PREFERENCE, FEED CHARACTERISTICS, AND FEATHER PECKING IN LAYING HENS

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By

Centaine R. Raginski

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ABSTRACT

Feather pecking has been controlled in the laying hen industry through methods such as beak trimming, breeding low feather pecking lines, providing enrichment items, and altering diets to promote satiety. Two recent studies have shown the positive impact of feeding silage (a relatively low nutrient feed item) in addition to (or as supplement) a nutritionally balanced hen ration in terms of decreased aggressive and feather pecking behaviour and improved feather score. The objectives of this thesis were to determine which specific characteristic(s) hens prefer, and if the preferred characteristics were responsible for improving hen welfare. Initial experiments consisted of a preference test, which provided a supplement in a removable insert in front of half a cage, and a balanced ration in the other half. In the first experiment, hens were shown to have a locational preference in the feed trough, not resulting from the presence of the insert, nor from intraspecific competition. Experiment two tested the preference for an ensiled or non-ensiled, and moist or dry material using barley greens, dried barley greens, barley silage, and dried barley silage. Results indicated that hens show preference for moist, non-ensiled materials, but ensiled materials are not rejected. Further investigations took place focusing on physical characteristics as experiment two results suggested a preference for unfermented products. Experiment three examined the provision of a forage-able substrate which was edible (wet or dry barley) or not (plastic lace), and found that edible materials increase the time spent at the feeder. Experiment four tested the preference for particle size of various edible materials (oats, silage and alfalfa) and found hens spend more time with smaller particles sizes of silage and oat materials. The final experiment (Experiment five) used a supplement with all preferred characteristics (wet pea fibre) to determine its impact on feather pecking in birds housed in conventional and enriched cages. Birds given supplemental material did not increase time spent at the feeder, and feathers showed no more or less wear than would be expected from the housing system. Vent scores (areas typically affected by pecking) tended to improve under the presence of pea fibre. Feather pecking decreased when hens were offered pea fibre when housed in large group and enriched cages. Decreased body weight, heavier gizzards, and improved feed efficiency (balanced feed per dozen eggs) point towards improved digestive efficiencies with consumed fibre, even though elongation of the ileum and jejunum seem to contradict this hypothesis. While highly preferred, moist, non-ensiled, low nutrient pea fibre did not impact hen welfare to the degree seen in previous studies with silage. However, similar feedstuffs are still a possible means of enrichment to reduce feather pecking and increase welfare in laying hens, and future research should continue with different bird strains and materials for enrichment, as well as examining hen digestive efficiencies and effects on production costs.

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LIST OF ABBREVIATIONS

BG – barley greens

BS – barley silage

CA – cubed alfalfa, dry

cm - centimeters

d – day(s)

DBG – dried barley greens

DBS – dried barley silage

DM – dry matter

g – grams

GA – ground alfalfa cubes

GBS – ground/mulched barley silage

GO – finely ground whole oat grains

h – hour(s)

min – minute(s)

sec – second(s)

Trt – treatment

WO – whole oat grains

1.0 INTRODUCTION

Feather pecking, an industrial dilemma in poultry production, is a common occurrence in laying hens. Feather pecking results in the deterioration of feather condition and coverage, thereby reducing a bird's welfare by causing distress and difficulty in maintaining body temperature. It can also lead to cannibalism.

Current prevention methods in the commercial industry include, but are not limited to, beak trimming, providing enrichment devices, reducing light intensity, and using low feather pecking lines. Feather pecking has been hypothesized to be the result of redirected foraging behaviour. Since birds kept in conventional cages lack any kind of substrate to forage in, except for their feed, this hypothesis is plausible. Birds may direct activity towards objects in their environment such as the cage floor and sides, feed, and feathers when lacking a suitable foraging substrate. While enrichment items have been advertised as a solution, dietary changes have been tested, show promise, and should not be overlooked.

Diets can be diluted with fibre, which extends the time to nutritional satiety as birds are required to eat more of a diluted diet to achieve similar nutritional gain from the undiluted diet. Providing additional higher fibre content may also impact physical satiety (i.e., a distension of the digestive tract). Birds are then less likely to look or forage for more edible items they may be missing in a concentrated diet, and are less motivated to peck at other birds as a result of this foraging motivation.

Studies have also found that feed form can impact feather pecking behaviour. Mash diets take more time to consume and improve feather condition as compared to pelleted diets. Feeding wet and wet fermented feeds have also indicated an improvement in feather condition. In the majority of these studies however, birds are only given one diet which they are obligated to consume and are not at liberty to choose their preferred level of intake of moist/moist-fermented feed product.

Two recent studies have opted for a preference layout in contrast to the typical one-diet experimental design. These two studies offer a supplemental material as an additional option to the balanced hen ration. In both cases, hens were provided with silage (the product of fermented plant greens containing a plethora of microbial synthesized compounds). The supplemental silage improved feather coverage, and decreased aggressive and feather pecking behaviour without affecting production parameters. The intriguing aspect of this is that the hens are actively choosing to consume silage over their balanced ration despite its comparatively low nutritive value.

Silages are not an ideal material to feed to laying hens because of difficulties in its distribution in a commercial barn. There are also problems with the storage of silages since the material can spoil. However, studies suggest there are behavioural benefits to providing hens with silage, and so exploring the silage characteristics preferred by laying hens could identify important feed properties. These properties in turn can be utilized to discern new nutritional enrichment materials with similar materials but more convenient for commercial use than the silage. The following paper investigates a hen's preference for the various characteristics of silage that are either a product of its characteristic fermentation, or its physical properties. The paper also describes the impact on feather pecking of having provided a material encompassing the determined preferred characteristics.

2.0 LITERATURE REVIEW

2.1 Feather Pecking Behaviour

Feather pecking is a behaviour considered to be any pecking with the beak by an individual bird directed at itself or other individuals, resulting in the removal of feathers. This behaviour is not associated with establishing dominance unlike aggressive pecking (Savory, 1995). Feather pecking behaviour should not be confused with allopreening; a behaviour where the preening (cleaning and alignment) of feathers is performed by one individual upon another. Allopreening, unlike feather pecking, does occur in order to maintain social bonds or social status in social and enriched living animals. In commercial poultry, feather pecking is considered a behavioural disorder (reviewed by Flisikowski *et al.*, 2008) that is defined by the actions of pecking at and pulling out of feathers of neighbouring birds (Savory, 1995). The study of feather pecking has come to denote two types of this behaviour in commercial flocks, gentle and aggressive (or severe). Gentle feather pecking is often considered pecking at the feather of another bird without any removal of feathers, while aggressive (or severe) feather pecking results in the removal of feathers, and a reaction from the individual being pecked (i.e., moving away from the attacker; Keeling, 1994). Aggressive feather pecking should not be confused with aggressive pecking, a behaviour whereby pecking is intently directed at the head region of another individual in order to establish or reinforce social status (Savory, 1995).

Feather pecking in laying hens is common in the poultry industry, where its occurrence and number of birds affected varies with the flock and management practices. Huber-Eicher and Sebo (2001) observed a high rate of feather pecking (over 30 interactions per 30 birds per 30 min) to occur in 77% of 25 commercial flocks by the end of the rearing period. However, the majority of the pecking was considered gentle and so did not necessarily have detrimental effects on hen welfare. The presence of feather pecking is considered to reduce laying hen welfare in commercial systems because high incidences of pecking can often lead to cannibalism. This unwanted behaviour results from continuous pecking of previously wounded areas. The steady pecking attacks may lead to stress in individual birds, as well as pain from the pecking or the resulting wounds. The loss in feathers reduces feather coverage, thereby causing difficulty in maintaining thermal homeostasis. This in turn can affect productivity due to the prioritisation of body maintenance over egg production.

2.1.1 Causal Factors

Feather pecking behaviour can occur in many breeds and strains of poultry as well as at any age. Management practices influence the occurrence of the behaviour and thus, are proposed causal factors. For example, high densities or large group numbers show increased pecking damage compared to relatively less dense groups (Savory *et al.*, 1999; Bestman *et al.,* 2009). Hetland *et al.* (2004) noticed larger group sizes in furnished cages worsened plumage condition compared to conventional cages with smaller groups housed together. Similarly, the size, placement and use of equipment such as feeders and nest boxes, can reduce the number and nature of interactions that promote the behaviour (van Krimpen *et al.*, 2005). In addition, the social nature of the chicken can cause amplification in the occurrence of this behaviour by social facilitation, where one individual learns the behaviour from another by observation.

Feather pecking behaviour has been hypothesized to be the result of redirecting other behaviours, specifically foraging, where birds peck at other objects to satiate an internal want (Blokhuis and Arkes, 1984; Blokhuis, 1986). It has been demonstrated that birds floor housed on slats direct more severe pecks at conspecifics (and less at the ground) than birds housed in same environment with straw litter (Aerni *et al.* 2000). Aerni *et al.* (2000) found that providing straw as a foraging substrate helped to reduce the occurrence and severity of the feather pecking. In addition, birds housed without the litter also show higher heterocyte-to-lymphocyte (H:L) ratios indicating an increased stress level under this condition (El-lethey *et al.*, 2000). Thus, birds given the ability to forage decrease the occurrence of feather pecking and show less stress symptoms. Research with Muscovy ducks also supports the theory of redirected foraging. Riber and Mench (2008) presented Muscovy ducks with various means of feed and water enrichment (placement of items into the water or feed source) to promote foraging. They found that enriched birds engaged in feather pecking less, foraged more and were overall more active than control birds which were given an empty feeder. Thus, in general, studies support the redirected foraging theory by demonstrating that the provision of some foraging material can promote foraging activity instead of feather pecking, thereby reducing feather pecking behaviour.

There is evidence that genetics underlie feather pecking behaviour. Kjaer *et al*. (2001) were able to select for and against feather pecking based on behaviour. The ability to artificially select feather pecking behaviour supports a genetic basis of the behaviour. Feather pecking has been shown to be associated with variations in the DRD4 and DEAF1 gene regions (Flisikowski *et al.*, 2008). The DRD4 gene was chosen because of its influence in early exploratory behaviour in passerines. Sequencing of this gene between high and low feather pecking lines revealed a difference in the number of non-functional genotype and allele variations between the lines. In addition, a neighbouring gene (DEAF1) exhibited greater variations within high feather pecking lines. The DEAF1 gene showed more promise of being a causal factor in feather pecking because of its importance in regulating the serotonin (a chemical found in the body related to the feeling of comfort and well-being) receptor. Flisikowski *et al.* (2008) do not dismiss the possibility of other genes being as much or more involved in feather pecking than the DRD4 and DEAF1 genes. The identification of a gene(s) that controls this behaviour may lead to better breeding or molecular strategies in the future to control, or even remove, the behaviour from a population.

2.1.2 Control Methods and their difficulties

Controlling feather pecking behaviour by genetic methods alone isn't yet perfect, so the industry has come to investigate new environmental or managerial and enrichment methods as ways to reduce the occurrence of this behaviour. Feather pecking and cannibalism are frequently managed by beak trimming birds. Birds with trimmed beaks (either trough hotblade or infrared methods) can impart less damage to feathers and skin. Low light intensity is also often found to decrease the occurrence of feather pecking, as indicated by better plumage condition (Hughes and Duncan, 1972; Kjaer and Vestergaard, 1999).

However, welfare concerns arise in regards to all the previous methods. Beak trimming, while helpful, can cause neuromas (a bulbous formation of the nerve tip) depending on the age of the bird and severity of the treatment in the trimmed area of the beak (Breward and Gentle, 1985; Lunam *et al.*, 1996; reviewed by Gentle, 2011). This can cause sensitivity of the beak and pain as indicated by a decrease in pecking force in beak trimmed birds (Dennis and Cheng, 2010), perhaps another reason for the birds not to peck their cage mates. Low light intensities can make it difficult for hens to see, and may cause and enlargement of the eye organ as indicated by weight (Blatchford *et al.*, 2009; Deep, 2010). Albeit lighting management is less of a welfare concern as its effects have relatively less impact on overall bird well-being, however, both methods (beak trimming and low light intensity) are currently in practice.

Enrichment items, such as string and straw, are in common use among zoo animals. Enrichment items provide new stimuli to promote foraging and play behaviour, as well as the overall mental well-being of animals (Swasigood *et al.*, 2001). The benefit of using manufactured materials is their cleanliness. Using materials such as straw or sand can result in material settling in manure and machinery, thereby causing potential problems to operations. Hens with untrimmed beaks in litter pens decreased the number of severe feather pecks when white bailing twine was hung in the cage as compared to no twine given, even when both treatments were provided additional foraging material (McAdie *et al.*, 2005). McAdie *et al.* (2005) also tested the same devices with hens in a commercial two-tiered system and observed reduced feather condition in birds without the string devices as compared to birds with the devices. Gvaryahu *et al.* (1994) also observed the benefits of enrichment devices (in this case, coloured plastic rings attached to the cage) on pecking behaviour. The experiment showed reduced aggressive hen pecks in hens given the rings. Providing inedible, reusable, materials may be a way to improve hen welfare as indicated by behavioural changes and improved feather condition.

In commercial poultry systems, use of enrichment items is not as common as in zoos. While they may reduce feather pecking behaviour, they may also be difficult for a producer to implement and also require maintenance. Edible and non-edible materials have been shown to have similar attraction to hens. Huber-Eicher and Wechsler (1998) observed no difference in feather pecking in hens given either straw or polystyrene blocks. Thus the use of an edible material for the hens, while an added cost, would require less maintenance than an enrichment device (the birds would consume the material) so long as it were relatively economical, easily distributable and not prone to spoilage.

2.2 The Role of Diet and Nutrition in Feather Pecking

Feed is an important aspect of poultry production and is a factor that can affect bird behaviour and can potentially be used to modify the frequency of feather pecking. Different aspects of feed and diet have been observed to have an impact on the occurrence of feather pecking behaviour (reviewed by van Krimpen *et al.*, 2005). The following sections outline key aspects of the diet that can affect animal behaviour.

2.2.1 Nutritional Deficiencies

Nutritional status has shown to impact behaviour. In the simplest example, a hungry individual exhibits different behaviours from a satiated individual. More specifically, the nutritional status of an individual can cause an alteration in the animal's behaviour, often a change in behaviour that would result in the increased probability of consuming the missing nutrient (Hirsch, 1982). Hence, a nutrient deprived individual will actively search out a new source (investigative pecking) and should be more acceptable of the presentation of a novel diet.

A range of dietary nutrients including amino acids, lipids, and carbohydrates can influence animal behaviour (Bosch *et al.*, 2007). Ambrosen and Petersen (1997) concluded that higher levels of protein in the diet (ranging from deficient to sufficient) of poultry result in better plumage condition and less mortality due to cannibalism. This suggests that feather pecking is a means to increase protein uptake. Alternatively the attraction to the protein rich feed may be due to an attraction to the misshapen feathers that result from a protein deficiency.

The amino acid tryptophan is important in regards to aggressive behaviour like feather pecking as it is a precursor to serotonin (Savory *et al.*, 1999), a calm-inducing compound in the body. Feathers are a protein structure, formed of keratin, which contain amino acids such as tryptophan. Birds that consume feathers could possibly be searching for amino acids lacking in their diet such as tryptophan to help form serotonin and increase their levels in the body, thereby creating a calming effect on the bird and reducing the drive to feather peck. While logical, the consumption of feathers for tryptophan is unlikely as feathers are found to be high in serine, proline, and cysteine and low in tryptophan (Murphy and King, 1982).

Mineral nutrition can also have an impact on behaviour. For example, research has demonstrated that low sodium diets were increase cannibalism (Hughes and Whitehead, 1974). However, the results from this study showed no difference in feather scores, indirectly indicating no difference in the occurrence of feather pecking behaviour between the two sodium level diets. When Hughes and Whitehead (1979) conducted a similar study, they noted an insignificant change in terms of feathering, but a change in feeding behaviour. An increase in pecking behaviour at novel objects and fecal material was also observed when birds were subjected to a low sodium diet compared to nearly none associated with feeding diets containing adequate dietary sodium. Low sodium (or mineral) levels change bird behaviour by increasing exploratory behaviour (indicated by decrease plumage condition and increase pecking behaviour), the same behavioural changes that occur from low nutrient diets.

Mineral malnutrition can also alter mental capabilities and cognitive function, which results in an alteration in behaviour (Hirsch, 1982). Disciplined school children who were placed on a complete vitamin-mineral tablet with 100% of the required daily intake, performed less infractions and were disciplined less when compared to those given placebos, indicating that insufficient vitamin concentration influences brain function and subsequent behaviour (Schoenthaler and Bier, 2000). Gesch *et al.* (2002) conducted a similar study in prisoners, with the addition of an Omega-3 and -6 fatty acid supplement. Results demonstrated that supplementation caused a significant decrease in the number of misconduct reports compared to the placebo group, who were behaviourally similar to the test group before the trial. However, neither experiment determined whether the subjects were deficient in nutrition prior to testing, but still indicate that nutritional deficiencies can have a marked effect on behaviour.

2.2.2 Feed Form

Poultry diets are typically fed in one of two forms, mash and pellet. Pelleted diet processing consists of grinding (either coarse or fine) and mixing the dietary ingredients, before forming the pellet shape through heat, pressure and moisture. Mash diet ingredients are more coarsely ground and mixed, without any further processing. The pellet form results in a greater intake of nutrients with one beak full compared to one beak full of the mash form. This is due to the greater concentration of nutrients stemming from the compaction of the diet during pellet formation. Mash diets, therefore increase the time hens must manipulate and consume enough feed to equal the same nutrient intake of the pellet.

If feed form affects nutrient intake and manipulation time, can it impact feather pecking? Hartini *et al.* (2003) tested feed form effects on the outbreak of cannibalism resulting from feather pecking. Results showed increased feather pecking with the pelleted diets, indicating that the increased time needed to consume a mash diet leaves less time for other activities like feather pecking. Savory *et al.* (1999) also noted that feather pecking worsened in birds fed pelleted diets over mash diets. This shows a similar effect to the presence of an enrichment material, where the mash diet appears to act like the foraging substrate by reducing the time available for other behaviours such as feather pecking. It then seems that the increased time occupied in activities other than feather pecking is important in the reduction of pecking behaviour. El-lethey *et al.* (2000) found higher H:L ratios in pellet fed birds, indicating greater stress in these hens, maybe resulting from higher rates of feather pecking. In horses, those fed hay spent significantly more time eating and less time chewing wood (a stereotypic behaviour) in comparison to horses receiving a concentrated pellet diet (Willard *et al.*, 1977). These results indicated an unpelleted diet can modify and/or reduce unwanted behaviour.

It can be suggested that the increased time associated with mechanically digesting a pelleted feed is not the factor affecting behaviour (as pellets dissolve rapidly into the ground material upon ingestion; reviewed by Svihus, 2011), but the time associated with manipulating the feed (occurring more with mash diets). The greater the time associated with feed handling, the less time for feather pecking. This concept is often stated as the redirected foraging theory (Blokhuis, 1986). The ingredients in a mash diet are not always ground the same, leaving a variety of sizes and colours. This variation may stimulate pecking at and sorting through the feed, choosing different ingredients to consume. The horses in Willard *et al.* (1977) were also observed to spend less time engaging in coprophagy and searching when fed a pelleted diet. Thus the horses are redirecting their behaviour, or looking for an alternative source for a missing nutrient, when no suitable foraging materials are present.

2.2.3 Fibre

Fibre is considered “any dietary component that reaches the colon without being absorbed in a healthy human gut' (Ha *et al*. 2000). It is often considered to be one entity, but it is composed of multiple components and different aspects of the fibre are often characterized based on the method of analysis, its chemical characteristics, and its physical properties (such as the size of fibre particles). Broadly, it can be considered as non-starch polysaccharides (NSP), which are indigestible in the chicken’s digestive tract, and can be either soluble or insoluble (Cummings, 1981).

Particularly in humans, fibre intake is promoted due to its benefits to overall health. Fibre promotes microorganism growth in the digestive tract and the production of volatile fatty acids (Cummings, 1981). So, if fibre is an important requirement in the human diet, it may also be important in the diet of chickens. Perhaps, feather pecking may then be a behavioural change as the result of low fibre in the diet since, as previously mentioned, hens fed pelleted diets (of a quick-dissolving nature) quickly reach nutrient satiety.

The obstacle when researching the importance of fibre is that the addition of fibre to a diet dilutes the nutrients causing individuals to consume more diet to gain the same energy intake as a diet without the additional fibre material. Bearse *et al.* (1940) experimented with the oat hull, a component previously observed to reduce cannibalism. Bearse *et al.* (1940) compared corn based diets containing fibre, ash and water extract components of the oat hull. They found the fibre added diet resulted in the best plumage condition and lowest mortality due to cannibalism but essentially diluted the diet when the bulky oat hull fibre component was included. van Krimpen (2008) found the addition of sand and fibre to the diet (which caused a nutrient dilution) significantly reduced feather pecking. Thus, birds had to spend more time consuming feed to gain the same energy intake, which left less time for feather pecking activity. Hartini *et al.* (2003) found higher cannibalism rates and shorter feeding times in birds fed low-fibre diets. A higher active escape rate from pecking was seen in low-fibre-fed birds, while higher-fibre fed birds tended to freeze when pecked by a conspecific, indicating either a tolerance to pecking, or a decrease in aggressive pecking to more gentle pecking by the pecker (Hartini *et al.*, 2003). The increased time spent feeding on high-fibre diets also indicate that less time would be available to engage in feather pecking behaviour as well. Kjaer and Hansen (2007), after supplementing the partridges' usual diet with roughage (maize silage, wheat sprouts, or rucola salad), found the roughage to elicit some changes in behaviour. Partridges fed silage spent more time lying and less time foraging compared to the other treatments and controls (no roughage). They suggested that their inclusion of a sand box (a material easily ingested) may have fulfilled the need to forage (a behaviour often associated in dustbathing bouts) without the roughage, and so the effect of the roughages on behaviour was not very noticeable.

Dietary fibre may affect behaviour in a variety of animals other than poultry. This effect improves welfare by decreasing undesirable behaviours, supporting the idea that fibre plays an important role in the diet. For example, de Leeuw *et al.* (2008) reviewed research demonstrating that fibre causes a decrease in stereotypies in pigs immediately after feeding. Redbo *et al.* (1998) found the amount of roughage supplemented to thoroughbred and trotting race-horses to be related to the amount of wood-chewing performed. However, the strength with which they concluded this statement does not seem valid when they noted that the amount of roughage consumed between the two horse types did not significantly differ. And lastly, Ramonet *et al.* (1999) found non-lactating sows fed high fibre diets to stand less, spend more time eating and masticating, and less time engaged in non-feeding oral activities (or stereotypies, a repetitive behaviour performed when animals becomes stressed or deprived of some stimuli) compared to those fed lower-fibre diets.

