	5.5	0.71	15.4	41.9	38.5	88.0	12.7
	SBP	2	2,650	36.2	5.5	0.73	13.7	44.0	39.1	89.9	11.7
			2,750	35.3	5.4	0.67	13.4	41.5	37.0	85.2	12.4
		4	2,650	36.9	5.6	0.75	14.8	44.3	41.5	90.8	12.3
			2,750	34.9	5.5	0.68	12.9	42.7	37.1	86.2	12.9
Main effects											
Rearing phase diets											
Fiber source											
	Straw			35.9	5.5	0.70	14.1	41.8	38.2	87.1	12.7
	SBP			35.8	5.5	0.71	13.7	43.1	38.7	88.1	12.3
Fiber level, %											
	0			34.8	5.4	0.69	13.9	41.1	37.5	86.4	12.0
	2			36.1	5.5	0.70	13.6	42.4	37.9	87.1	12.4
	4			35.6	5.5	0.71	14.2	42.6	39.0	88.0	12.5
Laying phase diets, AMEn/kg											
	2,650 kcal			35.5	5.4	0.70	13.9	42.0	38.4	87.2	12.4
	2,750 kcal			35.9	5.5	0.70	13.9	42.2	37.9	87.0	12.3
SEM[2]				1.23	0.17	0.024	0.79	1.58	1.66	3.05	0.91
								Probability			
General model[4]				0.796	0.897	0.218	0.705	0.732	0.765	0.926	0.960
Main effect											
Fiber source during rearing phase				0.961	0.943	0.771	0.543	0.242	0.734	0.644	0.567
Fiber level during rearing phase				0.494	0.725	0.738	0.527	0.551	0.540	0.792	0.756
Energy level during laying phase				0.552	0.448	0.691	0.975	0.769	0.543	0.915	0.823
Contrast											
Straw, 0 vs. 2 and 4				0.313	0.479	0.550	0.763	0.624	0.589	0.782	0.376
Straw, 2 vs. 4				0.362	0.546	0.565	0.197	0.833	0.632	0.741	0.630
SBP, 0 vs. 2 and 4				0.875	0.441	0.435	0.725	0.145	0.404	0.511	0.657
SBP, 2 vs. 4				0.333	0.645	0.395	0.823	0.643	0.449	0.753	0.498
Straw Linear				0.679	0.761	0.804	0.366	0.144	0.483	0.693	0.615
SBP Linear				0.367	0.382	0.226	0.986	0.754	0.276	0.480	0.491

