Behavioural and Nutritional Management of Non-Beak Treated Hens Housed in Furnished Cages

by

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ABSTRACT

BEHAVIOURAL AND NUTRITIONAL MANAGEMENT OF NON-BEAK TREATED HENS HOUSED IN FURNISHED CAGES

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This thesis is an investigation of how to manage flocks of non-beak treated laying hens without increasing damage and mortality due to injurious pecking. Three studies, outlined in four chapters, assessed the effects of breed, beak treatment, extra enrichments, and dietary alterations on mortality, behaviour, feather condition, beak shape, and production parameters. In the first study, it was demonstrated that breed affected mortality, bird-to-bird pecking, feather damage, and enrichment use (i.e. Lohmann Classic Brown > Hyline Brown). Hens with intact beaks had more feather damage, caused more damage to the extra enrichments, and had longer upper mandibles. In the second study, dietary alterations affected both behaviour and production parameters. Hens fed animal protein laid more eggs with brown spots, laid lighter eggs, had higher excreta dry matter content, and caused more damage to the extra enrichments than hens fed a purely plant based diet. Hens fed a high fibre diet required a larger volume of feed, laid fewer dirty and/or bloody eggs, had higher excreta dry matter content, and performed fewer spot pecks near the end of the study than hens fed a diet with a standard level of fibre. In the third study, breed affected upper mandible length and beak tip angle (i.e. Columbian Rocks had longer and sharper beaks). The use of extra enrichments was carried out over all three experiments and overall the results were encouraging. Hens with extra enrichments performed less bird-to-bird and spot pecking, had less feather
damage, tended to have shorter beaks (cuttlebones only), and tended to have blunter beak tips (with increased cuttlebone use), but had fewer ‘good’ quality eggs.
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Chapter 1 General Introduction

Animal welfare is becoming more and more important to the general consumer. Issues like confinement housing have been at the forefront of relatively recent ballot initiatives, legislation, or industry-set standards. For example, Proposition 2 in California restricted the use of confined housing for veal calves, sows, and laying hens (Ballotpedia, 2008), Council Directive 1999/74/EC in the European Union effectively banned barren conventional cages for laying hens (European Union Council Directive, 1999) and most recently, the Egg Farmers of Canada released new standards in 2016 giving producers less than 20 years to eliminate conventional caging for laying hens (CBC News, 2016a). Another major welfare concern for laying hens is beak alteration, which is especially important with the move away from conventional cages. Some European countries have already restricted or prohibited any type of beak alteration. In particular, the English government recently reviewed a derogation that permitted beak alteration, with a view for the practice to be banned in 2016. Though the ban was cancelled (Defra, 2015a), the future goal is still to move away from the need for beak treatment as a means to reduce injurious pecking and the associated damage. With the recent European and upcoming Canadian change in housing systems and the need for more humane alternatives to beak alteration, there is an ever-growing need to investigate best practices for housing and management for flocks destined for alternative systems. The welfare of many future flocks is dependant on discovering a solution for injurious pecking, so that flocks of hens with intact beaks can be housed successfully.


1.1 THESIS OBJECTIVES

The main objectives of this thesis are to identify some behavioural and nutritional management techniques that can be used in commercial systems to reduce the occurrence of injurious pecking without the need for beak alterations. For the purpose of this thesis, any mechanical procedure performed on the beak will be referred to as ‘beak alteration’, with beak trimming referring to the hot-blade technique and beak treatment referring to the infra-red treatment, as described in the following chapter.

The focus of this thesis was on brown laying hens housed in furnished cages, with group sizes of 21 or 80 hens/cage, however the final study was carried out using two Shaver breeds, housed in individual cages. Mortality, behaviour, feather condition, and beak morphology was used to assess the relative success of any given treatment. Three key factors were investigated: breed, dietary alterations, and the presence of extra enrichment, which included abrasive surfaces to shorten or blunt the beak tip.

The first study was carried out on a large commercial farm and thus, changes to the diet were not possible. Therefore, the focus of the first study was to assess differences in mortality, behaviour, and feather damage in two commercially available breeds, that were beak treated or not, and that were housed with extra enrichment or not. The main hypotheses were that the ‘alternative’ breed (Lohmann Classic), the beak treated hens, and the hens housed with extra enrichment would have less mortality, perform less injurious pecking, and have better feather cover. In addition, one of the extra enrichments in particular was expected to shorten the beaks, so beak morphology was also measured. The experiment was designed as a factorial to assess the individual, as well as additive effects, of each of the main factors.