2.2.4 Moisture

Poultry feed is typically fed in a relatively dry form (~10% moisture content) but there is considerable evidence that chickens will consume more of a moist feed than a dry feed. El Kaseh and Forbes (1995) fed broilers diets without and with water (1.5 to 2.0 times that of the weight in feed) and found higher intakes of wet feed on a dry matter basis. Similarly, Whitehead and Scott (2005) found broilers consumed more wet and wet fermented feed (similar intakes) compared to dry feed on an as-is basis. The higher intake of the wet feeds in this research may demonstrate a preference for moisture. The high intake could also logically demonstrate a dilution effect as seen in high fibre diets, however, high water intakes could result in digestive upset.

If chickens have a preference for a moist feed, how wet does the feed have to be? Beyer *et al.* (2002) found birds preferred feed with 0.5 and 1.0 parts added water over dry feed but the addition of more water resulted in feed avoidance (determined by measuring the amount of feed consumed when given the choice between the dry and one level of moisture). In the same study, adding 0.25 parts water was preferred over 1.0 part water. The birds preferred a wet feed over a dry, but demonstrated a limit to the degree of wetness. The information provided in this report is of value for understanding the impact of feed moisture levels, but the validity of the scientific approach used in this research is unclear because it is only published in abstract form.

In contrast to the above data, Riber and Mench (2008) found a water-grain mixture was consumed less by Muscovy ducks than a dry diet. Interpretation of these data is difficult because the ratio of feed to water was not given. The explanation for this observation could include a dislike of moist feed or, more likely, the addition of too much water to the feed which caused an aversion.

In general, it appears that a wet feed may be more palatable as indicated by a higher consumption than dry feed. However the effect of moisture content of feed on behaviour has not been evaluated.

2.2.4.1 Fermented Feed Effects on Plumage and Behaviour

Fermentation is the result of the microbial action of converting carbohydrates (or sugars) from a substrate to alcohols and organic acids. An example of a fermented feed is silage, where the moist environment promotes microbial growth and fermentation creating warm and anaerobic conditions. Very few studies have investigated the impact of typical fermented feeds on poultry plumage condition and behaviour, but two recent studies have shown the impact of supplementation of a related material in laying hens.

2.3 Studies on silage and feather pecking behaviour in laying hens

Silage has a high moisture content (approximately 66-77% moisture based on the silage and ensilation conditions; Steenfeldt, 2007; Johannson, 2008); thus, silage consumption could be expected to be similar to wet feed. A number of recent studies have shown the benefits of feeding silage to poultry. These benefits may be related to silage moisture content or other factors such as the presence of fermentative products (e.g., volatile fatty acids) may also affect animal preference and intake. Research has established that laying hens will eat considerable amounts of silage when given access in addition to a nutritionally adequate dry diet and despite its relatively low nutritional value for monogastric species (Steenfeldt *et al.*, 2007; Johannson, 2008). Both these studies examined behavioural responses in addition to feed intake.

Johannson (2008) offered barley silage *ad libitum* to White Leghorn hens kept in community cages in addition to their nutritionally balanced mash concentrate, and the birds readily ate the fresh silage offered. Results indicated that hens eat less balanced feed and spend more time feeding when supplemented with silage when compared to non-supplemented hens. Behavioural observations (by scan sampling) demonstrated that silage-fed birds performed less aggressive behaviour and more gentle feather pecking. Silage-fed birds also had better feathering compared to controls. These data support the concept that the silage is occupying the hens in activity and leaving less time for pecking behaviour (indicated by better plumage and time spent at the feeder). Hen production, similar to fermented feed and other studies, was not affected by the consumption of silage other than a significant increase in yolk colour in silage fed birds.

Johannson (2008) also studied the impact of feeding silage on bird stress by using H:L ratios and found no difference between the two diets.

Johannson (2008) suggested (with the support and evidence of other studies) that silage, while lacking in apparent nutritive value, may be more palatable to poultry as a result of its high moisture, fermentation products (vitamin B_{12}, volatile fatty acids), or particle size. In addition, its high fibre may promote satiety as the gizzard becomes enlarged and digesta passage rate is decreased.

Steenfeldt *et al.* (2007) found that supplementing a regular diet with some forage material (maize and barley-pea silage, or carrots) reduced feather damage and cannibalism-associated mortality compared to hens only fed a pellet diet. Feed intake of the laying diet was reduced when forage supplement was present (also seen in Johannson, 2008). The time required to manipulate the supplement may account for the time taken away from eating the balanced diet or feather pecking. These forages supply a large amount of fibre as well as nutrients. Carrots in particular supply another energy source as its soluble sugars are available for absorption. In contrast feed sugars would be fermented by microorganisms during the ensiling process (Steenfeldt *et al.*, 2007).

Silage has not only been observed to have an effect on poultry behaviour, but in other animals as well. Silage has been noted to increase the feeding time of cattle (Wilkinson, 2005). Sows supplemented with silage racks spent less time performing sham chewing behaviour (O'Connell, 2007). These behavioural changes under times of stress may indicate that silage induces a calming effect in the sows. This was not the case with poultry as Johannson (2008) found no difference in H:L ratios, a parameter not measured in the sows. The hypotheses presented by O'Connell (2007) to account for the change in stereotypic behaviour turn out to be similar to those postulated by Steenfeldt *et al.* (2007) and Johannson (2008); silage forms a foraging substrate and/or the addition of fibre causes an alteration in animal activities.

These studies, however, leave unanswered questions about why hens find silage particularly attractive, and what causes an alteration of behaviour. Being able to isolate the causal factor would lead then to the potential application of this factor to practical production methods in the industry, as silage *per se* is not a very convenient or straightforward distributable product as poultry feed.

2.3.1 What is silage?

Ensiling is an ancient technique used to preserve whole crops (stems, leaves, and influorescences) for extended periods of time. The end product is typically used in today's agriculture industry as a forage component in the diet of feedlot or housed cattle and dairy. The anaerobic conditions as a result of storage (in silos, wedges, or bails) promote the fermentation of the plant material by anaerobic microorganisms. The result is silage.

Previous research demonstrated the preference of poultry for silage (Steenfeldt *et al.*, 2007; Johannson, 2008); however, what is it about silage that could possibly influence their preference? In other words, what components are characteristic about silage and have the potential to modify animal behaviour?

2.3.1.1 Silage's Unique Characteristics

The previous studies of Johannson (2008) and Steenfeldt *et al.* (2007) indicate that hens readily consume silage in addition to their balanced ration even though silage has a relatively low available nutritional content compared to other feedstuffs. For example, McEniry *et al.* (2008) investigated the changes associated with the stage of crop ensiling. As ensiling continued over time, DM decreased as did water soluble carbohydrates (WSC), an expected occurrence as WSC are utilized as a substrate by microorganisms. Acid detergent fibre showed an increasing trend with ensilage time, while neutral detergent fibre showed a decreasing trend (McEniry *et al.*, 2008), indicating that silage is a source of fibre even though the resident microorganisms have altered the original composition during the fermentation process.

2.3.1.1.1 Compounds Produced by Bacteria

The fermentative process of bacteria involves glycolysis (Carr, 1968). Carbohydrates, such as glucose and fructose, are converted to pyruvate, a process which releases ATP and CO_2 (Carr, 1968; Woolford, 1984). Pyruvate can be converted to other compounds depending on the microorganisms present. Such products include lactic, acetic, butyric, and propionic acids. With the exception of propionic acid, these acids are characteristic of silage (Wolford, 1984; McDonald *et al.*, 1991). Acetic, butyric and propionic acids are commonly termed volatile fatty acids (VFAs) and are short-chain fatty acids (SCFA). Volatile fatty acids can be utilized for different functions including as an energy source by being used in the Kreb's cycle. The metabolization of VFAs and their use as an energy source was reported by Yang *et al.* (1970) who found that labelled carbon, from radioactive labelled VFAs, was expired with the breath in rats approximately 2.5 hr after ingestion. The amount of energy derived from VFAs and contributable to an animal's needs varies with the species; SCFA metabolism contributes 6-9% of maintenance energy in humans, 2-7% in dogs, 9.4% in rats (Yang *et al.*, 1970) and 10-31% in pigs, compared to 60-80% in cattle (Bergman, 1990; Stevens and Hume, 1998; Jozefiak *et al.*, 2004; Bosch *et al.*, 2007). In chickens, Annison *et al.* (1968) estimated that VFAs could provide up to 11% of the metabolizable energy requirement of mature chickens. However others have found that caecal fermentation only contributes 3-5% of the total energy needs of the broiler chicken (Choct *et al.*, 1992; Jorgensen *et al.*, 1996; Jamroz *et al.*, 2002).

Volatile fatty acids may also have metabolic effects other than energy contribution. VFAs appear to mainly be absorbed by the intestinal eptithelium (Bolton and Dewar, 1965), particularly in the ceca (Annison *et al.*, 1968). From there, VFAs are rapidly transferred into the blood (Annison *et al.*, 1968; Argenzio, 1981; Bergman, 1990; Stevens and Hume, 1998) by passive transport (Argenzio, 1981; Bergman, 1990). Measurements of the VFA levels in the portal and peripheral blood indicate that the portal vein and liver are constantly receiving VFAs. Acids such as acetic, formic, butyric, and propionic acids are not taken up and metabolised by the liver, as illustrated by their presence in peripheral veins (Annison *et al.*, 1968; Bergman, 1990). This indicates that VFAs present in silage (acetic, butyric and propionic acids) circulate in the blood stream, leading to the potential to cross the blood-brain barrier. This crossing possibility could potentially induce a calming effect, thereby modifying behaviour such as reducing aggression and feather pecking behaviour in poultry.

Fermentation can also involve the breakdown of protein structures (proteolysis) to amino acids and ammonia, as well as other compounds such as the odorous amines putrescine and cadaverine (Woolford, 1984; Bergman, 1990) and a variety of VFAs (Bergman, 1990). Putrescine and cadaverine are formed by the decarboxylation of amino acids; in this case, ornithine and lysine respectively (reviewed by MacPherson and Violante, 1966). Analysis of a number of silages found these amines to be present at variable but significant levels (MacPherson and Violante, 1966), therefore their presence could alter silage intake.

What about in poultry, do VFAs influence feed intake? Cave (1984) found the addition of propionic acid (100g/kg) to a basal diet decreased broiler feed intake of chicks in comparison to a diet without proprionic acid. A similar comparison of diets containing 0 or 30g/kg of lactic acid failed to show a significant difference in feed intake. Pinchasov and Jensen (1989) looked at the feed intake of broiler chicks when fed by intubation diets containing propionate, acetate, and lactate. Intubation results show that propionate was the only compound to significantly decrease feed intake (up to 20%) at 600 mg/kg. When the three VFAs were combined and incorporated into feed, it was observed that propionate was more effective at reducing intake than the other two acids (only significance). The intubation procedure prevents the chicks from making decisions based on flavour, but ensures choices are made by post-ingestive feedback. The study indicates that poultry can detect VFAs in diets and make choices accordingly, however the VFA's present in silage appear not to be deterrents or attractants to the consumption of silage.

2.3.1.1.2 Vitamin B_{12} and Coprophagy

Vitamin B_{12} (or cyanocobalamin, the most common form) is a water-soluble vitamin that contains cobalt. It is one of the few vitamins that animals cannot synthesize, but is synthesized by microorganisms like bacteria (aerobic and anaerobic; Leeson and Summers, 2001; Martens *et al.*, 2002; Green and Miller, 2007). Thus, animals can only obtain this vitamin by either ingesting it in the food, engaging in coprophagy, or absorbing it from the caecum or colon in the GIT where B_{12} producing microbial populations exist (Flodin, 1988; Baker, 1995; Martens *et al.*, 2002; McDowell, 2006). Silage would be a good source of this vitamin because of its abundant microbial population synthesizing vitamin B_{12}.

A vitamin deficiency in poultry should seem to be inevitable because of this reliance on an external source; however, the body requires relatively small amounts (McDowell, 2006). Vitamin B_{12} is stored in the body unlike the other B vitamins. The liver and the kidneys are the storage sites of vitamin B_{12} as indicated by the detected abundance of the vitamin (Leeson and Summers, 2001; Birn *et al.*, 2003; Green and Miller, 2007).

Laying hens require roughly 0.004 mg/kg (or 4 micrograms/kg) of vitamin B_{12} (NRC, 1994). McDowell (2006) points out that the required levels may simply be the minimum level to prevent signs of deficiency and so may still result in suboptimal performance, which is an interesting comment since humans require 3-5 micrograms per day (Lloyd *et al.*, 1978b), which is similar to that of the chicken even though a human is a much larger animal. Studies with vitamin and coprophagy deprived poultry (deprived of B_{12} sources) indicate that hens require between 2.2 and 4.0 micrograms of vitamin B_{12} to support hatchability (Milligan *et al.*, 1952), thus the hen's B_{12} vitamin status is important to chick health. Anderson *et al.* (1958) found that the quantity of B_{12} in the yolk increased with increased dosage of B_{12} in the hen. While hatchability isn't a concern in laying hens, the amount of vitamin transferred to the yolk is as eggs can be a vitamin source for humans.

Vitamin B_{12} deficiency, often manifested as pernicious anemia, is seen in humans as well as animals. It often occurs in vegans since the vitamin is abundant in animal food sources that they don't eat. It can also be the result of a genetic disorder (i.e. mistranslations resulting in a lack of the R-factor) which can be passed on to children (Flodin, 1988; Leeson and Summer, 2001; Halsted, 2003; Green and Miller, 2007). Inheritance of vitamin B_{12} deficiency also occurs in poultry, where the amount present in the egg reflects the vitamin condition of the hen (Lloyd *et al*., 1978b). Milligan *et al.* (1952) calculated 30-48% of the vitamin fed to the hen was transmitted to the egg yolk. Anderson *et al.* (1958) also noted a correlation between the amount of B_{12} in the hen and her chick, where chicks from hens deprived of B_{12} showed high offspring mortality rates even when they were supplemented post-hatching. Surviving chicks from deprived hens also grew more slowly compared to chicks from hens with higher B_{12} supplementation. Thus it would be evolutionary advantageous for hens to detect and utilize sources of vitamin B_{12} to increase the likelihood of offspring survival.

Vitamin B_{12} deficiency leads to reduced growth, poor feathering, nervous system disorders, as well as decreased hatchability of fertile eggs (reviewed by Leeson and Summers, 2001). It can result in central and peripheral nerve damage that may or may not be reversible (Flodin, 1988). Vitamin B_{12} supplementation has been shown to be useful in some central nervous system, or psychiatric/mental disorders such as hallucinations, irrational behaviour (Ordonez, 1977; Flodin, 1988), and aging related disorders (Green and Miller, 2007) in humans. These neurological changes may be dissociated from the more regular occurring physiological symptoms; nerve damage such as demyelization and axonal degeneration of sensory nerves occur, which can manifest itself as impaired sensory abilities, and be as severe as poor mental status (McCombe and McLeod, 1984; Savage and Lindenbaum, 1995). Treatment of such disorder with B_{12} supplements can cause a variety of responses in humans, from no effect, to improved neural functions (McCombe and McLeod, 1984). Vitamin B_{12}'s impact on the nervous system could indicate that such damage could underlie feather pecking behaviour. Contrary to this possibility, Schiable *et al.* (1947) found feather pecking not to be related to a vitamin B deficiency as feed supplemented with dried yeast (replacing an equivalent amount of corn) had no effect on the rate of cannibalism.

Despite the fact that vitamin B_{12} deficiency has the potential to alter behaviour and underlie some nervous disorders in humans, it does not necessarily underlie feather pecking behaviour. Since poultry are often reared in cages, or in the presence of slatted flooring, they do not gain access to faecal material, and thus may become more deficient in vitamin B_{12} than when housed in a system that does gives them access to their own faeces (McDowell, 2006). Marquering *et al.* (1969) found cockerels to ate 4-5 g of faecal matter when deprived of vitamin B_{12} versus 0.6-0.8 g when the dietary level was sufficient. It must be noted that although it seems very plausible for a vitamin B_{12} deficiency to be the cause for attraction in silage, vitamin B_{12} is not often lacking in a diet because of its inclusion in the vitamin-mineral premix at the required level, thereby making a deficiency an uncommon occurrence. Vitamin B_{12} however, is transferred to the egg making it a constant requirement to regenerate the daily loss associated with production.

2.3.1.1.3 Probiotic Effect

The microbial populations which have created a home in the silage may also impart some probiotic effect in poultry. Lactic acid bacteria cause a reduction in pH in the intestine as a result of the acids they produce, forming a "hostile environment" thereby replacing some of the other species populations (reviewed by Engberg *et al.*, 2009). The more acidic environment would cause a shift in the microbial population of the intestine as some organisms are more tolerant than others to low pH. Fermented feed diets for poultry had higher lactic acid bacteria faecal (Loh *et al.*, 2007) and caecal counts because of their ability to better survive the low pH environment they produce (Engberg *et al.*, 2009). Thus Loh *et al.* (2007) concluded that the inclusion of fermented feed may be useful for promoting good health.

It has been hypothesized that a low pH digestive tract habitat may form a barrier to some of the major poultry pathogens. Heres (2003b) investigated the infection of poultry to *Salmonella enteritidis*. Results showed that birds fed a fermented liquid feed required a greater dose of pathogenic organisms to achieve initial infection (by oral inoculation), it took longer (days) for all birds to begin shedding pathogens in their faecal material, but that no difference in enriched numbers was found in the intestine. Similar results were seen with *Campylobacter jejuni* (Heres, 2003a). Thus, fermented feeds may impart a probiotic effect.

2.3.1.2 Silage Preferences

Hens show a preference for fresh fermented feed, and not leftovers from the previous day as indicated by their interest in new feed (Johannson, 2008; Engberg *et al.*, 2009). This may indicate that poultry prefer components of the silage that are lost after exposure to air, such as volatile fermentation products or water.

Silage possesses a distinct, pungent, and sour odour. In the past, flavour and odour compounds in silage have been of interest because of their potential to impart a smell or taste to milk from dairy cows. Morgan and Pereira (1962) confirmed the presence of a number of VFAs such as propionate, acetate, methanol, ethanol, propanol, among others, (basically a number of fermentative products) as compounds imparting an odour to the silage.

The various volatile, and thus potentially odourous, compounds found in silage have also been investigated in regards to feeding behaviour. Forbes (2007) overviewed the characteristics of silage, which show a relationship to increased consumption in cattle. He found that ammonia and amines are inversely related to intake, and aversion to amines decreases after some time, whereas van Os *et al.* (1995) found no difference in intake in dairy cattle when amines and ammonia were added to a silage preserve.

Cadaverine and pustrescine are odorous amines produced in fermentation that have strong, characteristic odours (O'Neil, 2008). While these odours are detectable to humans, it is uncertain whether poultry are able to detect the same odours. The short chain fatty acids also have characteristic odours that are considered unpleasantly pungent and rancid to humans (O'Neil, 2008) and therefore, could also affect poultry intake of silage. Of interest though, is that Steenfeldt *et al.* (2007) found carrots to be preferred over silage. The question then is, what is it about silage and carrots that make it attractive to hens? Such characteristics include water content, palatability, particle size, colour and nutrient content.

2.4 Possible Factors Affecting Regulation of Silage Consumption

2.4.1 Detecting dietary differences

2.4.1.1 Novelty

Novel foods are often accepted as a result of learning from parents or peers (Rogers and Blundell, 1991; Forbes and Kyriazakis, 1995), which is logical as new foods may be toxic and so avoidance would be a preventative mechanism. Foods would then be accepted or rejected based on feedback after ingestion; malaise or satiety based on the presence of poisons or nutrients affecting the nature and amount of feed consumed (Nolan *et al.*, 1996). Post-ingestive feedback plays a role in determining if something is beneficial by positive feedback (Rogers and Blendell, 1991). Animals also demonstrate their knowledge through choice of feed based on their nutritional status. In other words, to eat for a nutrient that is limiting in the body. Animals also can learn to avoid diets which contain an imbalance of nutrients or toxic compounds (Nolan *et al.*, 1996).

Overall, animals are able to and do make appropriate food selections based on a number of criteria (Nolan *et al.*, 1996), these criteria based on the sensory bodies such as visual appearance, taste, palatability and post-ingestive feedback.

Studies in a variety of animals have shown that learning what to eat is affected by social facilitation. Galef (1993) observed that rats will eat more of a novel flavoured food than a familiar food when exposed to another rat which has already eaten the novel food. When the rats were then exposed to two novel diets, one eaten by a demonstrator and the other not, rats tended to eat food that had previously been eaten by the demonstrator and not the diet novel to both individuals. In hens, Sherwin *et al.* (2002) was able to support previous studies where hens observing were more likely to eat feed a demonstrator ate. In contrast, the hens would not discontinue consuming food a demonstrator would not eat. Rozin and Rodgers (1967) found rats deficient and sufficient in thiamine often initially ate a novel diet over a familiar diet when given the choice without a demonstrator, indicating that the deficiency causes an attraction to novel diets.

2.4.1.2 Ingestive Feedback

Choices may be influenced by positive or negative post-ingestive feedback effects after ingestion, absorption, and post-absorptive metabolism. The principle behind post-ingestive feedback is that animals associate the metabolic properties with the sensory properties of a feed material (Forbes, 2000). Post-ingestive feedback is a learning mechanisms which allows individuals are then able to choose their consumption of following materials based on the positive or negative association with a particular sensory property (flavour, texture, colour, etc.). Thus, what an animal learns post-ingestion from nutrient composition in consuming one material will be applied to whether or not it eats this material in the future (Forbes, 2000).

2.4.1.3 Sensory Systems and Poultry Preferences

Animals can use their senses as cues to distinguish one diet from another. For example, visual and positional cues are used to recognize and remember the diet which gives the greatest positive post ingestive feedback (Forbes and Kyriazakis, 1995). Scott and Verney (1947) recognized that there may be some characteristics of a diet that may act as an immediate cue for associating its post-ingestion effects. They used flavour as a cue to a B vitamin sufficient diet (thiamine, riboflavin, and pyridoxine). When given the choice, rats always preferred the flavoured diet regardless of its vitamin status, indicating that cues, and previous experience, have an impact on diet selection. The authors suggest that the use of the flavour as a cue in detecting a preferred diet must be the result of the animals receiving a beneficial signal after ingestion, which becomes associated with the flavour and no longer the nutrient.