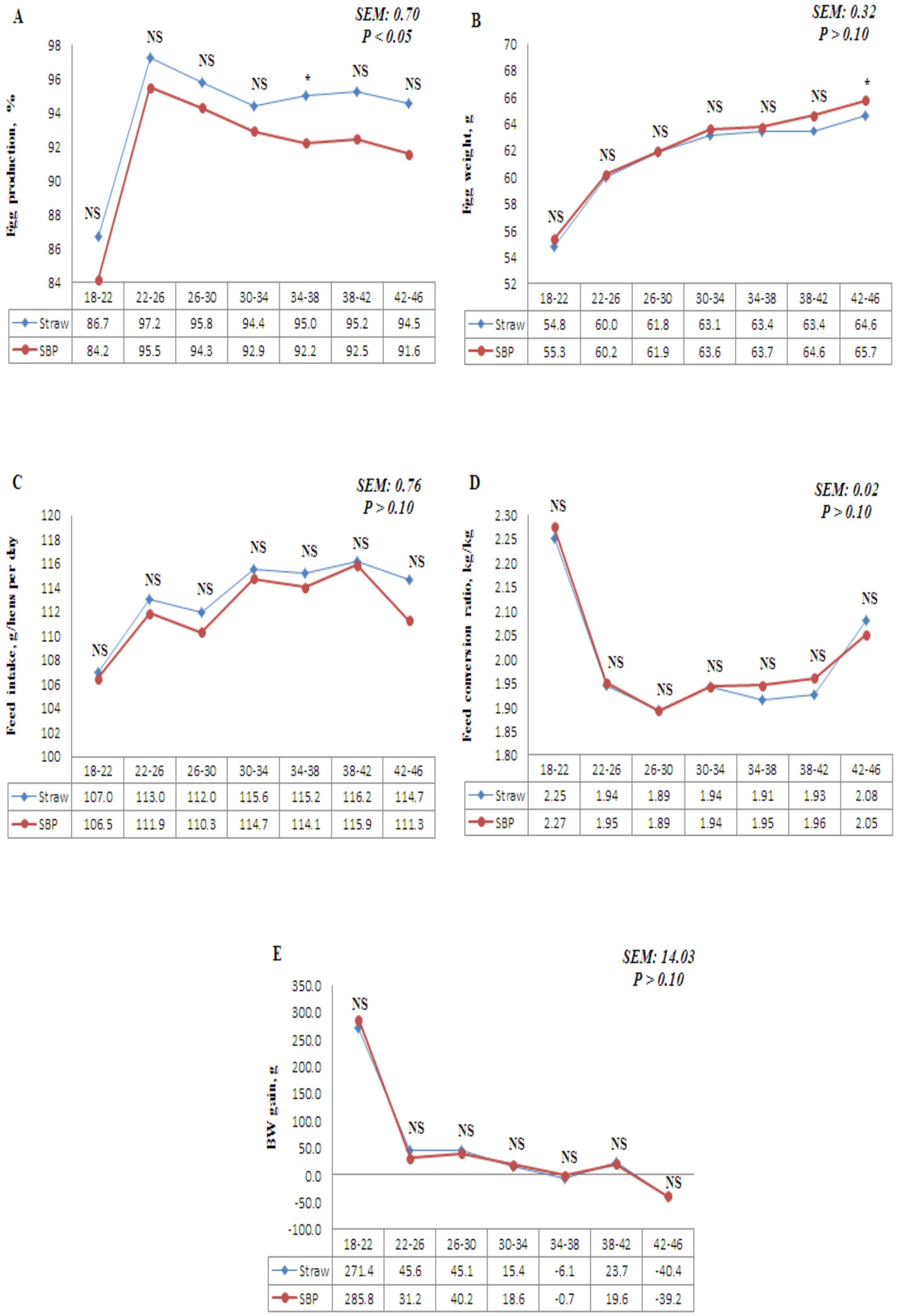
[1] Small intestine: was evaluated from data from the duodenum, jejunum and ileum

[2] Standard error of the mean (4 replicates of 10 hens each per treatment)

[3] The BW of hen was indicated in table5

[4] All of the interactions studied between level and source of fiber, and energy level were not significant (p>0.10)

Figure 1. Effect of fiber source of the rearing phase diets on egg production (A), egg weight (B), feed intake (C), feed conversion ratio (D) and BW gain (E) from 18 to 46 wk of age.



A

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Straw	86.7	97.2	95.8	94.4	95.0	95.2	94.5
SBP	84.2	95.5	94.3	92.9	92.2	92.5	91.6

B

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Straw	54.8	60.0	61.8	63.1	63.4	63.4	64.6
SBP	55.3	60.2	61.9	63.6	63.7	64.6	65.7

C

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Straw	107.0	113.0	112.0	115.6	115.2	116.2	114.7
SBP	106.5	111.9	110.3	114.7	114.1	115.9	111.3