2
In the second study, the extra enrichments were carried over (though size and placement were slightly modified) and used in a factorial combination with two different dietary alterations (the inclusion of an animal-based protein source and/or extra fibre). The second study was carried out on the SRUC research farm, and so it was easier to implement dietary treatments as the hens were hand fed and the feed troughs had dividers between the cages. Similar welfare outcomes were measured as in the first study (i.e. mortality, behaviour, feather cover). In addition, production parameters were also measured to ensure that the alternatives diets did not negatively impact on important economic production traits (e.g. egg production). The main hypotheses were that the animal-based protein diet, the extra fibre diet, and the extra enrichments would improve welfare outcomes, such as reduced mortality, less injurious pecking and associated feather damage.

The third study was performed at the University of Guelph’s research farm. Though beak measurements were recorded for the first study, they were not recorded in the second (where contact with the beak blunting board would have been greater as it was placed in the feed trough rather than hung vertically). Therefore, the goal of the final study was to quantify changes in beak morphology and peck force over time and between hens housed with or without two different types of beak blunting material (one of which was the same as the first two studies) in two different locations within the cage. The hypotheses were that the beak blunting devices would reduce beak length, blunt the beak tips, and reduce peck force.
Chapter 2 Injurious pecking, beak alterations, and housing management in furnished cages: A review

2.1 BACKGROUND

The egg production industries in both North America and the United Kingdom (UK) represent a significant contribution to the respective agricultural economies of these countries (Promar International, 2009; Fröhlich et al., 2012). The number of animals used in these industries is staggering, with Canada housing over 27 million laying hens in 2011 (Statistics Canada, 2012) and the UK housing approximately 39 million in 2015 (EEPA, 2016). Therefore, there are massive implications for both animal welfare and economic outcomes relating to regulation and legislation regarding animal husbandry.

Commercial strains of laying hens are often prone to high levels of injurious pecking (IP), which encompasses feather pecking and cannibalism. Though some aspects of conventional cages may increase the risk of IP, managing outbreaks in small groups can be easier for producers. Therefore, the move away from conventional cages throughout parts of world may limit the ability to cope with outbreaks of IP for flocks housed by the thousands. To limit the impact of IP on feather and tissue damage, laying hens are routinely subjected to beak alterations during rearing around the globe, including in Canada and the UK. The main goal is to reduce the performance and damage caused by IP (as described further in following sections). However, a handful of European countries, including Sweden, Finland, Norway, Iceland, and Switzerland have banned all types of beak alterations (Jendral and Robinson, 2004; FAWC, 2007). Under EU Council Directive 1999/74/EC, all mutilations, including beak alterations, are prohibited for laying hens (European Union Council Directive, 1999). However, an amendment was included that allows
individual Member States to permit beak treating as long as the chicks (intended for laying purposes) are less than 10 days old (European Union Council Directive, 1999; FAWC, 2007). Though there are no indications that Canadian egg producers will experience pressure to ban this practice in the near future, the UK recently attempted to review their derogation to allow beak treating, with a view to ban all types of beak alteration in 2016. Recently, this proposal to ban was cancelled so beak alteration is still permitted within the UK (Defra, 2015a) using infra-red technology only. However, reducing the need to regularly alter beaks is still considered important. Regardless of beak status, IP is still an important problem for commercial laying hens and thus worthy of investigation.

The following sections of this literature review will discuss different types of housing for laying hens both in Canada as well as the UK, with particular focus on furnished cages, causes and potential solution for IP, and finally beak alterations and their effects on welfare outcomes.