The sensory organs of the birds are important in aiding discrimination of feedstuffs. Chicks learn to distinguish feed items by pecking at various objects to determine if they are edible. Vision plays a role in feed discrimination as a chick observes its mother and siblings pecking at various objects. The attraction to certain objects can be based on visual stimuli such as shape, size, and colour. The author has often observed chicks readily peck at novel items, particularly shimmering items. Studies have shown colour preferences of both chicks and hens (Jones and Carmichael, 1998; Hammershoj and Kidmose, 2006; Ham and Osorio, 2007).

The ability of birds to discriminate between smells by olfaction is rather uncertain, however, other sensory systems are in place to help. Food is picked up by the beak and manipulated by the tongue, which allows for the detection of textures and tastes. Taste buds are located throughout the buccal cavity but the majority are found on the tongue (Mason and Clark, 2000), as well as on the upper beak of the anterior mandible, the epithelium posterior to the tongue, and the ventro-lateral side of the anterior of the tongue (Denbow, 2000). Gill (2007) reports that chickens have 24 taste buds (but up to 200 in birds such as parrots; Sturkie, 1976) compared to the some 10,000 in humans. Whatever the exact number, it appears that a bird's taste senses are small compared to mammals. Birds are generally accepted as having some sense of taste because taste buds are present, and because they do show aversion to some substances. Taste recognition would be an important evolutionary trait in animals, as it would likely help in detecting dangerous substances and thereby aid in finding suitable feed.

It is generally accepted that birds do not show the same preferences for certain tastes as mammals, humans in particular. For example, a number of birds often tolerate capsaicin (the compound that makes hot peppers spicy), gingerol and zingerone (the irritants in ginger) and piperine (the irritant in black pepper) (reviewed by Mason and Clark, 1995). Flavours aversive to chickens have been detected by the performance of specific behaviours such as head shaking, bill-wiping and beak-tongue movements

(Ganchrow *et al.*, 1990). It seems befitting that sweet taste is more strongly preferred by bird species that consume sweeter substances, such as nectivores and frugivores, while other birds tend to reject a sweet taste (Sturkie, 1976; Mason and Clark, 2000). Response to salty diets by birds is also variable, with some species tolerating higher salt concentrations than other species, particularly those that have salt glands (indicating their exposure to salt) such as gulls (Mason and Clark, 2000). Acidic, or sour, solutions are tolerated well by birds, but once again, species' preferences differ (Mason and Clark, 2000). Bitter flavours show no trend themselves, some evoke similar aversive responses as in humans and others are accepted. The acceptance of bitter flavours is likely associated with the bitter compounds plants produce in defence of animal consumption.

Other studies reporting taste preferences and aversions include Kare and Maller (1967) who found neither domestic poultry nor junglefowl to prefer 10% sucrose solutions to water. Jukes (1938) performed taste experiments on poultry to determine their preferences for sweet, salt, bitter and sour tastes. The unflavoured diet was consumed the most. Poultry did not show an aversion to sugars, but nor were they preferred. An increased aversion occurred with increased addition of salt or citrate to the diet. Gentle (1972) compared a variety of salts, sugars and acids in water solution and determined the chickens' preference based on the number of pecks to the drinker. Results showed an aversion to the majority of solutions and as solution concentration increased, except for sugars which seemed tolerable. Some solutions in the same taste category (e.g. sweet, salty, etc.) were preferred over others; for example, sodium salt received more pecks than potassium salt. The only solution that surpassed 50% for an extended period, indicating more than half the time was spent drinking this solution over the other, was sucrose. Although the authors stated it was significant, it does not seem that such intake levels should be considered significant since the birds didn't spend that much more time at the "preferred" solution. Tanagers were able to finely discriminate sugar concentrations of a difference of 1% up to 12% (Schaefer *et al.*, 2003), however these birds are frugivores, and may prefer higher levels of sugars and have better discriminatory preferences than chickens.

Scott and Quint (1946a) found rats showed no preference or aversion to a variety of compounds even though the authors noted that the odours were strong for human senses – diacetyl, oil of anise, monosodium glutamate, butyric acid. In this trial, the setup was well planned as both non-flavoured and flavoured feeds were given separately to the rats for a period before the choice test. That is, the rats were previously exposed to the experimental diets before testing to eliminate the effect of novelty and the cups were alternated to prevent the effect of location on choice. Balog and Miller (1989) tested whether chickens could discriminate among bitter, sour, salt and sweet tastes by flavouring diets. The unflavoured diet was consumed more than the flavoured diet, with the exception of the aspartame flavour indicating either the inability to detect a flavour or that it is very similar in flavour to the unflavoured diet. While aspartame was eaten the most of all the flavoured diets, the salt diet was the most avoided. Gentle (1972) compared the preference of poultry to various water solutions. Results showed that increased concentrations of solutions containing sodium, potassium and calcium chlorides, hydrochloric and acetic acids, as well as glucose fructose and sucrose all lead to aversion, however sugars appear to be least aversive and possibly preferred.

The chicken's ability to detect and discriminate flavours appears to be present. The flavour preference that has evolved in birds differs among species as a result of the diet they have adapted to (i.e., sweet flavours are preferred by frugivores). This ability to detect flavour and discriminate among diets has allowed for bird species to choose what they want to eat or reject what they don't. Poultry show a preference for unflavoured diets, but also show an inclination towards water solutions with sucrose, and aversions to high concentrations.

Interestingly, Hughes and Wood-Gush (1971a) noticed that feed palatability take precedence over nutrients in feed ingredient selection. Diets flavoured with the unpalatable quinine cause a rejection of calcium-supplemented diets for the unflavoured deficient diet in calcium deficient birds. It was also observed that calcium supplemented diets were of a slightly lighter colour. When colour was matched, the low calcium diet was still preferred by calcium deficient birds over the calcium sufficient quinine treated feed, indicating taste plays a larger role than visual cues.

2.4.1.3.1 The Nature of Preferences

The appetite for feed is classified based on its origin (Scott and Quint, 1946b). The simple preference has no relationship to nutritive value, that is, it is based on feed quality or characteristics such as flavour or consistency. In a human example, we could relate this to potato chips, a food not considered abundant in nutrients but one we enjoy for its flavour and texture. Learned preferences are where animals must learn from experience that a food results in a good feeling, which is often aided by the presence of a flavour to aid in associating the benefits. We could relate this to trying a mango for the first time, a nutritious food with a good taste. The result of this association would be the consumption of anything mango flavoured because we assume it has the same post-ingestive consequences. On the other hand, if mango causes you digestive upset, an aversion to mango and its flavour occurs. True hunger appetites (or preferences) are based on physiological need, and require no learning. This would exemplified by nutrient deficient rats consuming a nutrient rich food that has no flavour. Based on these classifications, Scott and Quint (1946b) considered the B-vitamin pantothenate (B_5) to be a learned appetite in deficient rats because of the flavour associated with the vitamin, unlike the B-vitamins thiamine, riboflavin, and pyridoxine. Thiamine, riboflavin and pyridoxine appetites were found not to be expressed in normal animals, indicating they are not simple appetites. But why did they not classify these as a true hunger? Since all deficient animals did not show the appetite, and because it took some time to develop the appetites, Scott and Quint (1946b) have a greater belief of the appetites being learned. Hughes (1979) supports these classifications, however, refers only to "simple" and "innate" appetites, and that such mechanisms are present in order to maintain "nutritional homeostasis" in the body. This would imply that the nutrients for which there is an appetite would be important or essential for the body and its biological functions.

Harris *et al.* (1933) found the detection of B vitamins in a diet to be a learned appetite even though rats nearly exclusively ate the vitamin rich diet when given the choice between it and a vitamin deficient diet. It was shown to be a learned appetite as Harris *et al.* (1933) could deceive the rats into choosing a vitamin deficient diet by moving the vitamin supplement from the original flavour to the other, differently flavoured, diet. Thus, the authors concluded that the choice in the diet is likely the result of the previous experience of a characteristic (in this case, flavour) of the diet and not the nutrient itself. It was also suggested by Rose and Kyriazakis (1991) that the length of this feedback to learning depends on the number of cues present to distinguish the dietary differences.

Davison and Sullivan (1963) determined the seed preferences of mourning doves by ranking one species relative to another based on the feeding behaviour of the doves when seeds of 10-20 different species were scattered in their own area. Feed was determined as choice, fair or uneaten. Doves were observed to try all seeds before choosing even if it was a seed never before encountered. It was after tasting that a seed was rejected. Preferences were chosen by the observed eagerness or neglect in eating a specific feed. Choice feeds were chosen over all others when present, while fair seeds were eaten when chosen seeds were not present. Rejected seeds were simply not eaten after the initial taste (Davison and Sullivan, 1963). Thus, the presence of an individual at a feeder does not mean they prefer that diet, as all novel food is tested. However, it has been suggested that by offering of a variety of different nutrient status diets, animals are better able to regulate their own nutritional status (Nolan *et al.*, 1996). In other words, animals are able to consume what they want based on their physiological need for nutrients at that particular point in time to better fulfill the missing gap.

Investigating preferences of feed particle size, Portella *et al.* (1988) found that the rate of disappearance of larger feed particles in crumble diets was much greater than smaller particles. The smaller particles sizes, however, disappeared faster when there was a lack of large particles. It can then be insinuated that laying hens have a preference for larger particle sizes (1.18-2.36mm) since they sort through their feed and pick out the larger sizes. This may related to the ease in manipulating the larger particles with the beak.

2.4.2 Nutrient Specific Appetites

Since silage does have an odour that may affect choice, this odour may be used to help associate it with the benefits derived after ingestion. Poultry would then learn that diets with similar identifying characteristics would be beneficial, and thus would be consumed. This implies that birds have a nutrient appetite and are seeking a particular benefit, which they can detect in diets varying in concentration. The benefit of evolving a specific nutrient appetite would be a conservation of time and effort in finding diets sufficient in the desired nutrient.

The main nutrient for which specific appetites have been investigated is calcium. Calcium is an important nutrient particularly for laying hens that are involved in virtually constant egg shell synthesis. Numerous studies have found that chickens are able to select sufficient calcium to meet their requirements (Hughes and Wood-Gush, 1971a;

reviewed by Hughes, 1979; Joshua and Mueller, 1979; reviewed by Mongin and Sauver, 1979; Classen and Scott, 1982).

Other nutrient appetites have been observed in poultry and other animals. Hughes and Dewar (1971) have also observed an appetite for zinc when deficient birds were given a selection. Although selection was observed, confounding factors in this research do not substantiate whether this is a learned behaviour. Rose and Kyriazakis (1991), and Hughes (1979), reviewed numerous studies that support the ability of both pigs and poultry to select diets based on protein and energy content. Thiamine was shown to be selected for by deprived birds (Hughes and Wood-Gush, 1971b; reviewed by Hughes, 1979). Some animals deprived of vitamin B_{12} have been shown to recognize between diets supplemented and deficient in the vitamin; for example, in rats prevented from coprophagy (Harris *et al.*, 1933). Diets insufficient in levels of B-vitamins thiamine, riboflavin, pyridoxine, and pantothenic acid fed to rats will cause the rats to choose the diets with higher levels of B-vitamins. It was demonstrated in this experiment that this choice only occurred for a certain length of time, after which they ate from both diets presumably because sufficient levels of the vitamins had been established in the body (Scott and Quint, 1946b). However, Harris *et al.* (1933) used natural sources of B-vitamins which may have imparted a flavour to be utilized for recognition, indicating a learned and not innate detection of the vitamins. Sodium-deprived birds did not show a distinct selection of adequate sodium-level diets after being placed in a low sodium diet (Hughes and Wood-Gush, 1971b; Hughes and Whitehead, 1979), but when presented with a saline solution and water, birds gradually avoid the saline solution, preferring water at all times (indicated by the amount consumed; Hughes and Wood-Gush, 1971b).

2.5 Preference Testing and Choices

Animals do have innate preferences for certain flavours (Rogers and Blundell, 1991). From studies gathering human feelings, it seems that food loses its pleasantness after time; thus, the presentation of a choice of foods would likely increase the pleasantness associated with a food, to prevent creating an aversion over time (Rogers and Blundell, 1991). A novel diet such as silage could maintain interest in the balanced feed and may propose another reason why poultry choose to eat both concentrate and silage (Johannson, 2008; Steenfeldt *et al.*, 2007). Presenting a choice also allows for individuals to meet their personal requirements, as well as allowing all individuals to maintain their requirements as they change with age (Manteca *et al.*, 2008). But, early and previous experience can also impact dietary intake through preference.

Preference testing in its simplest form is to provide individuals with a free and continuous access to two choices while assuming that the one chosen is the more preferred of the two options. Preference is also often obtained by measuring the amount of time at the feeder, or the amount of feed consumed. However, this is not necessarily the most fool-proof as more time spent may be a result of greater handling time. Equal amounts consumed is often interpreted as no preference to either food, but may indicate that the individual prefers a combination of the two options. Balog and Miller (1989) were able to demonstrate a position preference in poultry; one half of a feeder was next to a heater, and the other half was next to the drinker. One group was exposed to a rotation of feed position either every day, every other day, or not at all. Results showed that birds preferred to eat on one side of the cage, but when the diets were rotated, they were consumed equally - indicating a location preference. Positional preference was also found by Ueda *et al.* (2005) in chicks. This study also showed the use of feeder colour as a visual cue in the chicks' ability to recognize an non-aversive diet, although this was soon lost over time as the birds gradually began to eat from both feeders and recognize no difference between the two diets when the aversive agent was removed. It seems logical then to accept Hughes (1979) statement that in a preference test, the "two diets must be equally acceptable in terms of positioning and palatability".

The amount of work done to gain access to an option is an alternative method in measuring preference, where individuals will work more for a more preferred option. In other words, birds won't mind exerting more work for a greater benefit. In contrast, Forbes and Kyriazakis (1995) mention that when one option needs work to be applied so that the goal will be attained, there will be an automatic bias towards the option with the least effort thereby causing an incorrect assessment of preferred materials.

While preference tests are useful, it must also be noted that all individuals do not always choose unanimously, and that sometimes, one should be considerate of the individuals in the minority choice. As an animal grows, it makes sense for the nutritional status of the individual to change because of the physical changes occurring. Thus selection in diets will differ with age and sex in order to accommodate the changes and still meet requirements.

2.6 Concluding Remarks

The observation that poultry readily intake silage and spend more time eating when it is supplemented to a balanced diet is a remarkable occurrence. It seems more puzzling when one thinks about this choice in terms of the optimal foraging theory. According to this theory, animals face tradeoffs by choosing to eat a particular prey item or ignore it in favour of finding another. In other words, the animal chooses a food item based on the costs incurred to obtain it balanced against the benefits of ingesting the food item. Thus, a chicken that chooses to spend more time eating silage instead of partaking in other important behaviours such as maintenance, or being vigilant in evolutionary terms, seems illogical, especially since this food appears to be of low nutritive value. Thus, it must be hypothesized that poultry, which are eager to eat silage, must be obtaining some sort of benefit from it; it either contains a nutritive value that is not readily accountable, or it imparts satiety to a physiological system.

While it is not immediately certain what it is about silage that poultry appear to like, a review of the literature sheds some light on what information poultry are potentially gathering when consuming silage, and what benefits they may accrue in terms of positive feedback or nutritive value. Having explored the constituents of silage that distinguish it from other feeds, numerous questions arise. Do poultry prefer a fermentative product? Does the type of fermentation matter? Is the preference related to moisture or particle size? What is it that poultry can detect? Are they able to identify different flavours and smells? Do they prefer moist diets? What is the extent of their post-ingestive feedback detection? Can they identify the nutritional composition of the diet when either deficient or sufficient in a specific nutrient themselves? Are they able to detect changes in GIT microflora populations, their benefits, or improved cellular functions that result? Thus, the objective of the following study is to investigate what aspect of silage poultry find attractive by performing preference tests for diverse silage mixes when offered as a supplement to a balanced diet, and to determine if providing a material encompassing the preferred characteristics can impact hen welfare in terms of feather pecking.

3.0 Creating the Experimental Model and Determining Preferences

3.1 Abstract

In recent years, research on feeding silage to poultry has been conducted with positive results on bird welfare. Hens were observed to eat between 25 g and 60 g of fresh silage per day in addition to their nutritionally balanced ration. This amount is surprising since, comparatively, silage is lower in nutritional value. The objective of the following thesis was to determine which characteristic(s) of silage hens find appealing through a series of preference tests. A preference test model was developed unlike conventional preference tests in that the novel material was given in addition to the balanced ration. Feed consumption was assumed to relate to the strength of the preference. A series of experiments testing different qualities of silage (moisture, ensilation products, particle size, and forage-ability) was conducted. Results showed an increase in consumption of wet over dry products, unensiled over ensiled products, and edible over non-edible products, and a slight preference for a smaller particle size. The preferences of hens established throughout these experiments could be of use in the future for establishing an easily distributable material in poultry production for improving hen welfare.

3.2 Introduction

Feeding silage to laying hens has been shown to improve hen welfare by reducing negative feather pecking behaviour (Steenfeldt, *et al.*, 2007; Johannson, 2008). It was observed that the hens readily consumed the silage material offered, eating from

25 to 60 g as-is per hen per day. This amount of silage consumption in addition to the balanced hen ration is surprising as silage is a relatively low nutritive feed material. However, it does have some potential characteristics that could influence its intake. Silage has a high fibre content, a high microbial population which may impart a probiotic effect, compounds such as volatile fatty acids and amines that are produced during the fermentation process, and a high moisture content. These characteristics are potential attractive factors to laying hens which could account for their consumption of silage. However, distributing silage in a commercial system has challenges. Providing a high moisture diet means it is prone to spoilage, and requires specialized storage of the volume required for feeding. By determining the aspect(s) of silage poultry prefer, it may be more advantageous to incorporate this aspect(s) in an alternative feeding option. This research will focus on identifying factor(s) in silage that makes it attractive for hens to consume.

3.3 The Experimental Model

The chosen model was a preference test approach. The feeder of a cage was divided into two, proving hens with the option to consume either the balanced ration or the test material. All experiments utilized the same basic setup (Figure 1). Two cages made up a single replication. Each replication (every two cages) was divided by permanent metal dividers in the feed trough that reached a height above the trough (1.5 cm in height furthest from the cage and 4 cm closest to the cage). The dividers prevented the hens from eating from a neighbouring replication. A removable insert was fashioned from eavestroughing material in the exact shape and height of the permanent feed trough, but half the length of the replication. The insert was positioned in the middle of the permanent trough (or feeding area) of each replication. This resulted in half the feeding area in front of a single cage being the permanent trough, and the other half being the removable insert.

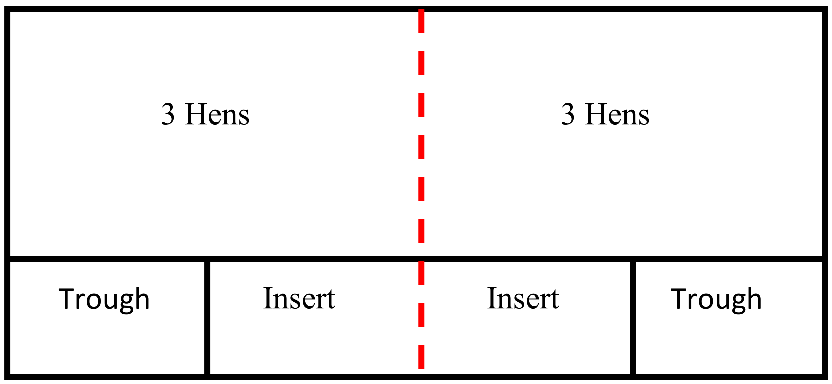


Figure 1. One replication consisting of two cages where the permanent trough was divided into two in front of each cage; the middle section was the insert used to feed test supplements and the edge sections (permanent trough) contained balanced laying hen ration. The dashed line indicates the division of the replication into two cages.

The chosen model utilized conventional cages to determine the impact of feed materials in a common North American commercial laying hen system. Lohmann LSL Leghorn hens were used in all experiments because of their common use in poultry production in Western Canada. The number of birds in a cage was modified from six (in a conventional commercial situation) to three, so that all birds were able to feed at once from either the balanced diet or the supplement if they so chose.

In this model, a direct choice was not made between supplemental feedstuffs (i.e., given the choice between silage and barley greens). This method is in contrast to a traditional choice test, where birds are often required to make a choice between two nutritionally balanced feed items. The current model was developed to permit testing of a bird's desire to eat a low nutrient supplement in the presence of a nutritious diet. The birds were given the choice between a balanced diet (nutritionally balanced with all required nutrients at sufficient levels) and a novel, supplemental feed item (not nutritionally balanced or required).

The constant presence of a balanced diet provided the nutritional requirements of the hen and made it possible to initiate the introduction of the novel supplemental feed without a preliminary adjustment period. Removal of an introductory period also eliminated the effect of previous experience on consumption. Thus, the immediate gravitation towards a novel diet may indicate either a deficiency missed in the balanced diet formulation, or demonstrate that chickens are naturally curious towards novel objects. The long term consumption of the test supplement would indicate palatability or another factor that maintained bird interest and intake. Providing a balanced diet in addition to a novel feed allowed for the hens not to become deficient in any one nutrient if they chose not to eat the novel feed offered. It also helped to demonstrate that there was no need for the birds to eat the novel item since nutrient and energy requirements could be met by solely eating the balanced diet. All supplemental (or novel) feedstuffs contained low nutrient levels and tended to be high in fibre content relative to the balanced ration.

The intake comparison between the balanced and supplemental material can be used to determine the preference of a supplemental material. The assumption made is that the most consumed material is indicative of a stronger preference for that particular supplement. Some novel feeds, may be interpreted as frightening or undesirable, and are avoided. The time frame of all experiments was 21d.

3.4 Experimentation

3.4.1 Preface

The first step in determining which aspect(s) of silage poultry prefer were to validate the experimental set-up and to decide which characteristics to investigate. Then, a series of experiments followed a sequential progression of questions, beginning with the most general and becoming more specific (Figure 2).