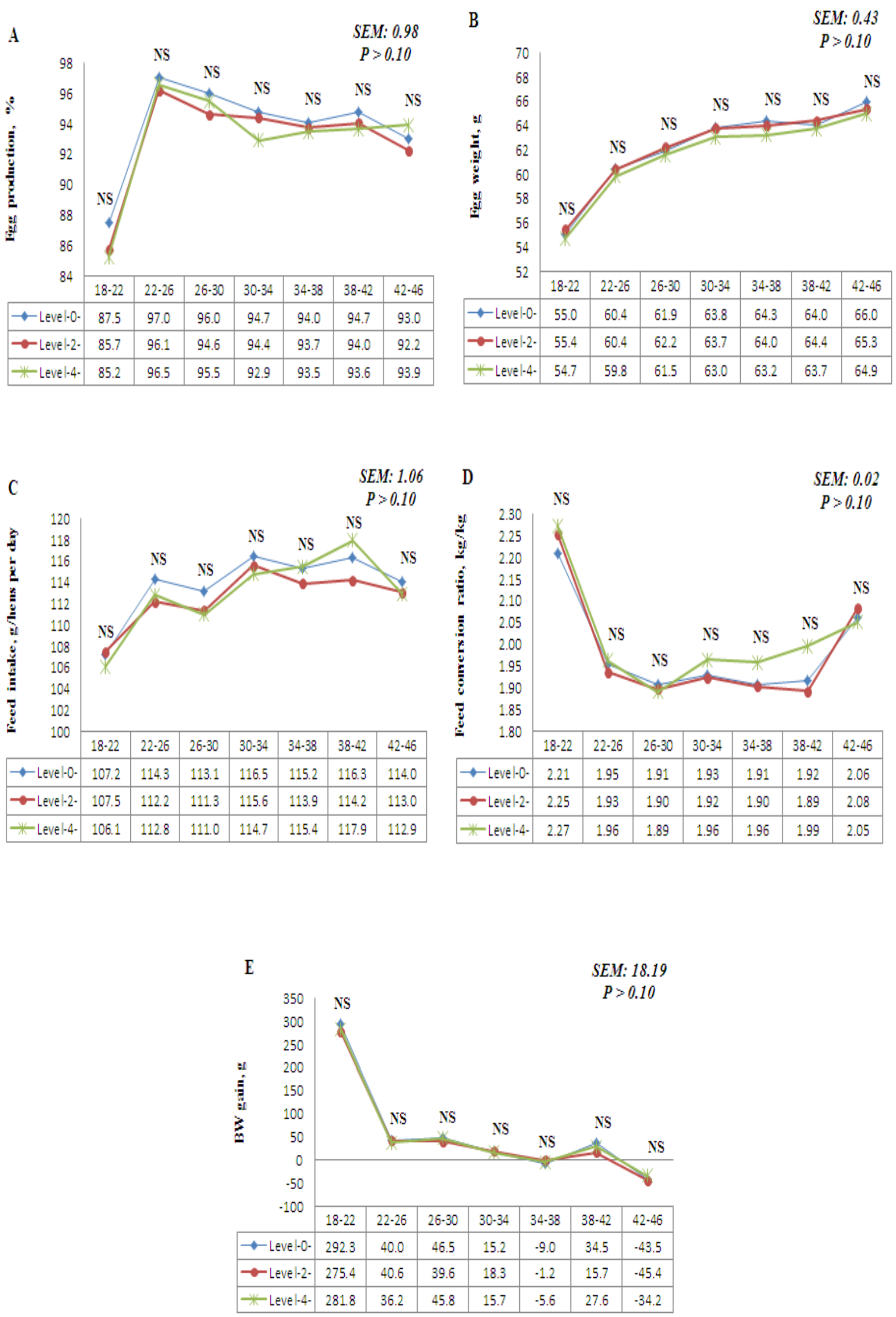
D

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Straw	2.25	1.94	1.89	1.94	1.91	1.93	2.08
SBP	2.27	1.95	1.89	1.94	1.95	1.96	2.05

E

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Straw	271.4	45.6	45.1	15.4	-6.1	23.7	-40.4
SBP	285.8	31.2	40.2	18.6	-0.7	19.6	-39.2

Figure 2. Effect of level of fiber of the rearing phase diets on egg production (A), egg weight (B), feed intake (C), feed conversion ratio (D) and BW gain (E) from 18 to 46 wk of age.