2.2 HOUSING

There are two main methods of housing laying hens: cages or non-cage systems. Whether a system is defined as a cage (or not) is not dependant on its size as such, but rather management style – where cages are systems that are managed without human entry (AHAW, 2005; Fröhlich et al., 2012). Conventional battery cages are small (usually <10 hens/cage) and barren, providing only feed and water. The term alternative housing generally refers to any housing system that is not a conventional battery cage. Furnished (or enriched) cages are larger and offer furnishings to support natural behaviour (e.g. nest areas, perches, scratch mats). Non-cage systems can be single-tier or multi-tier indoor systems with or without access to a range (“free-range”). Most non-cage systems provide access to a littered area, though some have wire or plastic slatted flooring.
throughout (all non-cage farms in the UK must provide litter, however this is not necessarily the case in North America). Single-tiered systems (also called floor, barn, or free-run housing) provide feed, water, nest boxes and perches on a raised platform and a littered area on the floor. Multi-tiered systems (also called aviaries) provide resources on multiple levels in a wire frame structure with a littered area on the floor (Figure 2.1). Free-range and organic systems are similar to the previously mentioned systems, but they must provide the hens with access to an outdoor area. However, vague language in the organic standards leaves the door open to interpretation regarding certain requirements. For example, outdoor access must be provided except in “extreme weather conditions” for some UK standards (Soil Association Organic Standard, 2007), or when “weather permits” in Canada (Government of Canada, 2015). Each of these systems has benefits for poultry welfare (discussed in following sections). In addition, management style within each system can also impact the level of animal welfare. However, it is their drawbacks that will limit their adaptability and suitability to the North American and British economies (for example, harsh winters may limit the use of free range housing in certain geographical areas).
Although conventional battery cages are the most widely used form of hen housing across the world (CIWF, 2016), they are not the only commercial option. In the last few years, the use of enriched cages has increased, especially in Europe, where the use of conventional cages was no longer permitted after 2012. Though the European Directive (1999/74/EC) uses the term ‘enriched cage’, the term ‘furnished cage’ will be used in this thesis henceforth as it is a less emotive and more objectively descriptive term. As of 2011 in Canada, the majority of hens were housed in conventional cages, with only a small fraction of the market taken by alternative housing systems (Table 2.1). The egg market in the UK differs greatly, as conventional cages are
no longer in use. Approximately half of the eggs are produced in furnished cages and the other half are produced in free-range operations (EEPA, 2016).

Table 2.1 Proportion of hens housed in certain environments

<table>
<thead>
<tr>
<th>Region</th>
<th>Conventional Cages</th>
<th>Furnished Cages</th>
<th>Barn Systems</th>
<th>Free-Range</th>
<th>Other</th>
</tr>
</thead>
<tbody>
<tr>
<td>Canada*</td>
<td>90+%</td>
<td>&lt;10% (all non-conventional cages combined)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>UK**</td>
<td>0%</td>
<td>42%</td>
<td>5%</td>
<td>50%</td>
<td>3% (organic)</td>
</tr>
</tbody>
</table>

*Statistics from BCSPCA (2012)
**Statistics from EEPA (2016)

2.2.1 History of Hen Housing

Up until the 1800s, chickens were kept on small mixed family farms, and ate what they could find (Elson, 2011; Fröhlich et al., 2012). The first known poultry club started in the 1800s and focussed on breeding for more robust poultry with improved performance (Elson, 2011). By the 1930s, poultry farming evolved into something more intensive with more confined housing conditions for egg-laying hens (Elson, 2011; Fröhlich et al., 2012). As a means of increasing hygiene, egg quality and production per unit of land space, while decreasing cost of production, hens were moved from outdoor free range systems to indoor caged environments, where negative production traits associated with extensive living (i.e. parasitic load) could be controlled and reduced (Elson, 2011). The use of conventional battery cages was first introduced commercially in California, USA and also in Switzerland in the 1930s (Arndt, 1932; Fröhlich et al., 2012). This was deemed a success for the industry as mortality and morbidity decreased significantly and, as the hygiene of the hens and eggs were improved by limiting direct contact with excreta, saleable
egg output increased (Appleby et al., 2002; Pohle and Cheng, 2009; Fröhlich et al., 2012). By the 1950s and 1960s, in both North America and the UK, more than 90% of eggs were laid in conventional cages (Elson, 2011). Concern for the welfare of the hens housed in these small, barren cages began to develop in the 1960s, especially after Ruth Harrison’s “Animal Machines” was publicized (Harrison, 1964).

Now, in North America, still more than 90% of all eggs come from hens housed in conventional cages (Street, 2012). However, as consumers become more attentive to certain farming practices, the pressure for change is being heard. Many large retailers are demanding a larger portion of the egg supply to come from hens housed in alternative systems. For example, in Canada, McDonald’s and larger grocery retailers have set goals of switching to alternative suppliers by 2025 (CBC News, 2015, 2016b). Even the egg industry itself is anticipating large changes in housing standards and have set goals for themselves as well. Egg Farmers of Canada released a statement indicating that conventional cages will no longer be allowed by 2036 (Egg Farmers of Canada, 2016). Though the phase-out of conventional cages in Canada will bring about unprecedented change on a large scale for Canadian producers, it is not new for British egg producers. Large retailers in the UK, like McDonalds and Marks & Spencer are already sourcing 100% of their eggs from free-range farms (McDonald’s, 2013; Marks and Spencer, 2016). And, as mentioned previously, the European Council Directive (1999/74/EC) effectively banned the use of conventional cages for laying hens, which took effect in 2012 (European Union Council Directive, 1999).