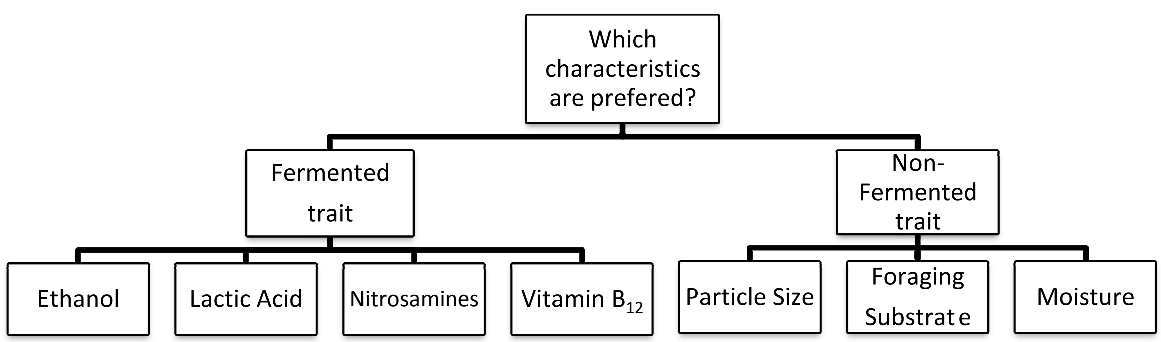


Figure 2. Scheme of sequential questions asked in regards to the preference of laying hens to consume silage.

Four experiments were performed to determine the favourable feed characteristics in silage with each experiment leading to a subsequent study. The final list of experiments was as follows, (Section 3.5):

- **Experiment 1:** Validating the Experimental Model
- **Experiment 2:** Moisture and Ensilation
- **Experiment 3:** Foraging Theory
- **Experiment 4:** Particle Size

3.5 Materials and Methods

3.5.1 General Methods

All experiments were conducted in accordance with the University of Saskatchewan University Committee on Animal Care and Supply (19940248).

3.5.1.1 Housing

Birds were housed in Specht cages (60.96 cm x 39.37 cm; forward sloping floor with 1858 cm^2 floor space per hen) in the middle row of a three tier system. Light intensity was maintained at 10 lux (at the feeder of the middle tier) and provided for 14 h per day in one continuous period. Room temperature was maintained at 21°C. The water was provided on an *ad libitum* basis by a Lubing nipple drinker in the rear of each cage. The balanced feed was also provided on an *ad libitum* basis. Three hens were placed per cage, and two cages composed one replication. Six replications were randomly allocated to each treatment. Supplement naïve birds were utilised in all experiments. Hens that had been previously housed together in the same type of cage were placed together in the experimental cages, and allowed two weeks to adjust to their new social and physical situation before commencement of the 3wk trial. Peak egg production was ensured before commencement of the first trial period.

3.5.1.2 Balanced Diet

A commercial laying hen diet in crumble form was fed in all experiments. Diets were formulated to equal or exceed the bird's nutrient requirements (NRC, 1994). The diet ingredients and nutrient content are shown in Table 1.

3.5.1.3 Laboratory Analysis

A sample of nutritionally balanced laying hen diet was collected at the onset of the trial and analysed for dry matter (DM; AOAC method 930.15, 15^{th} Edition, 1990) content to convert consumption to a DM-basis. Supplemental material was sampled at the onset of the trial (or at packaging) for initial moisture content, and at weigh back on a weekly basis (beginning on day 1 of the trial) for moisture content after the material had been in the feeder for 24hr. Dry matter content of all sample replicates were combined for a weekly average used in consumption calculations. DM analysis of supplemental, non-forage samples was conducted according to the AOAC method to determine moisture in feeds (AOAC method 930.15, 15^{th} Edition, 1990). Supplemental forage samples were treated with a two-method analysis (NTFA, 1993) where samples were partially dried at 55°C, ground, and then subjected to lab dry matter analysis (AOAC method 930.15, 15^{th} Edition, 1990).

Table 1. Composition of the balanced laying hen ration, in % except where noted

Ingredients:	Amount %
Wheat	51.983
Meat meal	5.000
Soybean meal	14.500
Corn	10.000
Limestone	9.500
Corn dried distillers grains with solubles	5.000
Tallow	2.500
Mono-calcium phosphate	0.740
Salt	0.270
DL-Methionine	0.200
L-Lysine HCL	0.130
Vitamin-mineral premix[1]	0.137
Sodium bicarbonate	0.040
<u>Calculated nutrient levels</u>	
AME (kcal/kg)	2,634
Crude protein	19.1
Crude fat	5.1
Crude fibre	2.54
Sodium	0.166
Calcium	3.92
Non-phytate P	0.38
Arginine	1.091
Isoleucine	0.695
Lysine	0.935
Methionine	0.487
Methionine and cysteine	0.808
Threonine	0.631
Tryptophan	0.223

[1]Supplied per kilogram of diet: vitamin A (retinyl acetate + retinyl palmitate),8900 IU; vitamin D_3, 2914 IU; vitamin E (dl-α-topheryl acetate), 27.9 IU; menadione, 1.4 mg; thiamine, 1.9 mg; riboflavin, 6.2 mg; niacin, 62 mg; pyridoxine, 3.1 mg; vitamin B_{12}, 12.4 mg; pantothenic acid, 12.4 mg; biotin, 155 mcg; iron, 170.1 mg; zinc, 83.9 mg; manganese, 125.5 mg; copper, 9.4 mg; iodine, 0.98 mg; and selenium, 0.23 mg.

3.5.1.4 Statistical Analysis

All statistical analysis was performed using Proc Mixed procedure of SAS, version 9.2, Statistical Analysis Systems Institute, Cary, NC. Means were compared and differentiated using Tukey's Range Test and pdmix800 was used to denote differences with letters (Saxton, 1998). Significance was established at $P \leq 0.05$.

3.5.2 Experiment One: Validating the Experimental Model

3.5.2.1 Preface

In order to feed a secondary feed item in a feed trough, without mixing feedstuffs, a removable metal insert was designed to easily slip in and out of the permanent feed trough of a conventional cage system. However, due to the novelty of the item, as well as its colour (white compared to the grey trough), it was appropriate to determine if the insert would influence feed consumption in any manner (demonstrated as any bias or preference in feeding location). It was hypothesized that hens do not show a locational preference in the feed trough, and that feed intake would not be modified as a result of feeder colour, or competition between adjacent cages at the feeder space.

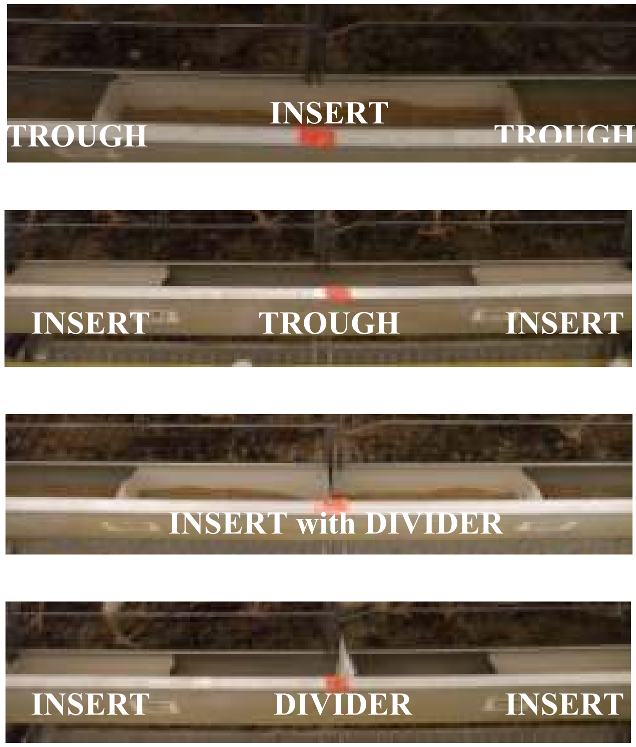
3.5.2.2 Treatments

The insert divided the feed area into two sections so that consumption in each area could be measured and thereby determine if a locational preference existed. The addition of a divider within a replication gave each cage composing the replication its own feeding area, thereby reducing the competition at the feed trough. The treatments were as follows and are illustrated in Figure 3: a single large insert located in the centre between the two cages with no divider in the insert separating cages within the replication (trt 1); two small inserts each located at the outside of the replication with no divider in the trough separating cages within the replication (trt 2); two small inserts located in the centre (inside) portion of the replication with a divider between the inserts separating the two cages within the replication (trt 3); and two small inserts each located at the outside of the replication with a divider in the trough separating cages within the replication (trt 4). The dividers were 1.5 cm (front) and 4.5 cm (back, nearest the birds) above the feeder trough and were designed to prevent birds from feeding outside their own cage area.

Balanced feed only was provided in both trough and insert areas. Feed consumption in each feed area was determined weekly and reported as average daily consumption per bird. Birds were 40 weeks of age at commencement of trial.

3.5.2.3 Statistical Analysis

The data were analysed as a completely randomized 2 x 2 x 2 factorial arrangement (insert location, dividers, and eating area). Means and standard error (SEM) were also calculated for each consumption area within each treatment (or insert placement).



Treatment 1

Treatment 2

Treatment 3

Treatment 4

Figure 3. Allocated feeder and insert treatments; treatment 1 with a single insert located in the middle of replication (composed of 2 cages), treatment 2 with inserts located on the edges of the replication, treatment 3 with two small inserts located in the middle of the replication with a divider in the insert separating the cages within a replication, and treatment 4 with inserts located on the edges of the replication and a divider in the trough separating the two cages within a replication.

3.5.3 Experiment Two: Moisture and Ensilation

3.5.3.1 Preface

The approach in this research, as mentioned previously, was to define preferences by working from general to more specific feed characteristics. The second experiment begins this process by looking at the two general characteristics of silage: moisture and the ensiling (fermentation) process. The objective of the following experiment was to determine the consumption of test materials in the presence of a nutritionally balanced laying hen ration to determine the hens' relative preference.

This experiment was completed on two separate occasions, when hens were 26 and 54 wks of age. Data were combined for analysis.

3.5.3.2 Treatments

The supplements tested in this experiment were barley silage (BS; 34% DM), barley greens (BG; 37% DM), dried barley silage (DBS; 79% DM), and dried barley greens (DBG; 86% DM). The materials selected were chosen because of their similar structure – barley could be prepared in four manners to create four different characteristics, with little physical or nutritional differences. Supplements were placed in the insert.

A control treatment was included only in the first trial period for a simple comparison of feed consumption (in section 3.6.2.1) and was not included in the statistical analysis. It consisted of hens given the balanced diet (90% DM) in both the trough and the insert.

Silage and greens were both barley based and collected from the University of Saskatchewan (U of S). The barley, all from the same field, was swathed at the mid-dough stage and separated among the ensuing treatments. Greens were defined as unfermented, not dried, barley plant material that was placed in appropriate sized packages for hen feeding (described later). The majority of the swathed material was utilized to produce silage for cattle feeding (Figure 4). The silage to be used in the hen feeding trial was collected after 6 wk of ensiling/fermentation.

Figure 4. University of Saskatchewan silage pit where material for trials was collected.

Open-wire penning raised above ground and covered with standard window screening in an empty barn wing was used to create dried greens and silage (Figure 5). The wet forages were spread out over the screen to a thickness of approximately 2.5 cm. The room's computer based heating and ventilation system was used to provide a stable room temperature of 25°C and to ensure the room was well ventilated. The forages were dried for at least 5 days before being packaged and frozen until use.

Supplements were stored in individual Ziploc bags weighed to 250 g and then placed in large plastic containers and frozen (-20°C) until use. Approximately one week prior to the trial, supplemental material was transferred to a cooler and stored at approximately 4°C for the duration of the trial.

3.5.3.3 Data Collection

Birds were fed the balanced diet once a day (at approximately 8:15 am) and the supplement was fed twice daily (8:15 am and 3:00 pm) by hand. Balanced diet and supplement intake were measured on a daily basis prior to the morning feeding.

Individual bird weights and average egg weight (the average based on weighing all eggs in a replication) were recorded the day before and the day after the trial period. Egg production was recorded to monitor hen health and production status.

3.5.3.4 Laboratory Analysis

Feed underwent DM analysis (detailed in section 3.4.1.3). The DM intake of the supplements was calculated by subtracting the leftovers (on a DM basis) from the initial fed amount (on a DM basis). The balanced DM intake was calculated using the initial sample taken before the onset of the trial as the material's moisture content did not differ after exposure to air for 24 hr.

3.5.3.5 Statistical Analysis

The balanced and supplement consumption data were analysed as a completely randomized block design (data were combined from the two experiments and blocked) with experiment as a random variable and a 2x2 factorial treatment arrangement (moisture and fermentation).

Figure 5. Aluminum penning set atop overturned garbage bins (above) with window screening rolled overtop to dry barley (below) and silage.

3.5.4 Experiment Three: Foraging Theory

3.5.4.1 Preface

It is has been proposed that feather pecking is the result of redirected foraging behaviour (Huber-Eicher and Wechsler, 1998). This idea is plausible because there is a lack of opportunity for hens to forage in a substrate other than their feed in a commercial cage setting. Previous studies conducted in this area have found that birds provided with a foraging material show a lower rate of feather pecking (Huber-Eicher and Wechsler, 1998). Providing environmental enrichment device other than foraging material also has a similar reduction in feather pecking in hens (Sherwin, 1995; Jones *et al.*, 2002; McAdie *et al.*, 2005).

Previous experimentation in the present thesis showed that laying hens highly scattered, but barely consumed dried materials such as dried barley greens (DBG) and dried barley silage (DBS). This led to the hypothesis that the birds find the dried material to be a suitable foraging and/or novel enrichment substrate. Observing behaviour at the feeder would determine the amount of time the birds spend with this material in comparison to the barley (wet and dried). It can then be determined whether or not the birds are fulfilling a nutritional or behavioural need with the silage supplement. The objective of this study was to determine whether laying hens spend time with a non-edible foraging material. It was hypothesized that performing foraging behaviour is a reward in itself.

3.5.4.2 Treatments

Supplemental materials in the third experiment consisted of: control (balanced diet), wet barley greens (BG), dried barley greens (DB), yellow plastic lace (YL), and white plastic lace (WL). A balanced diet was provided in addition to the supplemental material. Birds were 58 wk of age at the onset of Experiment Three.

3.5.4.2.1 Supplements

Barley greens were collected in July, 2009, and frozen until needed. Prior to the experimental period greens were pre-packaged into small re-sealable plastic bags and stored at 4°C. Dried barley remaining after the first supplement experiment (see section 3.4.3.1 for details) was collected and frozen until use. A plastic lace, similar in form to string (0.24 cm width), was obtained and cut into pieces measuring approximately 75 cm long. Forty-five pieces were grouped together and tied in the middle. When placed in the insert, each end of one length of lace was fed through the insert where joins occurred and knotted to prevent the loss of the entire lace bundle from the insert. Both white and yellow colours were used as published studies showed that hens prefer these colours over others such as red, blue, and green (Jones and Carmichael, 1998). Supplements are illustrated in their experimental setup in Figure 6.

Figure 6. Photos of supplemental items given in removable insert; white plastic lace (top left), wet barley greens (top right), yellow plastic lace (bottom left), dried barley greens (bottom right).

3.5.4.3 Data Collection

Birds were fed the balanced diet once a day (at approximately 8:00 am). The barley greens supplement was replaced once daily at the morning feeding time to an amount sufficient for *ad libitum* consumption. The dried barley was replenished as necessary. Feed intake was measured on a weekly basis by weighing the remaining balanced feed in the trough and can. Supplement intake of barley greens and dried barley greens were not measured. Egg production was monitored for supervising hen health.

Behaviour observations occurred three times (days 2, 9, and 16 of the trial). The number of birds partaking in activities in the feed trough and the insert were recorded by instantaneous scan sampling at 1 min intervals over a 10 min period (collection taking no more than 10s). The observer sat in a raised chair to allow clear observation of hen behaviour. Five minutes were allotted for adjustment after each move of the observer. Partaking in activity in a particular feeding location was defined by the bird having its head lowered in the respective feeding area with the beak no higher than trough level and the bird's attention focused downward, towards the feeder. Data were collected in two sessions; morning (9:00am to 12:00pm) and afternoon (2:00pm to 5:00pm). In each session, all replications of all treatments were observed. Three replications were observed at a time, with each grouping of three being observed in a random order each day and session to reduce the effect of time.

3.5.4.4 Statistical Analysis

The data were treated as a completely randomized design. The means and standard error (SEM) were calculated for each treatment for balanced feed consumption (on a dry matter basis) and time spent in activity in each feeding area.

Time spent at each feeder location (trough and insert) was set as the percent of time spent at each feeder, before being log+1 transformed and undergoing analysis as a randomized complete block design to minimize the effect of day.

3.5.5 Experiment Four: Particle size

3.5.5.1 Preface

Barley greens and barley silage were observed to have variable lengths of plant material. As a result of the hens tossing about this same material in the previous experiment, it was questioned as to whether or not the hens were actively sorting out the material, that is, tossing out one size of material and consuming another. As larger sized items generally take longer to break down in the digestive tract compared to materials of a smaller size, post ingestive feedback could result in the hens making a preference to eat one size over another. This led to investigating a preference for particle size.

3.5.5.2 Treatments

The following supplements were given in addition to the balanced diet: Control (balanced diet), whole oats (WO), finely ground oats (GO), cubed alfalfa (CA), ground alfalfa (GA), barley silage (BS), ground/mulched barley silage (GBS). The supplements were a variety of easily obtained, higher fibre, and lower nutritive materials. Birds were 63 wk of age at the onset of the trial period.

3.5.5.3 Supplement Preparation

Silage and ground silage were collected one week prior to the onset of the trial period and frozen until needed. Ground silage was obtained by placing the same silage to be used whole, on a cement pad and using an electric lawn mower to chop up the forage. Two wooden boards were placed on their length on either side of the mower to prevent the silage from scattering (Figure 7). The silage was mown for approximately 3 minutes over an area approximately two meters long and 20 cm wider than the mower itself. The particle size of the silage before and after mowing is shown in Table 2. Particle size was determined using a Penn State forage particle separator (3 pans instead of 4 as in Heinrichs and Kononoff, DAS 02-42).

Ground oats were obtained by grinding whole oats to a fine grind with a hammermill (Jacobson Ajacs Hammermill; model 170F8 series, fine screen 30 mm perforations). Cubed alfalfa was sourced from Early's Farm and Garden Centre (Saskatoon, SK) and originally derived from Elcan Forage Inc. (Box 55, Broderick, SK). A portion was ground using a full circle pulverator hammer mill (Model 160-D, Jacobson Machine Works, Minneapolis, MN, USA; 12.7 mm screen). These supplements were kept at room temperature. All supplemental materials are shown in Figure 8.

Table 2. Particle size proportions of whole and ground silages sorted using a Penn State forage particle separator

%	Whole Silage	Ground Silage
>19 mm	5.48	1.58
8-19 mm	70.39	73.91
<8 mm	24.13	24.51

Figure 7. U of S dairy barn barley silage where silage was collected for the particle size experiment (above) and mulching (grinding) of silage (below) to finer size using and electric lawn mower using wooden boards to prevent silage from escaping.

Figure 8. Supplemental materials fed to hens; whole oat grains (top left), finely ground whole oat grains (top right), barley silage (middle left), mulched/ground barley silage (middle right), cubed alfalfa (bottom left), and ground cubed alfalfa (bottom right).

3.5.5.4 Data Collection

Birds were fed the balanced diet and supplements daily at approximately 8:00 am. The silages were fed a second time at approximately 3:00 pm. Supplements were fed at levels sufficient for *ad libitum* consumption. Egg production was monitored to monitor hen health and production status.

Behavioural data were collected occurred three times (days 3, 10, and 17 of the trial). The number of birds partaking in activities in the feed trough and the insert were recorded with the same methods as Experiment 3 (Section 3.5.4) with the change that in each session, 3 replications per treatment were observed.

3.5.5.5 Statistical Analysis

The data were treated as a completely randomized design using the following *a priori* contrasts for supplement comparisons; WO vs. GO, BS vs. GBS, CA vs. GA. The means and standard error (SEM) were calculated for each treatment for balanced feed consumption (on a dry matter basis) and time spent in activity in each feeding area. Time spent at each feeder location (trough and insert) was established as the percent of time spent at each feeder, before being log+1 transformed (since data were not normally distributed) and undergoing analysis as a factorial design to determine an effect of day. The effect of day was present, thus, data were blocked by day to account for this effect.

3.6 Results and Discussion

3.6.1 Experiment One: Validating the Experimental Model

Average diet DM content was calculated to be 89.5% and the average dry matter consumption is shown in Table 3. Feeding location (eating area) was found to be significant; birds ate more from the inside than the outside feeding areas. All other main effects and interactions among them were not significant.

Table 3. Average dry matter feed consumption (grams per bird per day) of birds given different feeder positions and/or dividers

	Eating area		Insert location		Presence of Dividers		SEM
	Inside	Outside	Inside	Outside	Dividers	No Dividers	
Feed Consumption	59.49^{A}	48.70^{B}	53.64	54.56	54.43	53.76	0.7779

^{A,B} Means with different letters in the same row are statistically different ($P \leq 0.05$).
SEM – standard error of the means.

The hens were not alarmed to the presence of inserts since feed consumption did not decrease in areas where the insert was located. Intraspecific competition was not a factor that would influence where birds preferentially choose to eat as there was no increase in feed consumption in the middle areas when dividers were present. The preference of all birds to consume more feed in the middle (or inside) is either an arbitrary occurrence, or the result of another factor not evident. Thus, it can be concluded that the inserts have no effect on feed consumption of laying hens.

3.6.2 Experiment Two: Moisture and Ensilation

3.6.2.1 Feed Consumption

Whether or not the supplements were fermented did not affect the consumption of balanced diet and the interaction between moisture and fermentation treatments was not significant (Table 4). Consumption of the supplement (on a dry matter basis) was affected by moisture and fermentation main effects and their interaction was also significant. Birds ate the most BG, followed by BS, while birds given dried supplements ate little to no material at all. Birds fed BG and BS in a moist form ate less balanced diet and more supplement in comparison to birds fed these supplements in dry form. Supplement moisture level affected total dry matter intake (the combination of the balanced feed and supplement). Birds given DBG and BS consumed more feed that those given BG and BS. Fermentation did not affect total dry matter intake, and there was no interaction between the main effects.