A

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Level-0	87.5	97.0	96.0	94.7	94.0	94.7	93.0
Level-2	85.7	96.1	94.6	94.4	93.7	94.0	92.2
Level-4	85.2	96.5	95.5	92.9	93.5	93.6	93.9

B

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Level-0	55.0	60.4	61.9	63.8	64.3	64.0	66.0
Level-2	55.4	60.4	62.2	63.7	64.0	64.4	65.3
Level-4	54.7	59.8	61.5	63.0	63.2	63.7	64.9

C

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Level-0	107.2	114.3	113.1	116.5	115.2	116.3	114.0
Level-2	107.5	112.2	111.3	115.6	113.9	114.2	113.0
Level-4	106.1	112.8	111.0	114.7	115.4	117.9	112.9

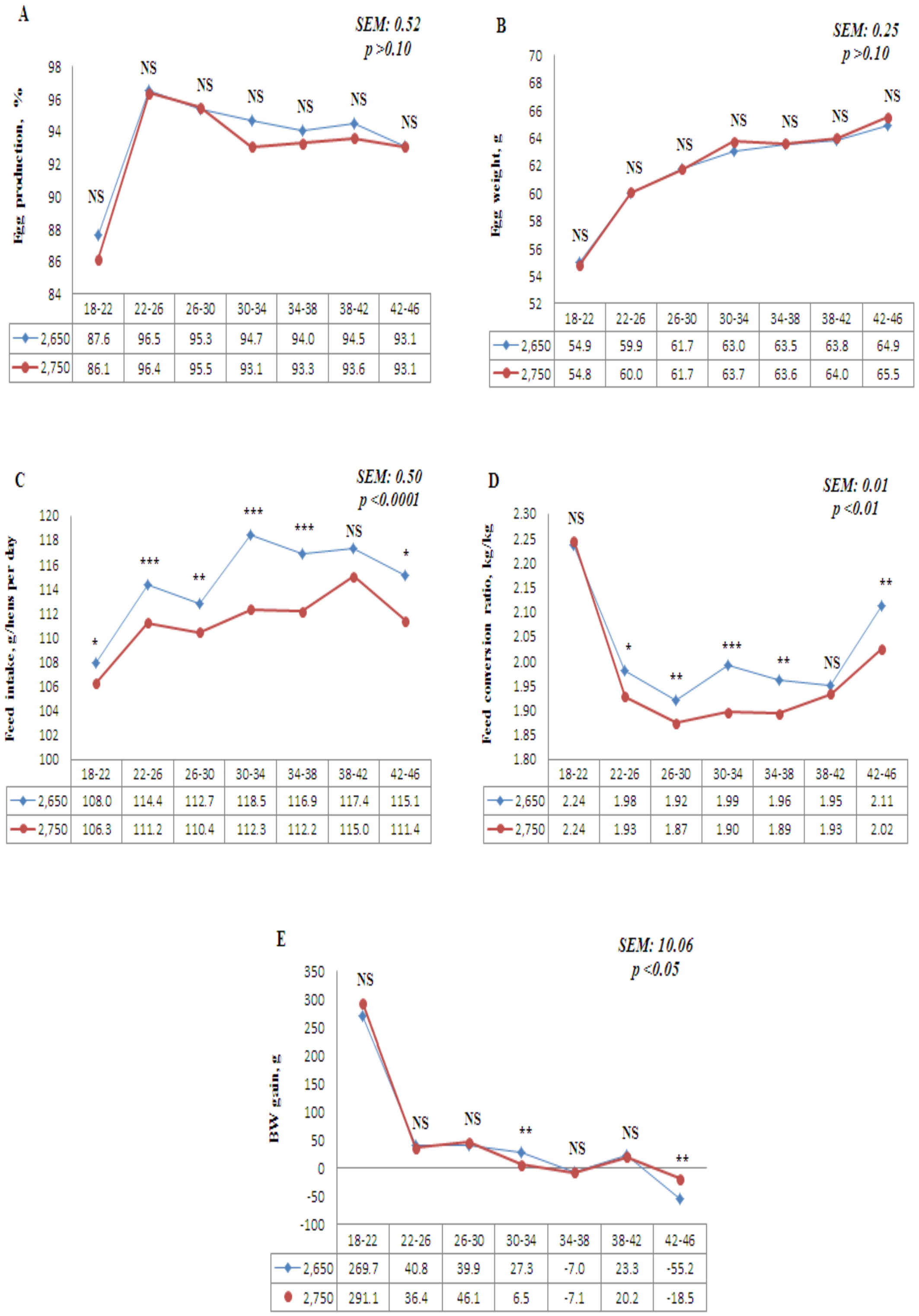
D

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Level-0	2.21	1.95	1.91	1.93	1.91	1.92	2.06
Level-2	2.25	1.93	1.90	1.92	1.90	1.89	2.08
Level-4	2.27	1.96	1.89	1.96	1.96	1.99	2.05