Table 4. Average balanced, supplement and total consumption of hens on a dry matter basis (g per bird per day) given a supplement of barley greens, dried barley greens, barley silage, or dried barley silage for a three week period. The supplement relates to the insert feeding location, the balanced consumption refers to the outer trough location, and total consumption refers to the combination of these locations

Consumption	Barley Greens	Dried Barley Greens	Barley Silage	Dried Barley Silage	SEM
Balanced diet[1]	85.04	93.93	84.84	97.89	3.6291
Supplement[2]	4.54^{A}	0.19^{C}	2.28^{B}	-0.20^{C}	1.6229
Total Intake[3]	89.59	93.83	87.13	97.40	2.3322

[1]Significant moisture effect.
[2]Significant fermentation and moisture, and interaction effects.
[3]Significant moisture effect.
[A,B] Means with different letters in the same row are statistically different ($P \leq 0.05$).
SEM – standard error of the means.

The factorial design used in this trial permitted the ranking of the supplemental materials in order of preference between moisture and ensiling products (as indicated by a significant interaction P-value). It appears that hens prefer a moist product over a dry product whether it is ensiled or not, but prefer a non-ensiled material over an ensiled one based on the intake of the high moisture supplements.

Less BS was consumed than was found in Johannson (2008) and Steenfeldt *et al.* (2007) trials (3.35 and 7.16 g in the current trials as compared to 13-14 g or more in the published studies, on a dry matter basis). This could be the result of different housing systems as the current studies are in conventional cages, while the published studies were conducted with enriched cages (grouped, enriched cages; Johannson, 2008) and litter pens (Steenfeldt *et al.*, 2009). The social dynamics and different stressors in these group housing units could influence the feed consumption where birds housed in large groups experience more stress (Mashaly *et al.*, 1984; Onbasilar and Askoy, 2005), thus motivating them to consume more supplemental material (Siegel and van Kampen, 1984; Nasir *et al.*, 1999). In addition, Johannson (2008) also used a different strain, which may have affected the results (Shaver White as compared to the Lohmann LSL in the current study). Johannson (2008) also provided a large particle calcium source separate from the balanced diet and the supplemental barley silage. This could have impacted intake as this grit would aid breakdown of fibrous materials in the gizzard, allowing easier passage and digestion and therefore greater intake if it were found to have positive post ingestive feedback. Another difference could be the silage itself. No two batches of silage turn out identical, the silages from Johannson (2008) and the present experiment could differ compositionally due to effects of the originating barley and silage production, or environmental differences.

Daily average supplement consumption per bird (grams of DM) was plotted to determine if there was an innate or learned preference for the materials. A control group was included for comparison, but was not included as part of the experimental trial. Control daily intake (balanced feed placed in insert) plotted over time (Figure 9) shows a stable intake over the trial period. An innate preference would be demonstrated by an immediate and relatively constant high consumption of the material, resulting in consumption similar to the control diet. The control diet, however, is not representative of a true innate preference since the hens had been previously exposed to the diet before the trial period.

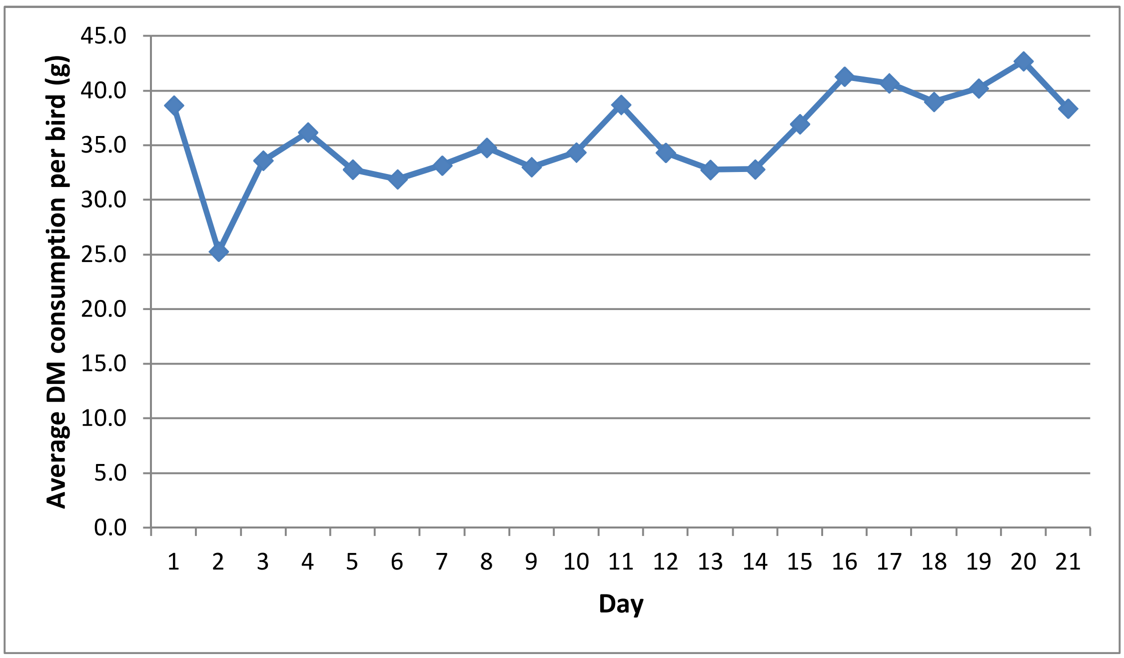


Figure 9. Control average daily dry matter intake of balanced feed placed in the removable insert (grams per bird per day) as measured over the three week trial period.

The daily intake of barley greens plotted over time (Figure 10) shows an initial consumption of approximately 3 g (DM intake) with a gradual increase in consumption over several days. After 7 days of the experiment, consumption remained relatively stable until the end of the trial period.

Barley silage supplement average daily intake plotted over the trial period (Figure 11) shows no consumption at the onset of the trial period, but is followed by an

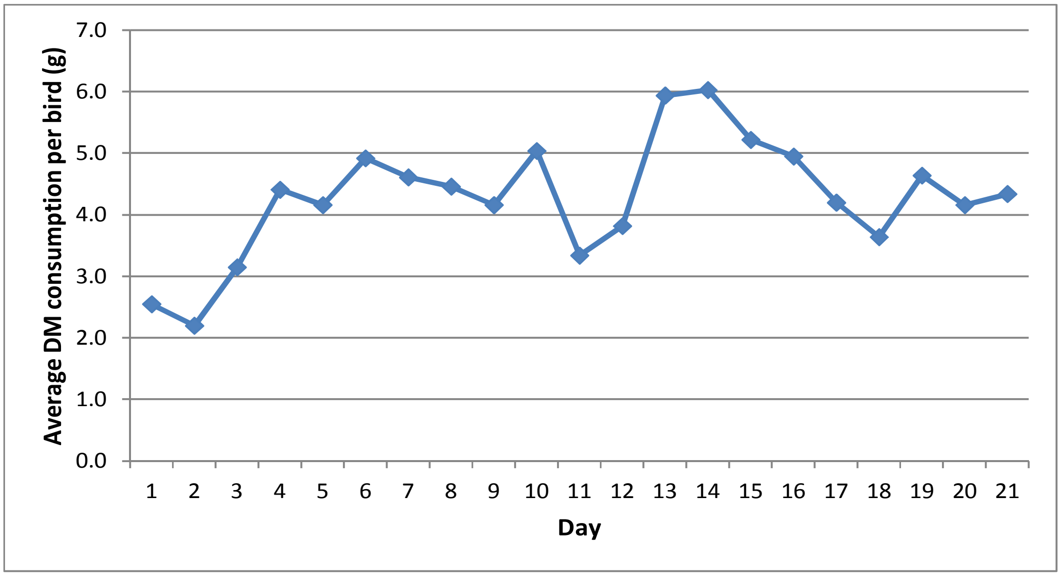


Figure 10. Barley greens average daily dry matter intake of balanced feed found in the removable insert (grams per bird per day) as measured over the three week trial period.

increase in consumption until day 5 at which point consumption remains relatively constant for the remainder of the experimental period. Thus, we see that some supplements, such as BG and BS, show increasing intake before becoming somewhat stable, indicating that ingestion, thus preference, for the material grew with time.

Daily consumption patterns of DBG and DBS are shown in Figures 12 and 13. The data demonstrate that hens do not show a preference for dried supplement through a variable pattern. Dried barley green consumption shows an overall decrease in intake over time, opposite to BG, indicating that a learning period was needed for hens to find the material unfavourable. The negative numbers may relate to the ability of such materials to pick up moisture from the birds and/or environment thereby increasing the weight. Perhaps, this incident results from balanced feed in the neighbouring trough being spilled into the insert because of bird feeding activity. This could indicate that the

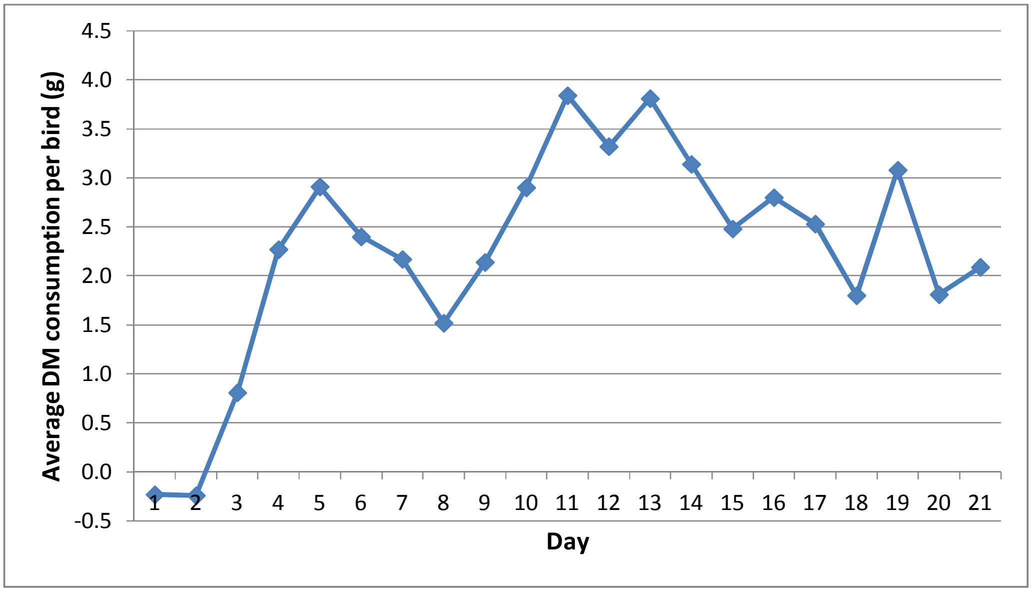


Figure 11. Barley silage average daily dry matter intake of balanced feed found in the removable insert (grams per bird per day) as measured over the three week trial period.

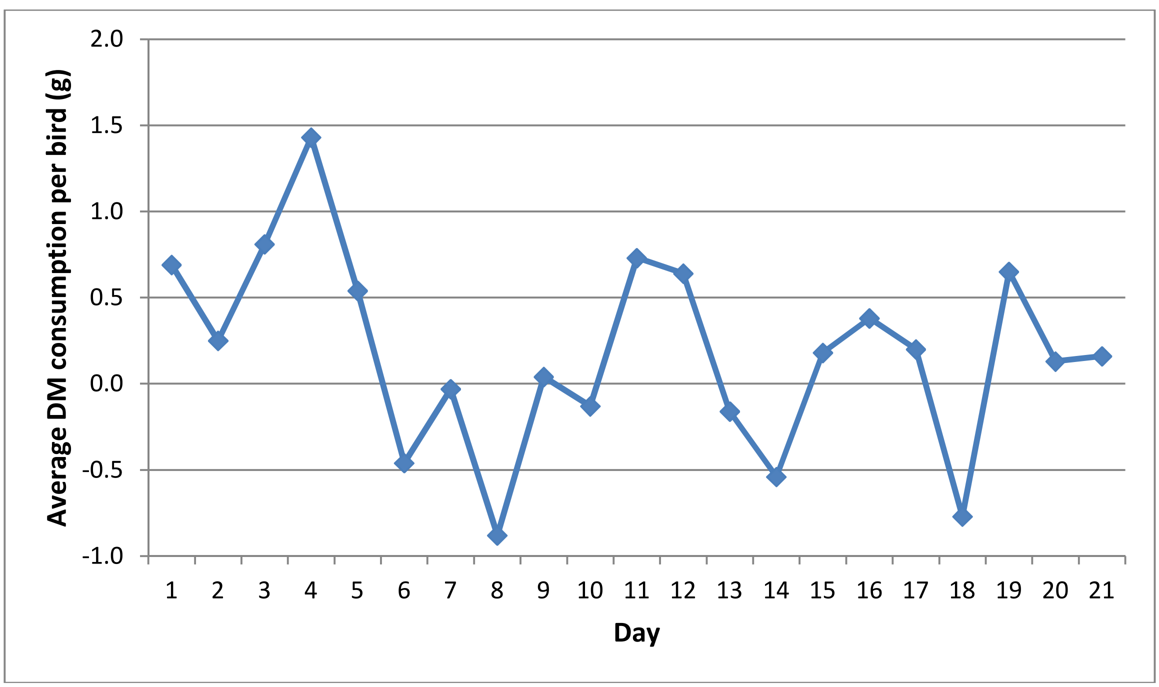


Figure 12. Dried barley average daily dry matter intake of balanced feed found in the removable insert (grams per bird per day) as measured over the three week trial period.

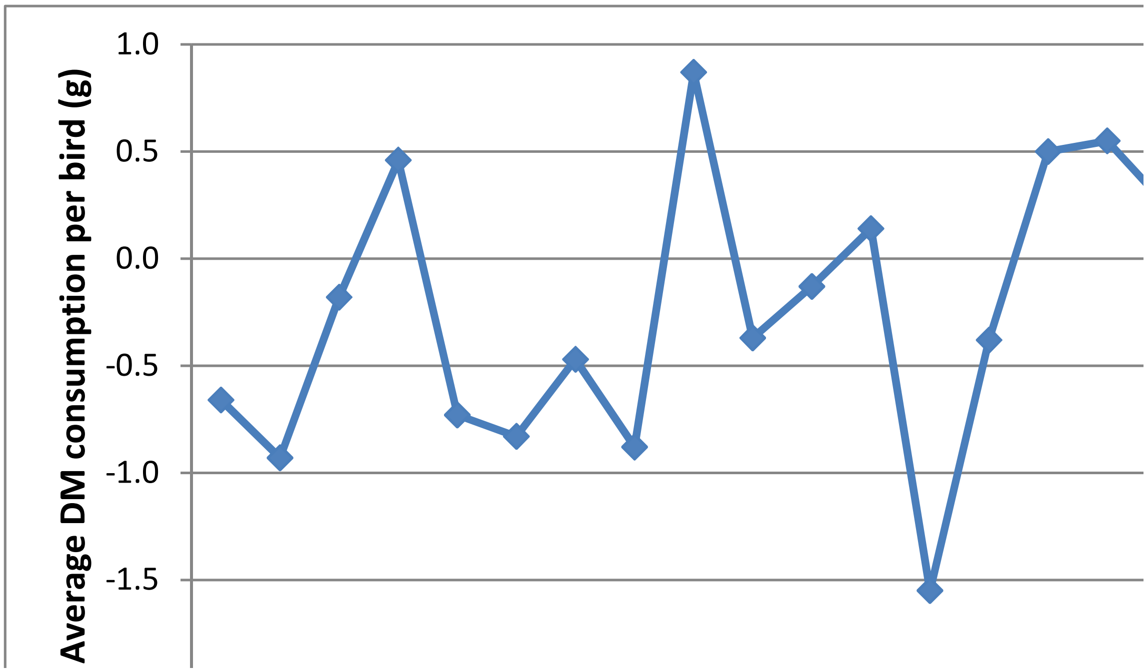


Figure 13. Dried silage average daily dry matter intake of balanced feed found in the removable insert (grams per bird per day) as measured over the three week trial period.

experimental design is flawed, however, scattering was for the most part approximately equal among all supplements. In subsequent trials the amount of supplement allocated was reduced, however, this tossing of the feed did promote ideas of hens foraging in the supplemental materials.

3.6.3 Experiment Three: Foraging Theory

3.6.3.1 Behaviour

Behavioural data are shown in Table 5. Percent of the observed time which hens spent at the insert (supplement) is greatest in hens given a balanced diet (the controls), followed by those given BG and DBG (which did not differ between each other), followed by hens given white and yellow plastic lace (which did not differ between each other). The percent of observed time spent at the trough (balanced diet in all treatments) was greatest in birds given BG as a supplement but did not differ from birds given other supplements (YL, WL, DBG). Control birds spent the least amount of time at the trough area, which differed from all the other treatments. Of the total time spent at the balanced feed (the trough area of supplement fed birds, but both the insert and trough area of control birds), control birds spent the least amount of time. This did not differ significantly from those given DBG or white and yellow plastic lace, but did differ from birds given BG (which did not differ from birds given DBG, WL, and YL). The total percent of observed time hens spent at the entire feeding area (the combination of both insert and trough feeding areas) was highest in birds given the BG supplement but did not differ from those given DBG. DBG hens spent the second highest amount of time at the feeding area, but this did not differ from hens given white plastic lace. In turn, hens given white plastic lace did not differ from those given yellow plastic lace but was higher than for the control hens. Values for hens given YL did not differ from the control birds.

Table 5. Behaviour collected from laying hens as percent of time spent at either, the insert feeding area, trough feeding area, time with balanced feed, or time at the general feeding area for birds given barley greens, dried barley greens, white plastic lace, yellow plastic lace, or balanced diet (control) as a supplement to a nutritionally balanced laying hen ration in the trough

% Time	Control	Barley Greens	Dried Barley Greens	White plastic lace	Yellow plastic lace	SEM
Insert	8.15^{A}	4.73^{B}	5.37^{B}	1.54^{C}	1.36^{C}	0.1309
Trough	10.15^{B}	21.31^{A}	18.41^{A}	19.90^{A}	18.63^{A}	0.1008
Balanced Feed	18.30^{B}	21.31^{A}	18.41^{AB}	19.90^{AB}	18.63^{AB}	0.1190
Feeding Area	18.30^{D}	26.04^{A}	23.78^{AB}	21.44^{BC}	20.00^{CD}	0.1370

^{A,B,C,D} Means with different letters in the same row are statistically different ($P \leq 0.05$).

3.6.3.2 Feed Consumption

Feed consumption of the balanced laying hen ration is seen in Table 6. Birds with no enrichment supplement ate the most balanced feed, but the amount was no different than for hens given WL, YL, or DBG. Hens receiving BG as a supplement ate less than other treatments but this was not significantly so for the DBG and YL treatments.

Table 6. Average balanced feed consumption (g per bird per day) of laying hens receiving barley, dried barley, white plastic lace, yellow plastic lace, or balanced diet (control) in the supplemental insert

Supplement	Control	Barley Greens	Dried Barley Greens	White Plastic Lace	Yellow Plastic Lace	SEM
Balanced ration consumption	103.44^{A}	93.88^{B}	98.45^{AB}	102.11^{A}	100.57^{AB}	1.9775

^{A,B} Means with different letters in the same row are statistically different ($P \leq 0.05$).

It was observed that hens do not avoid novel objects since hens spent time with all materials given in addition to the balanced feed. While the hens preferred a novel material with a nutritive value (BG, DBG), as indicated by a greater amount of time spent with such material, non-nutritive materials were not ignored. The hens did spend some time with the plastic lace, thereby stimulating their behavioural repertoire. The relatively greater focus on nutritive materials indicates that a reward for behavioural effort (nutrients) is more appealing. This finding supports the redirected foraging theory as a potential mechanism for feather pecking since in a wild setting, chickens would forage in various substrates for materials such as rocks for improved gizzard function, or feed items. Edible materials provide a reward for foraging behaviour, and act as reinforcement for foraging behaviour.

The hens find materials with greater nutritional value more attractive as indicated by the time spent with the novel materials. In turn, hens receiving supplements with nutritive value (BG = DBG) spent more time at the feeder (trough and insert) than birds from other treatments (YL and WL). The question remains whether the increased time occupied in activity at the feeder is the factor which reduces feather pecking in hens when fed silage (Steenfeldt *et al.*, 2007; Johannson, 2008). If we put this in practical terms, a change in behaviour of 1% is equal to 8.4 min in the 14 hours of light given. So, hens given supplements (BG and DBG) with nutritional value spend a total of 6 - 8% more time at the feeder, or approximately 50-67 min compared to hens given only a balanced diet. While the WL and YL treatments only increase their time at the feeder by approximately 2 – 3%, or 17 – 25 min compared to hens only eating a balanced feed, and less than half the extra time compared to BG and DBG treatments. This large increase in time should therefore have an impact on which activities a hen participates in each day. If an additional hour is spent feeding or foraging at the feeder, this is one hour less the hen could be occupied inflicting damage to a cage mate through feather pecking. The aspect of activity replacement through enrichment should be an area for further study in future research as a potential mechanism of reducing feather pecking.

3.6.4 Experiment Four: Particle size

3.6.4.1 Behaviour

Behavioural data (Table 7) show hens spent more time with GBS and GO over BS and WO, respectively. No differences were seen with particle size in total time at the feeder and time spent with the balanced feed. Control birds spent more time at the insert and less time at the trough, but the same total time spent at the feeder compared to supplement fed birds.

3.6.4.2 Feed Consumption

Greater supplement and total dry matter intake was only found with GO over WO (Table 8). All hens given supplements ate less total dry matter compared to control birds except for GO supplemented birds.

Balanced feed was consumed the most by birds given the ground oat supplement. This did not differ from the consumption of balanced feed by hens given the WO, GA, CA and BS supplements. Balanced feed consumption by hens given GBS did differ from hens given GO, but it did not differ from WO, GA, and CA.

Table 7. Percent of the observed time hens spent at the insert, the trough, at the area containing balanced feed (trough in supplement fed birds, and trough plus insert in controls), and at the total feeding (insert plus trough feeding areas) area when given cubed alfalfa, ground alfalfa, silage, ground silage, whole oats, ground oats or balanced diet (control) as a supplement to a nutritionally balanced laying hen ration in the trough

Supplement	Control	Silage	Ground Silage	Cubed Alfalfa	Ground Alfalfa	Whole Oats	Ground Oats	Contrast	P-value
% time observed at insert	12.71^{A}	2.98^{CD}	6.21^{B}	0.47^{E}	1.78^{DE}	2.40^{CDE}	4.12^{BC}	S vs GS CA vs GA WO vs GO	0.0052 0.0583 <0.0001
% time observed at trough	8.10^{C}	16.37^{AB}	16.95^{AB}	16.48^{AB}	12.64^{B}	17.37^{A}	17.54^{A}	S vs GS CA vs GA WO vs GO	NS 0.0812 NS
% total time at feeding area	20.81^{AB}	19.35^{AB}	23.16^{A}	16.95^{B}	14.42^{B}	19.76^{AB}	21.67^{A}	S vs GS CA vs GA WO vs GO	NS NS
% time observed at balanced feed	20.81^{A}	16.37^{AB}	16.95^{AB}	16.48^{AB}	12.64^{B}	17.37^{A}	17.54^{A}	S vs GS CA vs GA WO vs GO	NS 0.0852 NS

$^{A,B,\ C,\ D,\ E}$ Means with different letters in the same row are statistically different (P≤0.05).

Table 8. Average dry matter consumption (grams per bird per day) of a nutritionally balanced laying hen ration and supplemental material (on a dry matter basis) of hens given cubed alfalfa, ground alfalfa, silage, ground silage, whole oats, ground oats or balanced diet (control) as a supplement to a nutritionally balanced laying hen ration in the trough

Consumption	Control	Silage	Ground Silage	Cubed Alfalfa	Ground Alfalfa	Whole Oats	Ground Oats	SEM	Comparison	P-value
Balanced	58.02^{C}	92.10^{AB}	89.72^{B}	93.68^{AB}	93.57^{AB}	92.00^{AB}	95.49^{A}	0.9992	--	--
									S vs GS	NS
Supplement	40.20^{A}	2.35^{B}	2.78^{B}	-1.00^{D}	-0.11^{CD}	-1.15^{D}	1.21^{BC}	0.4062	CA vs GA	0.1197
									WO vs GO	<0.0001
									S vs GS	0.1281
Total Intake	98.22^{A}	93.38^{BC}	92.49^{C}	92.68^{C}	93.46^{BC}	90.84^{C}	96.70^{AB}	0.3772	CA vs GA	NS
									WO vs GO	<0.0001

^{A,B,C,D} Means with different letters in the same row are statistically different ($P \leq 0.05$).

While it appears that there are negative consumption rates, this is not entirely true. This simply indicates that more mass was present after 24hr than before, indicating that moisture from the air or birds was picked up by the dry material, or that the hens managed to toss some of the balanced feed into the insert (as indicated by the observed presence of balanced feed mixed with the supplemental material).

It appears that particle size has limited impact on consumption but increases the time spent with the supplement in laying hens. Birds appear to show interest in the novel feed items indicated by the time spent at the insert, however numerically, not as much as the balanced feed (also seen in previous experiments discussed). The hens spent more time with the smaller particle size in silage and oat supplements, but only oat fed birds consumed more of the smaller particle size. This indicates perhaps the hens are foraging more in the material. A smaller size may be more preferable because it is more manageable (in terms of handling with the beak) and thus easier to eat than a larger size.

3.7 Conclusion

The initial part of the current research dealt with an investigation into the laying hens' preferred characteristics of silage to account for the unexpected intake of this material which was linked to decreased feather pecking behaviour (Steenfeldt *et al.*, 2007; Johannson, 2008). The approach taken was preference testing and model development. The model created allowed for the hens to choose between a balanced hen ration and a novel and relatively low nutrient material instead of having to choose between two novel materials as in traditional preference testing models. The characteristics investigated include moisture, fermentation, particle size, nutritive value and foraging substrate. Hens consumed more wet than dry material particularly when it had not undergone ensilation (thus fermentation processes). Hens also spent more time with foraging substrates of nutritive value (edible materials), particularly if it is of a smaller particle size. It was discussed that the presence of a highly preferred material in addition to the typical hen ration has the potential to increase the hens' time spent engaged in activity at the feeder by up to 1 hour which may have an impact on the hens' other activities in a day. The next chapter of the present study delves into the area by taking a preferred material (incorporating the characteristics from the initial experiments) and examining its impact on feather pecking behaviour by observing behaviour as well as feather condition and coverage.

4.0 Application: Does a preferred material influence feather pecking?

4.1 Abstract

Recent studies showed the positive impact of feeding silage to hens on their welfare by reducing feather pecking behaviour. The previous chapter dealt with investigating the reasons for the observed high affinity of hens for silage. It was observed that hens preferred moist, unensiled, and edible materials of a small particle size. Hence, it was hypothesized that the provision of a feed item with the previously determined preferred characteristics in addition to a balanced ration, should reduce the occurrence of feather pecking and thereby improve feather condition and coverage. To test this hypothesis, pea fibre was fed in a preference test model as a supplement to hens housed in conventional (3 or 6 birds per cage) and enriched cages (20 birds per cage). Pea fibre was a material which possessed all the characteristics previously determined to have been preferred by hens. The two housing systems were treated as separate experiments and not compared statistically. Results showed that birds given the supplement (wet pea fibre) voluntarily consumed approximately 13g dry-matter of the supplement per bird per day and ate less balanced feed, but had a higher dry matter intake than control birds in both housing systems. Bird housed at 6 hens per cage ate more pea fibre than hens housed 3 per cage in conventional cages, 11g compared to 14g per bird per day (DM basis). This may be the result of increased stress from crowding, or increased competition for balanced feed. Pea fibre consumption decreased in quadratic fashion with time for birds housed in enriched cages, while consumption in conventional cages increased linearly. Lower body weights in supplement fed birds were

unrelated to improved efficiency in terms of feed per dozen eggs. No effect on egg production was found. Pea fibre consumption increased proventriculus and gizzard weight and ileum length. Feather pecking decreased when supplemental wet pea fibre was provided in enriched cages. Feather score showed no significant difference given supplemental material. Providing laying hens with supplementary pea fibre did not impact welfare parameters as hypothesised, however, this could be due to a number of factors such as bird age and strain. It was apparent that birds accepted the material as it was observed they readily consumed the wet pea fibre. This indicates that the feed characteristics chosen are highly preferred by hens. Future research should investigate the effects of providing supplemental material in different environments and examine the effect of supplemental provision on hen efficiency.

4.2 Background

Feather pecking is common in laying hen flocks. It is commonly managed through the use of low light intensity (Hughes and Duncan, 1972; Kjaer and Vestergaard, 1999), beak trimming and selective breeding of hens for reduced feather pecking (Kuo and Craig, 1991). A variety of dietary factors have also been manipulated to alter feather pecking behaviour. van Kripmen *et al.* (2005) reviewed the topic of manipulating the balanced diet by describing research involving tryptophan supplementation to increase serotonin levels in the body, as well as low energy diets, different feed forms, and fibre content to increase the amount of time spent feeding.

Recently, two studies (Steenfeldt *et al*., 2007; Johannson, 2008) have shown reduced feather pecking in birds presented with silage in addition to and separate from to their usual balanced ration. The voluntary consumption of the silage in these two studies prompted the investigation into silage characteristics preferred by laying hens and is reported in the previous chapter.

The experiments in the previous chapter shed some insight on what feed and material characteristics laying hens prefer to eat relative to others. It was observed that birds prefer edible materials with a high moisture content. Materials that have been ensiled or are non-edible enrichment objects are not necessarily rejected, but are less preferred, as shown by amount of edible material consumed and time spent at the feeding area, respectively. Although not definite, the research also suggested that hens appear to prefer a feed that is small in particle size. The question remains whether or not an attractive supplemental feed can increase the time at the feeder. The increased time at the feeder would take a hen's focus away from negative activities like feather pecking, reducing feather pecking directed towards conspecifics.

The final experiment described in the current chapter applies the general preferences demonstrated in the previous experiments (see Chapter 3.0). The objective was to use a low nutritional-value food (in this instance, pea fibre) that meets all previously determined criteria and to determine its impact on laying hen behaviour so that damage as a result of feather pecking is reduced.

4.3 Materials and Methods

4.3.1 Experimental Design

Two experiments (differed by housing type) were conducted to explore the impact of providing supplementary wet pea fibre with a commercial nutritionally balanced diet on feather pecking behaviour, feather score, and accordingly laying hen welfare in comparison to hens only provided with a nutritionally balanced diet. Both experiments were conducted simultaneously and will be discussed together, but were analysed separately. The first experiment was conducted in conventional battery cages using the same preference model previously devised (section 3.3). The conventional housing system compared two housing numbers (3 and 6 per cage), thus providing two feeder space allowances. Three birds were chosen to reduce any repercussions resulting from competition at the feeder, while six birds were chosen to mimic commercial conditions. The second experiment, conducted simultaneously, used enriched cages (nesting boxes and perches provided) with 20 hens and 2 roosters per cage to provide a better comparison to two previous studies (Steenfeldt, 2007; Johannson, 2008). The enriched cages also were used to replicate the high feather pecking levels noted previously in these cages by Schwean (1995). Data were analysed separately but will be presented and discussed concurrently.

4.3.2 Housing

Lohmann LSL pullets were housed in floor pens with wheat straw litter from hatch until 18 wk of age. At 18 wk of age, pullets were placed into Specht battery cages (60.96 cm x 39.37 cm; forward sloping floor with 1858cm^2 or 929cm^2 per hen at 3 and 6 hens per cage respectively, in the middle row of a three tier system. Water was supplied *ad libitum* via Lubing nipple drinkers located in the rear of each cage. The housing environment was kept similar to commercial conditions; temperature was maintained at 21°C and light was provided for 14h per day at an intensity of 10 lux. Mortality was recorded and hens were removed when signs of illness or cannibalism were evident. Removed birds were not replaced to prevent the changing of group dynamics from affecting production and behavioural characteristics.

Also at 18 wk of age, 20 pullets were placed in enriched cages (Figures 14 and 15), along with 2 roosters. These cages were locally manufactured (1.2m wide, 1.8m long, 1.2m high) and located above a shallow manure scraping track. Housing density in these cages was 982cm^2 of cage floor space per bird (not counting nest space). The floor consisted of a 75% open plastic grid. Separate panels of horizontal bars extend the length of the cage at floor level at the front and rear of the cage, as well as at the front of the cage at perch level. Two feeders were attached to the horizontal bars and were located at the front and rear of the cage at floor level. Three wooden perches were located at a height of 0.6m from the floor and ran along the length of the cage. The perches were spaced 0.38m from each other and allowed 16.2cm of roosting space per bird. Metal roosts were located in the front and rear of the cage to facilitate eating (no feeder at this level in this trial) and nest entry. Water was provided by nipple drinker lines (4 nipples per line) hung underneath the two outside perches. A roll-away nest box (1.2m long and 0.54 m wide, 7° floor slope) was located at the rear of the cage (0.6m above the cage floor). The floor of the nest box was composed of a 2.54cm x 5.08cm wire mesh covered with plastic, non-backed pliable broiler breeder Astroturf. Solid wall dividers visually separated each cage.

In the conventional cages, 3 pullets were placed per cage in half the cages, and 6 in the remaining half, with two cages composing one replication. Eight replications were allocated to each dietary treatment (control (balanced diet) and supplement (pea fibre plus balanced diet)) at each housing number. The enriched cages housed 20 hens and two roosters. Four replicates per treatment were allocated to each dietary treatment in enriched cages. The trial period began at 19 weeks of age and lasted 20 weeks (ending at 38 weeks of age).

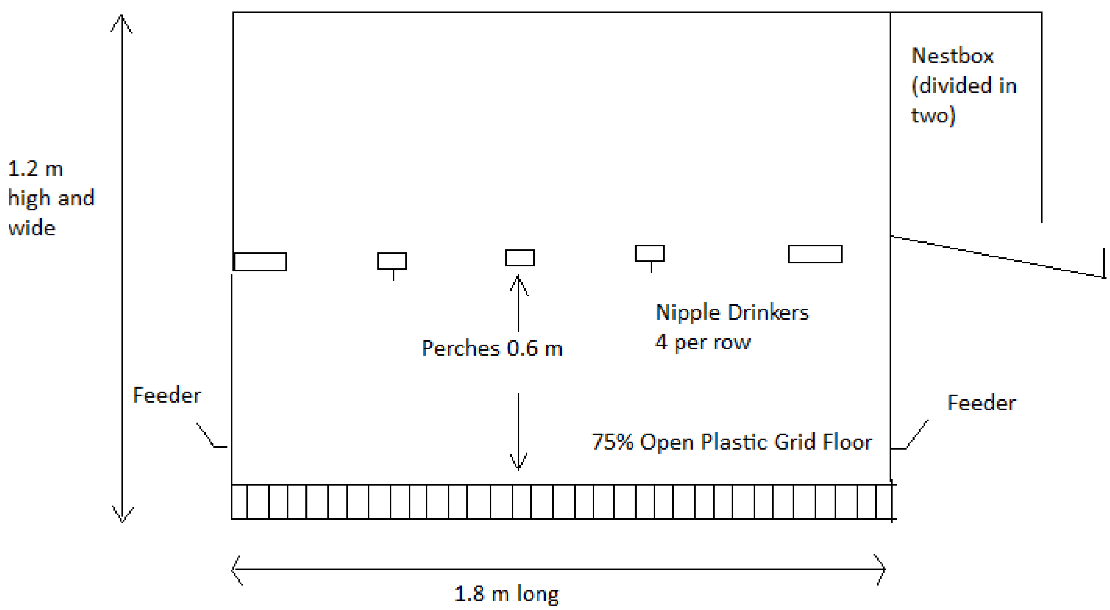


Figure 14. Profile view of enriched cage set up and measurements (Johannson, 2008).

4.3.3 Balanced Diet

A commercial laying hen diet that met or exceeded nutrient requirements (NRC Poultry, 1994) was supplied at levels sufficient for *ad libitum* consumption (see Table 1 in Chapter 3.0). Balanced feed was fed in troughs of all replications, in addition to the insert of the battery cages and rear feeder of the enriched cages in control treatments.

Figure 15. Top: enriched cage used in experimental design; bottom left: single back feeder located beneath the nestbox; bottom right: front view of enriched cage with single front feeder.

4.3.4 Supplement

Pea fibre (animal grade; obtained from Parheim Foods, Saskatoon, SK) was chosen as a supplement because it is low in protein and high in fibre, indicating its low nutrient value (Table 9). Pea fibre was also found to readily absorb water, be small in particle size, and edible. By adding water at a 1:1 weight to volume ratio (Beyer *et al.*, 2002), a high moisture material (56% moisture) was readily obtained, thus making it a material which should be preferred by hens based on the general research results of the previous chapter. The pea fibre was pre-weighed as needed to a desired weight to permit *ad libitum* consumption (30 g per bird per day of dry material) and stored dry at room temperature in re-sealable plastic bags.

Table 9. Nutritional composition of animal grade pea fibre (dry) obtained from Parrheim Foods (Saskatoon, SK)

	Composition (%) dry material
Dry matter	89.80
NDF[2]	62.83
ADF[1]	55.98
Crude Protein	8.48
Total Starch	6.37

[1]Acid detergent fibre.
[2]Neutral detergent fibre.

4.3.5 Feeding Regimen and Feed Intake Measurement

Birds were fed a balanced diet once a day at approximately 8:00am and the supplement (pea fibre) at approximately 11:00 am. Water was added to the pre-weighed pea fibre at a 1:1 weight by volume ratio. The supplement was replaced on a daily basis with fresh material. Balanced feed intake was measured once every two weeks and the average daily consumption per bird was calculated. Supplement intake was measured daily.

In the conventional battery cages, the supplement was provided in a removable insert (made of eavestroughing material) that was centered in the feed trough of the two cage replicate, thus making up 50% of the feeding space. Balanced feed was provided in the remaining trough on either side of the insert. The insert was removed, weighed, the stale supplement removed and the fresh supplement added and spread with a spatula before being replaced. The feeders for the control treatment were identical, but the balanced feed was provided in the insert instead of supplemental pea fibre. All inserts were removed at the 11:00am feeding regardless of treatment, thus treating all birds similarly.

The enriched cages had separate feed troughs for each feed type; the balanced feed was provided in the front feeder of the cage, and the supplement in the rear feeder of the cage at the same level. The control treatment had balanced feed placed in the rear feeder. All feed troughs in the rear of the cage were removed at the 11:00am feeding time, and feed (supplement and balanced) was added after replacement of the feeder to the cage.

Dry matter content of the balanced feed and the pea fibre was determined using AOAC (method 930.15, 1990) method. Samples were taken of the balanced diet and wet pea fibre once, while samples of pea fibre left in the insert for 24 hr were taken on a weekly basis. Consumption on a dry matter basis was calculated for both balanced feed and supplement by subtracting the DM of leftovers remaining after 24hrs from the DM content of the initial amount fed.

4.3.6 Data Collection

4.3.6.1 Body Weight, Egg Production and Digestive Tract Measurements

Body weight was measured at the onset (19 weeks of age) and end of the trial period (38 weeks of age). Egg production was monitored throughout the trial period. At the end of the trial period, 1 bird from each replication of 6 birds per cage in conventional cages was euthanized via cervical dislocation, and the digestive tract removed to determine the effect of pea fibre on digestive tract size. Weights of the crop, proventriculus, gizzard, and small intestine segments were recorded (nearest 0.01g). The small intestine length was also measured (nearest 1 mm with a standard ruler).

4.3.6.2 Feather Scoring

Feather scoring was used to assess damage as a result of feather pecking and was collected at the onset of the trial as well as at 10 week intervals. Two individuals scored feather condition at each session to reduce individual observer bias. The same two individuals scored each session. The scoring system was based on the method of Tausen, *et al.* (2005); a number from 0 to 4 was given based on completely nude (0) to full feather coverage (4) for the following six body areas: head (and/or neck) region, back, tail, vent, wings and breast.

4.3.6.3 Behaviour Data Collection

Behaviour was monitored at 5 week intervals beginning on day 13 of the trial period for a total of 4 observation periods. Feather pecking increases slightly in frequency in the afternoon (Kjaer, 2000; Preston, 1987), so observations took place between 1:00 and 4:00 pm. The order in which the cages were observed was rotated for each observational period (i.e., going in forward or reverse cage numerical order). Three replications of the conventional cages per treatment, and two of the enriched cages per treatment were observed in each session (with a day between each session). Three conventional cage replications were observed at one time, but only one enriched cage was observed.

Each 20 min observation period consisted of a 5 min habituation period, followed by 10 min of instantaneous scan sampling (Altmann, 1974) of all behaviours (Table 10), and ended with a 5 min continuous all-occurrence observation period of feather pecking before moving on to the next set of cages. A feather pecking bout was counted when one hen pecked at the feathers of another individual once or repeatedly until a pause of at least 3 sec occurred or a new bird was chosen as the object of pecking before being counted as a second bout.

4.3.6.4 Statistical Design

Statistical analysis was performed using SAS, version 9.2 (Statistical Analysis Systems Institute, Cary, NC) Proc Mixed procedure. The data were treated as a completely randomized design, and enriched and conventional cage experiments were analysed separately. Egg production, body weight, feed intake,

Table 10. Ethogram of behaviours observed in conventional and enriched cages

Behaviour	***Description***
Feeding	Head extended through the cage bars and pecking at the feed in the through, or gaze directed downward. Divided into areas depending on cage type (conventional: trough and insert; enriched: front and back feeder).
Drinking	Located at the drinker with head at the same level as the nipple with pecks at the nipple.
Resting	A crouched position where the legs and keel are located on the cage floor while the bird is not occupied in any activity. Head may be extended or withdrawn into the scapular feathers. May also place weight on one side with legs slightly extended away from the body.
Standing	Only the feet in contact with the cage floor, not involved in any activity.
Moving	Bird displacing itself from a previous location
Preening	The care of feathers as brought about by drawing each feather though the bill, cleaning and smoothing (Andrew, 1956). It is performed in both standing and sitting positions (Blokhuis, 1984).
Dustbathing	Similar to feather ruffling, as the feathers are raised and the body shaken, but is performed with the keel on the substrate (a sitting position). It is often followed by raking the bill in the litter, and/or pecking at the ground and/or scratching the ground with the feet.
Ground Scratch	Scratching of the cage bottom with the feet in either a standing or crouched position. Often observed in dustbathing bouts.
Wing Flapping	The rapid beating of fully extended wings multiple times in succession.
Feather Ruffling	The vibration of the entire axial body by shaking, with feathers raised and ending with a shake of the head and of the spread tail feathers.

***Continued,* Table 10.** Ethogram of behaviours observed in conventional and enriched cages

Behaviour	*Description*
Stretching	There are two types of stretching: 1. Where the wing and analogous leg of one side, are fully extended to the side but slightly to the back. 2. Where both wings are lifted skyward but not fully extended- left in the folded position.
Scratching	Extension of one leg towards the head, with the later slightly bent towards the approaching leg (occurs in the head and neck area of the bird). The foot is moved rapidly up and down while the digits are in contact with the bird's body.
Object Peck	Any peck directed to an inanimate object; often occurs repeatedly and may occur for an extended period of time.
Feather Peck	Any peck directed to any feathered area on another bird; may be gentle or aggressive.
Head Shake	Similar to feather ruffling and often accompanies it; the head alone is shaken.
Bill Swipe	The rubbing of a beak/bill against an object such as a perch; the bill is moved towards and away from the body while the side is rubbed against the object at a fast pace.
Perching	Birds located on a perch in the enriched cages, in a resting position (a crouched position), not engaged in activity other than head movements while looking around
Mating	Engaged in copulation, crouched, with the male on the female's back. Only observed in enriched cages.
Nestbox	Located in the nestbox; may be engaged in any activity within the box of the enriched cage (ie. Laying, resting, object pecking).
Comfort	Grouping of comfort behaviours for statistical analysis; includes dustbathing, ground scratch, wing flap, feather ruffle, scratch, head shake, and bill swipe.
Total Feeder	Combination of observations/time spent at all feeders and/or feeding areas.

feed efficiency, and digestive tract components were treated as a 2x2 factorial treatment design in conventional cages. Interactions for all variables were not significant. Daily supplement intake over time was analysed using regression. Feather score, feather pecking frequency, and behaviour were analysed as repeated measures for both housing systems. Behavioural data were converted to percent of time occupied before being log+1 transformed before repeated measure analysis. Data from the 2 observations sessions at each collection day were combined since no effect of day was observed. Means were separated using Tukey's and pdmix800 (Saxton, 1998). Significant differences were indicated by $P \leq 0.05$.

4.4 Results

4.4.1 Body Weight, Egg Production, and Feed Intake

Body weight, feed intake, egg production and feed efficiency data of birds housed in conventional cages are found in Table 11. Similar data of birds housed in enriched cages is found in Table 12.