E

	18-22	22-26	26-30	30-34	34-38	38-42	42-46
Level-0	292.3	40.0	46.5	15.2	-9.0	34.5	-43.5
Level-2	275.4	40.6	39.6	18.3	-1.2	15.7	-45.4
Level-4	281.8	36.2	45.8	15.7	-5.6	27.6	-34.2

Figure 3. Effect of AMEn concentration of the laying phase diet (kcal/kg) on egg production (A), egg weight (B), feed intake (C), feed conversion ratio (D) and BW gain (E) from 18 to 46wk of age.



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Anejo
Ingredient composition of the rearing phase diets (Guzmán et al., 2013)

Table: Ingredient composition (% as fed basis, unless otherwise indicated) of the experimental diets

	0-5 week				5-10 week				10-17 week			
	Barley	Corn			Barley	Corn			Barley	Corn		
		0%	2%	4%		0%	2%	4%		0%	2%	4%
Barley	35.0	-	-	-	45.0	5.0	4.9	4.8	50.0	25.0	24.5	24.0
Corn	-	40.0	39.3	38.5	-	40.0	39.2	38.4	-	40.0	39.2	38.4
Wheat	24.0	18.0	17.6	17.3	26.7	15.0	14.7	14.4	19.3	1.1	1.08	1.06
Soybean meal (47% CP)	32.1	35.0	34.3	33.6	23.6	25.2	24.7	24.2	18.0	18.8	18.4	18.0
Wheat bran	-	-	-	-	-	10.1	9.9	9.7	7.3	10.2	10.0	9.8
Fiber source[a]	-	-	2.0	4.0	-	-	2.0	4.0	-	-	2.0	4.0
Poultry fat	4.6	2.65	2.54	2.44	1.03	1.04	1.02	0.98	1.02	0.5	0.49	0.48
Monocalcium phosphate	1.96	2.05	2.01	1.95	1.16	1.35	1.32	1.3	1.97	1.96	1.93	1,89
Calcium carbonate	1.29	1.26	1.22	1.20	1.43	1.28	1.26	1.23	1.46	1.5	1.47	1.45
Sodium chloride	0.34	0.36	0.35	0.34	0.35	0.33	0.32	0.31	0.35	0.35	0.34	0.34
DL-methionine (99%)	0.21	0.18	0.18	0.17	0.17	0.16	0.15	0.15	0.1	0.08	0.08	0.07
L-Lys HCl (78%)	-	-	-	-	0.05	0.04	0.03	0.03	-	-		
L-Thr (98%)	-	-	-	-	0.01	-	-	-	-	0.01	0.01	0.01
Vitamin and mineral premix[b]	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5

[a] The fiber sources used were sugar beet pulp and straw, depending on experimental treatment.

[b] Provided the following (per kilogram of diet): vitamin A (trans-retinyl acetate),6,000 IU; vitamin D_3 (cholecalciferol), 1,200 IU; vitamin E (all-*rac*-tocopherol-acetate), 5 mg; vitamin K_3 (bisulphatemenadione complex), 1.5 mg; riboflavin, 3.5 mg; betaine, 67.5 mg; thiamin (thiamine-mononitrate) ,1 mg; vitamin B_{12} (cyanocobalamin), 15 µg; Se (Na_2SeO_3), 0.1 mg; I (KI), 1.9 mg; Cu ($CuSO_4 \cdot 5H_2O$), 4 mg; Fe ($FeCO_3$), 18 mg; Mn (MnO), 66 mg; and Zn (ZnO), 37 mg.

Muchas Gracias