Body weight did not differ between control and supplement treatments in either conventional or enriched cages at 19 weeks of age. Body weight similarly was not affected by the number of birds housed per cage in conventional cages. At 38 weeks of age, supplement fed birds had lower body weights in both conventional and enriched cages, and birds housed at 6 per cage had lower body weights than birds housed 3 per cage.

Hen-day egg production did not differ between dietary treatments in conventional and enriched cages, or as a result of the number of birds housed in conventional cages. Treatment or number of birds per cage did not affect the occurrence of double yolk, soft shell, cracked, broken or abnormal eggs in conventional cages. The levels of cracked eggs and abnormal eggs were greater for hens in enriched cages and fed supplemental pea fibre. The occurrence of double, soft, and broken eggs were not affected by dietary treatment. Mortality was greater in control than supplement fed birds and birds housed 6 rather than 3 per cage in conventional cages.

In conventional cages control birds ate approximately 100g dry matter per bird per day of balanced hen ration. Supplement fed birds ate less balanced ration (approximately 94g per bird per day dry matter) in addition to the 12.8 g (dry matter basis) of pea fibre supplement. Birds given the supplement consumed more total dry matter than control birds. Birds housed at 3 birds per cage ate less pea fibre than birds housed 6 per cage but there was no effect of housing density on balanced diet consumption.

Birds housed in enriched cages ate significantly more balanced feed when they were in the control treatment. Approximately 12.5 g (DM basis) of pea fibre was consumed per bird per day in cages with access to the supplement. Total DM consumption was higher in birds with access to pea fibre.

Control birds in conventional cages were more feed efficient in terms of the amount of total feed per dozen eggs than birds supplemented with pea fibre. Housing density did not affect feed efficiency, nor did dietary treatment for hens housed in enriched cages. In both housing systems, pea fibre fed birds were more feed efficient in terms of balanced feed per dozen eggs than hens only offered laying hen ration.

Supplement consumption was plotted to help decipher any patterns over time (Figure 16). At the beginning of the trial, birds housed at 3 and 6 birds per conventional cage consumed similar amounts of supplement. At the end of the 20 week period birds housed at 6 per cage consumed a higher amount of pea fibre than 3 birds per cage. Birds housed in enriched cages ate higher amounts of pea fibre than the conventional

Table 11. Body weight, egg production, feed efficiency, and feed intake of birds housed in conventional battery cages

	Diet		Birds per cage		
	Control	**Wet Pea Fibre**	**3**	**6**	**SEM**
Body weight 19 wk(kg)	1.41	1.41	1.40	1.41	0.0064
Body weight 38wk (kg)	1.76^{A}	1.71^{B}	1.76^{A}	1.71^{B}	0.0107
Hen day production (%)	94.07	93.76	94.32	93.52	0.3794
Double yolk eggs (%)	0.79	0.94	0.88	0.84	0.0636
Soft shell eggs (%)	0.31	0.37	0.39	0.28	0.0423
Cracked eggs (%)	0.03	0.07	0.05	0.05	0.0154
Broken eggs (%)	0.11	0.15	0.17	0.09	0.0247
[1]Abnormal eggs (%)	0.07	0.09	0.07	0.09	0.0199
Mortality (%)	0.05^{A}	0.01^{B}	0.01^{B}	0.05^{A}	0.0109
Feed Intake (g per hen per day) DM basis					
Balanced diet	100.35^{A}	94.24^{B}	98.37	96.22	0.5996
Pea fibre	--	12.76	11.38^{B}	14.15^{A}	0.1060
Total Intake	100.29^{B}	106.94^{A}	103.98	103.26	0.2117
Total feed/dozen eggs (kg)	1.26^{B}	1.36^{A}	1.32	1.30	0.0089
Balanced feed/dozen eggs (kg)	1.26^{A}	1.20^{B}	1.24	1.21	0.0082

[A,B] Means with different letters in the same row and within a main effect are statistically different ($P \leq 0.05$).

[1]Abnormal = atypical and malformed eggs not in another category.

cages at the beginning of the experiment, but at the end of the 20 week period were consuming less supplement (not compared statistically). Regression analysis of supplement consumption showed a linear increase in birds housed at 3 and 6 per conventional cage. Hens housed in the enriched cages showed a quadratic pattern during the 20 week trial period, with the lowest consumption near day 99.

Table 12. Body weight, egg production, feed efficiency, and feed intake of birds housed in enriched cages

	Diet		
	Control	**Wet Pea Fibre**	**SEM**
Body weight 19 wk(kg)	1.38	1.35	0.0090
Body weight 38wk (kg)	1.80^{A}	1.73^{B}	0.0136
Hen day production (%)	90.22	92.28	0.9836
Double yolk eggs (%)	1.18	1.02	0.0909
Soft shell eggs (%)	0.11	0.13	0.0290
Cracked eggs (%)	0^{B}	0.06^{A}	0.0138
Broken eggs (%)	0.06	0.17	0.0334
[1]Abnormal eggs (%)	0.08^{B}	0.47^{A}	0.0691
Mortality (%)	0.16	0.05	0.0293
Feed Intake (g per hen per day) DM basis			
Balanced diet	106.23^{A}	98.11^{B}	1.2110
Pea fibre	--	12.52	0.2680
Total Intake	106.23^{B}	110.56^{A}	0.3514
Total feed/dozen eggs (kg)	1.53	1.57	0.0152
Balanced feed/dozen eggs (kg)	1.53^{A}	1.38^{B}	0.0175

^{A,B} Means with different letters in the same row are statistically different ($P \leq 0.05$).
[1]Abnormal = atypical and malformed eggs not in another category.

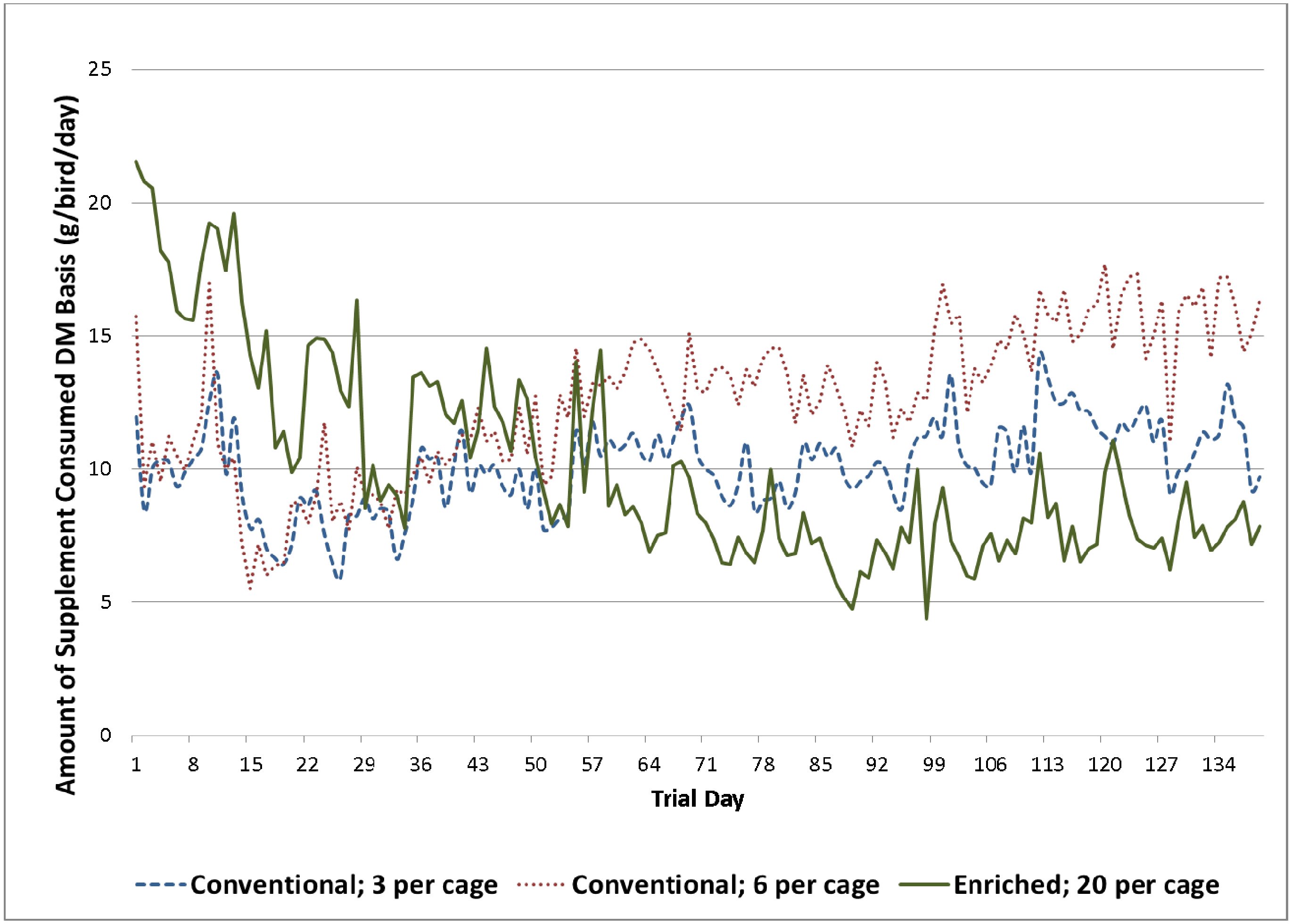


Figure 16. Average daily supplemental pea fibre dry matter intake of birds housed in conventional cages with 3 and 6 hens per cage, and in enriched cages with 20 hens and 2 roosters per cage.

4.4.2 Feather Score

Feather score did not differ between dietary treatments for individual score areas and for total score (Table 13). Hens housed at 6 per cage compared to hens at 3 per cage in conventional cages had a lower feather score for neck, tail, vent, and wing areas. Therefore the overall score was lower for hens housed at six per cage versus three hens per cage. As conventionally housed birds aged, feather condition worsened in the neck, tail, vent, wing, and breast areas, resulting in a lower total score. The treatment by age interaction was significant for vent where perfect scores decreased at a greater rate for control versus supplement fed birds (final mean scores; control = 3.75; supplement = 3.83). The age by housing density interaction was significant for neck (3 = 3.85, 6 = 3.65), tail (3 = 3.02, 6 = 2.76), vent (3 = 3.90, 6 = 3.68), wings (3 = 3.12, 6 = 2.98), and total score (3 = 20.94, 6 = 20.15); scores decreased at a faster rate over time when hens were housed 6 rather than 3 birds per cage.

Dietary treatment did not affect the feather score of birds housed in enriched cages (Table 14). As the hens aged, decreased scores (poorer feather condition) occurred in the neck, back, tail, vent, wings, and breast areas, and therefore the total score decreased. The treatment by age interaction was significant for neck (control = 3.82; supplement = 3.72), back (control = 3.57; supplement = 3.45), tail (control = 3.28; supplement = 3.20), and total score (control = 20.82; supplement = 20.55) areas where supplement fed birds' perfect scores decreased at a faster rate over time compared to control birds.

4.4.3 Feather Pecking Frequency

Feather pecking frequency (observed in 5 min of continuous sampling) did not differ between dietary treatments in conventional cages (Table 15), but frequency was higher in birds housed at 6 per cage than those at 3 per cage. Frequency also decreased with hen age. Feather pecking frequency in the enriched cages (Table 15) was lower in birds given pea fibre supplement and was unrelated to age. The treatment by age interaction was significant; feather pecking frequency was highest in control birds (20 weeks of age = 0.065; 25 = 0.015; 30 = 0.020; 35 = 0.023) and lowest in supplement birds (20 weeks of age = 0.003; 25 = 0.005; 30 = 0.027; 35 = 0.018).

4.4.4 Behaviour

Behaviour is reported as the percent of time that hens expressed each of the various behaviours. In conventional cages (Tables 16), birds given supplement spent less time at the insert area and more time at the trough area than control birds. No differences were found between dietary treatments for other behaviours.

Housing 6 birds per cage resulted in increased time at the insert area, total time at the feeder, drinking, standing, and feather pecking, and decreased scratching in comparison to hens housed at 3 birds per cage.

The age of the hens affected the time spent with the balanced feed, preening, resting, standing and dust-bathing in no particular fashion. Object pecking increased after the first observation period, only to decrease again at 35 weeks of age. Feather pecking decreased after 20 weeks of age. Dust-bathing increased over time, as did ground scratching.

Table 13. Average feather score and standard errors of the mean (SEM) for birds housed in conventional cages

Score Area	Diet: Control	Diet: Wet Pea Fibre	Birds per cage: 3	Birds per cage: 6	Age (weeks): 19	Age (weeks): 28	Age (weeks): 38	SEM
Neck	3.89	3.91	3.95^{A}	3.87^{B}	4.00^{A}	3.98^{A}	3.71^{B}	0.0081
Back	4.00	4.00	4.00	4.00	4.00	4.00	4.00	0.0013
Tail	3.27	3.29	3.36^{A}	3.25^{B}	4.00^{A}	3.00^{B}	2.84^{C}	0.0139
Vent	3.9	3.93	3.97^{A}	3.88^{B}	4.00^{A}	3.98^{A}	3.75^{B}	0.0073
Wings	3.38	3.37	3.44^{A}	3.34^{B}	4.00^{A}	3.09^{B}	3.02^{C}	0.0123
Breast	3.60	3.65	3.63	3.62	4.00^{A}	3.80^{B}	3.06^{C}	0.0131
Total Score	22.04	22.13	22.35^{A}	21.96^{B}	24.00^{A}	21.85^{B}	20.38^{C}	0.0410

^{A,B}Means with different letters within a main effect are significantly different (P<0.05).

Table 14. Average feather score and standard errors of the mean (SEM) for birds in enriched cages

Score Area	Diet: Control	Diet: Wet Pea Fibre	Age (weeks): 19	Age (weeks): 28	Age (weeks): 38	SEM
Neck	3.93	3.90	4.00	3.96	3.77	0.0437
Back	3.90	3.80	4.00^{A}	4.00^{A}	3.50^{B}	0.0127
Tail	3.51	3.56	4.00^{A}	3.34^{B}	3.24^{C}	0.0169
Vent	3.90	3.95	4.00^{A}	4.00^{A}	3.81^{B}	0.0087
Wings	3.55	3.56	4.00^{A}	3.32^{B}	3.33^{B}	0.0163
Breast	3.64	3.64	4.00^{A}	3.89^{B}	3.01^{C}	0.0182
Total Score	22.42	22.43	24.00^{A}	22.52^{B}	20.68^{C}	0.0685

^{A,B}Means with different letters within a main effect are significantly different (P<0.05).

Table 15. Feather pecking frequency (number of occurrences per bird per minute observed) as observed in 5 minutes of continuous observation in conventional and enriched cages

Frequency (average bouts /min per bird)	Diet: Control	Diet: Wet Pea Fibre	Birds per cage: 3	Birds per cage: 6	Age (weeks): 20	Age (weeks): 25	Age (weeks): 30	Age (weeks): 35	SEM
Conventional	0.04	0.03	0.03^{B}	0.05^{A}	0.06^{A}	0.04^{AB}	0.03^{AB}	0.02^{B}	0.0046
Enriched	0.03^{A}	0.01^{B}	-	-	0.03	0.01	0.02	0.02	0.0042

^{A,B}Means with different letters within a main effect are significantly different (P<0.05).

The dietary treatment by age interaction was significant for resting, feather ruffling and moving and the bird housing density by age interaction was significant for preening, both in no particular pattern. The dietary treatment by bird housing density by age interaction was significant for drinking. Drinking increased as the number of birds per cage increased and it increased more in control birds (data not shown).

In the enriched cages (Table 17), birds fed pea fibre supplement spent less time at the back feeding area, and more time preening compared to hens fed only the balanced diet.

The age of the hen affected the time spent at the front feeder and total time at the feeder as well as the expression of drinking, preening, resting, standing, object pecking, perching, dust-bathing, and the combination of comfort behaviours in the enriched cages with no obvious trend related to age. Feather pecking was highest for hens at 20 wk and lower for all later observation periods. Time spent in the nest box increased with hen age, reaching a plateau at 30 weeks of age.

The dietary treatment by age interactions were significant for time at the back feeder, total time at the feeder, feather pecking, dust-bathing, and total comfort behaviours. Time at the back feeder increased over time in supplement fed birds and while control birds spent more time at any given age at the back feeder compared to supplement fed birds, they showed no trend overtime. Total feeding time decreased overtime in supplement fed birds, while control birds showed no pattern over time and had similar consumption rates to supplement fed birds over time. Total comfort behaviour showed no pattern, and dust-bathing occurrence peaked at 25 and 30 weeks of age in supplement and control fed birds respectively. Feather pecking was highest in control birds at 20 weeks of age and then showed no further differences between treatments or age.

Dietary treatment did not affect crop, proventriculus, and small intestine (duodenum, jejunum and ileum) weights in relation to body weight in hens derived

Table 16. Occurrence of behaviours in birds housed in conventional cages expressed as a percent of time occupied in the behaviour within the observation period

	Diet		Birds per cage		Age (weeks)				
Behaviour	**Control**	**Wet Pea Fibre**	**3**	**6**	**20**	**25**	**30**	**35**	**SEM**
Balanced (trough area)	8.20^{B}	15.53^{A}	12.14	11.56	12.38^{A}	9.81^{B}	13.80^{A}	11.32^{AB}	0.3899
Supplement (insert area)	9.61^{A}	5.75^{B}	7.20^{B}	8.18^{A}	6.67	8.17	6.79	9.16	0.3138
Total Feeder	17.81	21.28	19.33^{B}	19.75^{A}	19.04	17.97	20.59	20.48	0.4726
Drink	6.90	7.34	6.50^{B}	7.74^{A}	7.08^{AB}	6.74^{AB}	8.22^{A}	6.42^{B}	0.3135
Preen	8.03	9.06	9.50	7.60	10.25^{A}	10.39^{A}	5.01^{B}	8.62^{A}	0.3479
Rest	8.18	9.22	8.88	8.51	9.72^{A}	9.03^{A}	4.93^{B}	10.87^{A}	0.3425
Stand	27.03	26.06	24.98^{B}	28.42^{A}	30.82^{A}	22.96^{B}	28.01^{A}	24.77^{AB}	0.5696
Object Peck	26.50	22.49	25.39	23.62	16.25^{C}	28.11^{AB}	29.11^{A}	24.70^{B}	0.5489
Feather Peck	1.69	1.61	1.23^{B}	2.06^{A}	3.31^{A}	1.10^{B}	1.40^{B}	0.76^{B}	0.1438
Scratch	0.39	0.27	0.54^{A}	0.11^{B}	0.24	0.51	0.17	0.39	0.0730
Feather Ruffle	0.19	0.18	0.14	0.23	0.21	0.07	0.32	0.14	0.0472
Move	2.68	2.07	2.94	1.82	2.79	3.00	1.79	1.97	0.1996
Head shake	0.04	0.05	0.08	0.02	0.03	0.00	0.06	0.08	0.0288
Bill swipe	0.07	0.00	0.07	0.00	0.00	0.00	0.00	0.14	0.0245
Dustbathe	0.09	0.14	0.07	0.16	0.00^{B}	0.07^{AB}	0.31^{A}	0.07^{AB}	0.0382
Stretch	0.05	0.04	0.04	0.05	0.00	0.04	0.07	0.07	0.0236
Ground scratch	0.14	0.12	0.21	0.05	0.00^{B}	0.04^{B}	0.07^{B}	0.42^{A}	0.0457
Tail shake	0.02	0.04	0.04	0.02	0.00	0.00	0.00	0.11	0.0200
Wing flap	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.0000
Comfort	1.00	0.83	1.18	0.64	0.49	0.73	1.01	1.41	0.1140

Means with different letters within a main effect are significantly different (P<0.05).

from 6 hens per conventional cage (Table 18). Similarly, duodenum and jejunum length were not affected by feeding pea fibre as a supplement. Gizzard and proventriculus weight and ileum length were heavier and longer respectively in supplemented hens. The length of jejunum was numerically longer for the supplement-fed birds (P=0.0653).

Table 17. Occurrence of behaviours in birds housed in enriched cages expressed in percent of time occupied in behaviour within the observation period

	Diet		Age (weeks)				
Behaviour	**Control**	**Wet Pea Fibre**	**20**	**25**	**30**	**35**	**SEM**
Balanced (front area)	9.58	17.52	16.63^{A}	13.21^{AB}	11.38^{B}	12.93^{B}	0.5548
Supplement (back area)	12.24^{A}	5.68^{B}	8.00	8.82	7.47	11.59	0.4316
Total Feeder	21.82	23.20	24.63^{A}	22.03^{AB}	18.85^{B}	24.52^{AB}	0.6223
Drink	6.87	7.98	9.13^{A}	7.23^{AB}	5.60^{B}	7.75^{AB}	0.3961
Preen	6.44^{B}	8.26^{A}	6.69^{B}	5.68^{B}	13.82^{A}	3.18^{C}	0.4175
Rest	0.25	0.37	0.06^{B}	0.77^{AB}	0.42^{AB}	0.00^{B}	0.0697
Stand	10.44	12.46	11.31^{A}	12.59^{A}	6.58^{B}	15.33^{A}	0.5393
Object Peck	22.28	17.83	20.13^{A}	25.10^{A}	11.58^{B}	23.52^{A}	0.6745
Feather Peck	1.13	0.35	1.63^{A}	0.38^{B}	0.71^{B}	0.25^{B}	0.1187
Scratch	0.28	0.57	0.19	0.32	0.79	0.41	0.0804
Feather Ruffle	0.22	0.19	0.25	0.19	0.00	0.38	0.0557
Perch	17.78	16.30	16.06^{B}	14.65^{B}	28.22^{A}	9.20^{C}	0.6692
Move	7.37	7.04	8.44	6.97	5.71	7.70	0.3572
Head shake	0.00	0.07	0.00	0.00	0.14	0.00	0.0242
Nest box	4.27	4.67	1.38^{C}	2.98^{B}	6.28^{A}	7.22^{A}	0.2596
Mate	0.06	0.24	0.06	0.06	0.14	0.33	0.0493
Bill swipe	0.00	0.10	0.06	0.00	0.14	0.00	0.2874
Dustbathe	0.20	0.10	0.00^{B}	0.20^{AB}	0.39^{A}	0.00^{B}	0.0492
Stretch	0.03	0.03	0.00	0.07	0.07	0.00	0.0233
Ground scratch	0.00	0.03	0.00	0.00	0.00	0.07	0.0165
Tail shake	0.16	0.03	0.00	0.06	0.26	0.07	0.0391
Wing flap	0.16	0.06	0.00	0.13	0.25	0.06	0.0414
Comfort	1.05	1.19	0.50^{B}	0.97^{AB}	2.03^{A}	0.98^{AB}	0.1348

^{A,B}Means with different letters within a main effect are significantly different (P<0.05).

Table 18. The effect of dietary treatment on emptied digestive tract segment **w**eights or lengths relative to body mass derived from hens housed in groups of six birds per cage in conventional cages

Organ	Measurement	Control	Wet Pea Fibre	SEM
Crop	Weight[1]	0.30	0.36	0.0206
Proventriculus	Weight[2]	0.27^{B}	0.31^{A}	0.0092
Gizzard	Weight	0.71^{B}	0.96^{A}	0.0559
Duodenum	Length	1.39	1.46	0.0337
Duodenum	Weight	0.72	0.72	0.0091
Jejunum	Length	2.95	3.28	0.0900
Jejunum	Weight	0.91	0.96	0.0324
Ileum	Length	2.62^{B}	3.11^{A}	0.0961
Ileum	Weight	0.70	0.78	0.0337

[1]Weight (g)/body weight (g)
[2]Length (cm)/body weight (g)
^{A,B}Means with different letters within a main effect are significantly different (P<0.05).

4.5 Discussion

As predicted by previous research, the hens demonstrated a strong and consistent appetite for the supplemental moist pea fibre (56% moisture). The strength of this appetite was demonstrated by the activity and crowding at the front of the conventional cage in anticipation of the fresh supplement, as well as the immediate consumption of the moist pea fibre. In contrast, control birds showed much less anticipation at feeding and rarely approached the front of the cage when the insert was replaced.

Supplement-fed birds housed in the enriched cages appeared to be very anxious when receiving the supplement. Hens would often peck at the author’s hands or the bag carrying the supplement, prior to feeding. The control hens in the enriched cages ignored the feeding of the balanced diet. However, part way through the trial period, they began standing near the back feeding area (analogous to supplement feeding area in pea fibre fed hens) while the feeder was removed, and eating immediately from the filled feeder when replaced. In summary, hen behaviour clearly illustrated their partiality for the fresh supplemental moist pea fibre, indicating that flavour, texture, or palatability did not deter consumption.

Supplement intake varied daily but overall consumption was similar for both cage systems. Hens voluntarily consumed approximately 12-13g of DM (or approximately 37-38g as-is) per bird per day, indicating the hens' partiality to the novel feed material. This composed 11-12% of the total dry matter intake in both cage types. The percent of total intake is one third of that observed in Steenfeldt *et al.* (2007) but is similar to the voluntary intake of silage found by Johannson (2008) (approximately 13g per bird per day, dry matter basis). Even though the supplemental material and strains of bird are different in this work than in the Johannson (2008) research, the hens ate the same amount, suggesting that pea fibre is just as preferred as barley silage. The supplement consumption differs from Steenfeldt *et al.* (2007), where ISA Browns consumed nearly 60g as-is of maize and barley-pea silage, and over 100g as-is of carrots. This high consumption could be due to a variety of experimental factors such a bird strain. Brown layers are typically heavier than white strains (like that used in this thesis), resulting in higher feed consumption.

The strain of bird could also impact behaviour, particularly feather pecking. In the current research only the Lohmann LSL strain was used. This bird strain was chosen because of its common use in western Canada, but it is possible that the response of hens to the provision of a supplement would differ in other strains. For example, when comparing the Schwean (1995) and Johannson (2008) studies in the same enriched cages to the current experiment, the frequency of feather pecking in the current research was low. This could be attributable to chicken strain since the current study used Lohmann LSL, whereas Johannson (2008) and Schwean (1995), both using a Shaver strain, found higher occurrences of feather pecking as indicated by poorer feather condition and a higher feather pecking frequency by Schwean (1995).

The high supplement consumption seen for birds in Steenfeldt *et al.* (2007) could also be due to housing type. Housing hens in large groups could potentially result in more frequent or larger stress responses due to the number of social encounters in a large and complex pecking order (Mashaly *et al.*, 1984). The resulting stress could be mediated by larger intakes of supplemental feed, also seen as increased feed consumption as a result of increased corticosterone (Siegel and van Kampen, 1984; Nasir *et al.*, 1999). Two hen housing systems were used to investigate the impact of feeding wet pea fibre on the behaviour of laying hens in the current research. Feather pecking is not as prevalent in conventional cages as in the enriched, large group cages (Schwean, 1995). Thus, it was of interest to study the impact on feeding the wet pea fibre in this type of housing system in addition to the commonly used conventional battery cages. Furthermore, the use of large group housing and aviary-style housing is increasing in some parts of the world, so the comparison of the two housing units has relevance for hen management from a broader housing perspective.

The difference in consumption between the three experiments noted above could also be explained by a difference in nutritional quality of the supplement; the nutrient content of maize and pea silage and carrots is higher than the pea fibre used in either the present work or the barley silage used by Johannson (2008). Therefore, more of the total nutrient requirement of the hen could be supplied by the silage and carrots, hence the greater consumption of supplements in the Steenfeldt *et al.* (2007) research compared to Johannson (2008) and the present experiment.

Regression analysis showed that with time, voluntary supplement intake decreased in enriched cages in a quadratic fashion, while it increased linearly in conventional cages. Although both groups of birds housed in conventional cages had similar supplement intake at the onset of the trial, it is interesting to note that by the end birds housed at six per cage were eating a greater amount than birds housed at three per cage. If we associate supplement intake with stress levels (Siegel and van Kampen, 1984; Nasir *et al.*, 1999) as a result of social interactions, it could be assumed that stress would be high at the onset of housing. This type of stress should decrease with time as social ranking is established even though high ranking birds still tend to peck others (Cunningham and van Tienhoven, 1983; Davami *et al*, 1987). Thus, if pea fibre consumption is an aid to alleviate stress, then we should see increased consumption at the onset of housing (and the trial period) and a decrease in intake with time. This matches the overall trend of moist pea fibre consumption in the enriched cages. In contrast, the 3 and 6 hens per conventional cage showed an increase in consumption with time and that birds housed 6 per cage ate more pea fibre than those housed 3 per cage. The difference between these treatments may be related to stress resulting from competition with others (Hughes and Wood-Gush, 1977) or from competition at the feeder (explained in more detail below). Why birds in conventional cages tended to increase consumption over time compared to the hens in the enriched cages is uncertain but the hens in both housing systems readily consumed the material.

In terms of important production and health indicators, birds fed pea fibre were lighter at 38 weeks of age compared to birds only fed the balanced diet in both conventional and enriched cages. Steenfeldt *et al.* (2007) also observed silage supplemented birds to be lighter in body weight compared to control hens. Body weights were near Lohmann guidelines (Lohmann, LSL 07/10/E) at 38 weeks of age, indicating that the hens were still in good health when consuming the supplement. Lower body weight may be a positive outcome, as supplying a low nutritive material may aid in body weight control. The lower body weight in supplement fed hens could be attributed better feed regulation by a developed gizzard (discussion to follow) as birds with underdeveloped, or small, gizzards tend to overeat (reviewed by Svihus, 2011). Body weight control may reduce the opportunity for obesity or any associated negative impacts on hen health from high body weights.

Conventionally housed birds were also lighter when housed at 6 hens per cage instead of 3. The body weight, again, is near Lohmann guidelines and is no concern for hen health. Increased competition at the feeder in conjunction with a greater number of birds in a pecking order means some birds are unable to ingest as much balanced feed (Davami *et al.*, 1987) and opt for pea fibre consumption in the feeding area where there is less competition, which is demonstrated by the increased supplement intake recorded in hens housed at 6 per cage instead of 3.

Egg production was not affected by feeding pea fibre in birds housed in conventional cages. This was not the case in enriched cages as providing pea fibre for consumption resulted in a higher occurrence of cracked eggs and eggs with other atypical characteristics. It is unlikely that the supplement is responsible for the change and it is more likely a chance result since the incidence of these conditions is very low.

Feed consumption was reported as balanced laying hen ration, pea fibre supplement consumption and total consumption (balanced intake plus supplement intake) all on a dry matter basis. In both housing systems, control birds ate more balanced feed than supplement fed birds (Steenfeldt *et al.*, 2007; Johannson, 2008), indicating that the hens forgo their balanced feed to consume pea fibre. This was also observed in the previous chapter where coarse particle supplements (B, BS, GS, WO) resulted in lower balanced feed consumption than finer particle supplements (GO). A review by Svhihus (2011) suggests that a decrease in feed intake is expected when the diet contains fibre in order for the gizzard to better grind the material. While this may be logical, what makes the intake measurements in the current research different is that the hens are voluntarily forgoing the balanced ration. The reduced balanced feed intake in addition to no differences in egg production with the provision of pea fibre results in greater feed efficiency in terms of the amount of balanced feed per dozen eggs. The lower balanced feed intake may also relate to the reduced body weight of supplement fed birds even though their total dry matter consumption is higher in both housing systems. Since pea fibre is low in nutritional value, it adds less to production parameters such as body weight and egg production than the balanced diet because the majority of the difference in dry matter intake would be fibre. The fibre component would not be digested but rather passed through the digestive tract while providing little in terms of nutrients to the hen, but it may aid indirectly to nutrient digestion by elongating the intestine.

Supporting the effect of fibre, measurements taken of the digestive tract show increased proventriculus and gizzard weight in birds fed moist pea fibre. As the pea fibre is a grainy and fibrous material, it is logical that birds given the supplement would use their proventriculus and gizzard muscles more to break down the material chemically and mechanically, respectively. This in turn increases the development of these muscles, thereby increasing gizzard weight. In addition, the ileum in supplement fed birds shows elongation. This also makes sense since the pea fibre is a fibrous material. While the majority of pea fibre is indigestible, fibrous material can bind with digestible diet components, and in doing so, make it temporarily unavailable to the gut wall for nutrient absorption (reviewed by Hartini *et al.*, 2003), thereby decreasing digestive efficiency. Elongation of the ileum (the last place for nutrient uptake before the colon) would then increase the time and area available for digesta digestion and absorption, thereby compensating for the nutrients temporarily bound by the fibre.

Feather score, a common indirect measurement of feather pecking, was not affected by the presence of supplemental moist pea fibre. There was an affect of age, which is no surprise since both housing systems cause damage and wear on the feathers. This explains the reduction in neck feathering as feathers are damaged by cage bars during feeding or observing the environment outside their cage. It also could explain the reduction in feather condition of the wings, tail and breast; the tail is consistently rubbed against the cage or other hens when turning, the wings also when stretching, and the breast when lying down. In addition, in the enriched cages the back and neck are affected with time as a result of mating since during copulation, the rooster mounts the hen's back and holds onto the neck feathers with his beak. However, cage wear does not explain the reduction in condition and coverage of vent feathers since they do not make contact with other objects as often, and hence must be affected by feather pecking. The tail, while also the result of the housing, is also affected by feather pecking as the author observed feather pecking occurring at this area on a regular basis.

There were some areas where feathering was more affected than others when the number of birds per cage differed. The neck, tail, vent and wings scores were lower (feather condition and coverage worse) when birds were housed at six per cage in the conventional cages. Wathes *et al.* (1985), Davami *et al.* (1987), and Bilcik and Keeling (1999) (even though a different strain housed in litter pens was observed) also found feather coverage to decrease when birds were housed at higher densities. In the current experiment, a reduction in feather condition is likely due to the more limited space for birds housed 6 birds per cage. Hens housed at three per cage were able to rest without having to touch or having minimal contact with another individual. While in cages with six hens per cage, an individual hen was in physical contact with another hen at any given point in time. Hens housed at six per cage had less effective space to move when compared to hens caged with three individuals. They were continuously having their feathers ruffled by their and other hens' movements through the cage, which resulted in abrasion against birds and the cage (Wathes *et al.*, 1985). This explains wearing of the tail, wings, and possibly neck feathers. The neck feathers in birds with six per cage could have also resulted from the increased competition for feeder space as birds would be putting their heads through the bars with more vigour or more frequently than when in cages of three hens, in order to establish a position at the feeder. Hens housed 6 per cage also feather pecked more frequently than hens housed 3 per cage (Hughes and Wood-Gush, 1977). The neck region is an easily reached area for feather pecking and a reduction in vent coverage is the result of pecking and less likely from cage abrasion. The worsened vent condition in birds housed 6 per cage instead of 3 corresponds to the increased feather pecking observed in these cages.

Feather score was not impacted by supplement use, including areas typically affected by feather pecking. This corresponds to the lack of supplement effect on feather pecking behaviour in the conventional cages, but not to the decrease in supplement fed birds in enriched cages.

More feather pecking occurred when six birds were housed per cage in conventional cages. Hughes and Wood-Gush (1977) also found more birds (6 vs. 3) in cages had a higher frequency of aggressive and non-aggressive pecking. This could be the result of the crowding (also suggested by Hughes and Wood-Gush, 1977; Wathes *et al.*, 1985). In the more crowded conditions, feathers are always present to satisfy any motivation a hen may have to peck, or may even promote the incidence of feather pecking since there are tousled feathers which stick out from the body as a result of such close contact. The crowding may also increase aggression or stress, which could be manifested as feather pecking (Davami *et al*, 1987).

Over time, a decrease in feather pecking occurred in the conventional cages. This is as expected if social dynamics stabilize and social conflicts decrease. However, this isn't the case in the enriched cages; there is no significant trend occurring over time.

Modifying the occurrence of feather pecking could also have been observed through adjustments in bird behaviour. If an impact towards feather pecking was possible by supplementing hens with moist pea fibre, not only should a decrease in feather pecking behaviour occur, but also a decrease in object pecking and an increase in total time spent at the feeder. An increase in time spent at the feeder could mean reduced redirected pecking motivation at objects and conspecifics (the redirected foraging theory of feather pecking behaviour). In both cage systems, no difference was found in any of the previously mentioned behaviours, however, the numerical decrease in object pecking and feather pecking, and the increase in total feeding time in supplement fed birds may have helped improve the feather score of the vent by the end of the trial period in conventional cages.

Control birds in conventional cages appear to spend approximately equal time between the two areas of the feed trough (with a slight preference for the insert area). However control hens housed in enriched cages clearly preferred eating from the back feeder. Supplement fed birds ate less balanced feed than controls and this was reflected in the time the same birds spent at the balanced feed (front trough or insert) compared to the higher total time at the feeder spent by control birds (combined both feeding areas).

Preening was observed to increase in birds housed in enriched cages and fed pea fibre, but a similar dietary treatment effect was not found in the conventional cages. This may have resulted from more time to engage in preening, or perhaps there was more space in the enriched cages to engage in longer uninterrupted bouts of preening.

The number of birds allocated per cage in conventional cages affected pea fibre intake. The increased time spent with the insert for birds housed 6 hens per cage may indicate an increased stressful situation associated with the increased housing density or increased feather pecking previously discussed (Hughes and Wood-Gush, 1977; Mashaly *et al.,* 1984; Bilcik *et al.*, 1998). If stress is mediated by increased pea fibre consumption (Siegel and van Kampen, 1984; Nasir *et al.*, 1999), then this may explain the increased supplement consumption of hens housed at six birds per cage. The increased intake associated with a greater bird number per cage could result if competition for space containing conventional feed is high (since there is a high drive to eat in order to compensate for the nutritional costs associated with egg production). Birds that are eager to gain access to the balanced feed but cannot access the feeding area because it is occupied by other hens, may opt to consume the supplemental moist pea fibre as long as it is more easily accessible – a material that will fill the crop and provide a positive short term feed intake control until the hen can reach the required balanced ration (Hodgkiss, 1981). Over time, dominance is established amongst hens in the cage, and dominant hens may command the balanced feed area, limiting its use by hens lower in the social hierarchy (also suggested by Appleby *et al.,* 2004). Subordinate hens must then rely more on consuming the supplement to reach physical satiety before they can obtain access to the balanced feed to gain nutritional satiety. This would then account for the increase in supplement intake over time observed in the enriched cages. Supplement consumption still increases linearly with time in birds housed at three per cage, possibly because there still is a dominance hierarchy amongst three birds, so even though there is still adequate space for all three birds to feed from one feed material at a time, a dominant hen can still prevent the other hen(s) from feeding in that area (Cunningham and van Tienhoven, 1983). Birds housed at six per cage also show an increase in drinking behaviour, possibly as a way of eliminating body heat associated with the increased housing density or as a way of meeting a temporary satiety (Hodgkiss, 1981) if low ranking hens cannot reach the feeder due to dominant hens (Cunningham and van Tienhoven, 1983). An alternate explanation could be that increased water consumption is associated with increased supplement consumption.

An increase in drinking behaviour was observed in pea fibre supplemented hens. This is worth noting as a hypothesis for this increased water consumption involves reaching greater satiety from combining fibre consumption and water (Hocking *et al.*, 2004). The water absorbent nature of pea fibre could absorb more water than that was provided at feeding. The further expansion of the particles from the additional water consumed by the hens may help to increase satiety by a greater distension of the crop and/or gizzard. This physical satiety may also relate to the observed reduction in balanced feed intake.

4.6 Conclusions

It was found that moist pea fibre, which possessed all the characteristics determined from previous experiments to be highly preferred, was readily and voluntarily consumed by Lohmann LSL hens. The hypothesized benefit of providing supplemental pea fibre on feather pecking behaviour and feather score was not apparent and may have been related to the relatively low levels of feather pecking in the flock. Results demonstrated little impact of feeding moistened pea fibre on behaviour and feather score even though hens consumed considerable quantities of the supplement. Although not significant, there was a trend towards a redirection of feather and object pecking behaviour towards spending more time at the feeder. Vent score was improved in supplement fed birds by the end of the trial period. Therefore, it may be too soon to reject the potential of wet pea fibre (or another supplement with similar characteristics) to reduce feather pecking. The effect of a supplemental material may have a greater impact in a different environmental and using a different strain of bird.

Balanced diet supplementation with wet pea fibre decreased the amount of balanced feed consumed but increased the total dry matter intake. Hens showed no difference in egg production, so were more feed efficient in terms of balanced feed per dozen eggs. The increased feed efficiency may relate to physiological changes in the digestive tract from pea fibre consumption. Heavier proventriculi and gizzards and longer jejunum and ileum segments of the small intestine were found in hens that consumed pea fibre. Increased power in the gizzard to breakdown feed which may better regulate feed intake may be related to the improved feed efficiency even though a longer intestinal tract to increase the opportunity for nutrient uptake seems contradictory.

Housing density can affect feed intake as a result of competition at the feeder or stress as a result of increased feather pecking (reflected in decreased feather scores), seen in birds housed 6 per conventional cage as compared to 3 hens.

5.0 Overall Conclusions and Discussion

The current study showed that hens voluntarily eat relatively large amounts of low nutrient material even when a nutritionally balanced diet is available. The reason(s) for this choice is not certain but could relate to taste and palatability as unensiled materials with high moisture content were observed to be preferred. It may also be an attempt to increase fibre intake. This does indicate that animals do not always make dietary choices with nutritional benefits, or at least benefits that are obvious to scientists studying the choice.

Preference testing, although in a less traditional set-up, was shown to be adequate in detecting the relative preference of hens for supplemental materials. This was determined by balanced hen ration and supplement consumption, as well as behaviour in terms of the amount of time spent at the feeding area. The original hypothesis to this thesis was that silage consumption was related to fermentation products produced during ensilation. The hens' readily disproved the hypothesis by consuming more of the unensiled barley greens than the barley silage in the first experiment. It then became apparent that physical characteristics were more influential in the immediate consumption of a feed. Hens showed increased consumption with moist, non-ensiled feed materials and there appeared to be a slight preference for a smaller particle size, although this aspect of the research requires validation using a more definitive range of particle sizes. It is also hypothesized that if hens are allowed an object to redirect their pecking activity, feather pecking will decrease. Results indicated that the hens spent more time with edible than non-edible materials similar in appearance. The choice of edible materials indicates that they provide a better reward than non-edible materials that only encourage foraging behaviour. Thus, a low nutritive, moist and unensiled edible material, such as wet pea fibre was provided to hens as a means to reduce negative feather pecking behaviour.

In laying hen flocks where feather pecking and cannibalism are a common occurrence and a problem for hen welfare, feeding a supplement readily consumed by the hens could potentially reduce feather pecking by increasing time hens spend at the feeder. However, the evidence that wet pea fibre reduces feather pecking behaviour is weak with only minor trends in the current report in terms of behaviour (drinking, total feeding time, object pecking, object pecking and preening) and feather coverage/condition. This may correspond to the lack of a significant increase in the total time hens fed the supplement spent at the feeder. If the success of providing a supplement is indicated by the time redirected from feather pecking to feeding (foraging), the findings suggest a negligible effect of the supplement. It is possible that the lack of difference in total time spent eating was related to the ease with which the hens ate the pea fibre; the smaller particle size made for easy consumption and it is likely that their crops quickly filled. However, it is relevant that hens highly preferred the wet pea fibre and voluntarily consumed it.

Providing an edible supplemental material increased the weight (proventriculus and gizzard) and length (jejunum, ileum) of the digestive tract. This may be related to the reduced amount of balanced feed required to produce a dozen eggs. Future research into this topic should include investigation of the effect of fibrous materials on feed efficiency including how much and what type of fibre is required to maximize feed efficiency. Using the preference model used in this study may permit hens to establish their own fibre requirements.

An attractive feed material can alter the time spent occupied between feeding and feather pecking related activity and in conjunction with fibre intake, could potentially be associated with reduced feather pecking behaviour. Future studies should continue to investigate the potential of feed supplementation as enrichment material for laying hens.

Providing a supplementary material to laying hens proves to be a positive idea, but in a production setting, economic benefits are a priority and producers will equate the costs associated with supplement purchase and distribution with the benefits obtained. Based on the research in this thesis, it would appear that feeding a moist supplemental material such as pea fibre to hens is not the most viable material because it has little impact. However, this should not eliminate the potential of nutritional enrichment for production systems. While not common, nutritional enrichment should be a more reasonable alternative over installing an object that must be managed and/or replaced. While it may appear that hens show the greatest interest in their balanced feed at all times, a supplemental material is still readily consumed. Future research should be conducted in feed enrichment to modify bird behaviour and potentially reduce production costs as a result of hen efficiency.

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