To date, federal legislation within Canada does not require producers to conform to particular housing specifications. The Animal Welfare Act states that animals must not be put into situations that cause distress, though routine husbandry procedures are exempted (Farm Animal...
Council Network, 2013). However, the Canadian Codes of Practice for laying hens, which are generally considered as the national standards for Canadian egg production, are currently being updated and will aim to include the most up-to-date scientific information and recommendations regarding welfare in different environments (NFACC, 2012). More specific requirements are only followed and enforced if producers participate in farm assurance schemes or if it is demanded from the product retailer. For example, the British Columbia Society for the Prevention of Cruelty to Animals (BCSPCA) has a certification label that requires a cage-free environment with outdoor access for hens (BCSPCA, 2015).

**2.2.2 Advantages and Disadvantages of Conventional Cages**

The term ‘conventional cage’ refers to barren (i.e. feeder, drinker, wire floor) cages that generally house 5 to 10 hens, and in Canada, the current recommended minimum space allowance is 432 cm$^2$/bird for white strains and 483 cm$^2$/bird for brown strains (Canadian Agri-Food Research Council, 2003), though the codes are currently under review. In the EU, before they were banned in 2012, the minimum space allowance for conventional cages was 550 cm$^2$/bird (European Union Council Directive, 1999). Their design is quite simplistic, in that they are made completely of wire, with angled flooring to allow for egg rollout and ease of egg collection. One of the main benefits of the cage over non-cage systems is that it increases hygiene and reduces the incidence of disease as it allows for the separation of the excreta from both the hens and eggs (Duncan, 2001). In addition, cages are more economically viable as they require less land space (vertical space is maximized), and reduce egg damage compared to non-cage housing (Van Den Brand et al., 2004). Small group size is another benefit of conventional cages, making management and daily inspections easier, plus they may limit the spread of behavioural problems such as feather pecking or cannibalism (Duncan, 2001).
Despite their obvious benefits to hen and egg hygiene, conventional cages are widely criticized for their negative effect on hen behaviour and welfare (Appleby, 1998; Duncan, 2001). Due to their small size, these cages impair hen mobility and do not offer the space or the resources to carry out natural behaviours, such as nesting – a highly motivated behaviour – as indicated by the fact that hens will work hard to gain access to nesting substrates (Cooper and Appleby, 1996). Hens housed in conventional cages often show behavioural and physiological symptoms of frustration prior to oviposition, including restless pacing and increased duration of egg retention, leading to extra-cuticular deposits of calcium on the egg shell (Yue and Duncan, 2003). In addition, limited space allowance and lack of perches reduces the opportunity for exercise, general activity, perching, and roosting. This often results in poor bone density and strength, both in turn leading to an increased risk of osteoporosis and subsequent bone breakages at depopulation (Fleming et al., 1994, 2006). Given the chance, poultry will spend a large portion (up to 75%) of the time foraging; scratching and pecking at the ground, looking for food (Savory et al., 1978). Though this daily proportion is probably an overestimate for modern strains of laying hens, as Schütz and Jensen (2001) and Lindqvist et al. (2002) showed that modern White Leghorns performed less foraging behaviour and less contra-freeloading than Red Jungle Fowl or a less highly selected strain (e.g. Swedish Bantam). Still, this behavioural limitation can lead to increases in feather pecking, cannibalistic pecking, and poor feather condition, as has been observed for hens housed on wire compared to those housed in pens with litter (Hughes and Duncan, 1972; Blokhuis and Arkes, 1984).

2.2.3 Advantages and Disadvantages of Furnished Cages

Modern furnished cages are generally larger than conventional cages and contain furnishings that aim to support and promote some of the behavioural needs that have been deemed
important, whilst maintaining the health and economic benefits of housing hens in cages. For example, furnished cages contain a nesting area, comprised of a plastic flooring (usually plastic mesh or AstroTurf) enclosed with plastic curtains. In addition, perches and an area designated for foraging and scratching are also usually provided.

Though the conventional cage helped improve hen and egg hygiene as well as improve production efficiency (Duncan, 2001), the behavioural and welfare-related problems associated with severe confinement and barrenness led to the development of alternative systems that included benefits from both outdoor and indoor housing. The first design of a furnished caging system was developed in the 1970s, and was deemed the ‘get away cage’ as it included multiple tiers to which subordinate hens could escape to different levels (Elson and Tauson, 2012). The specific design of this cage, with feeders on two levels, resulted in problems maintaining good hygiene, and thus lead to further developments. The Edinburgh modified cage was developed in the UK in the 1980s (Appleby, 1998; Elson and Tauson, 2012), and included perches as well as a nesting area. Further modifications to these types of furnished cages included a scratching or foraging area as well (Appleby, 2003). Commercial use of furnished cages was first seen in Sweden, where more than 40% of the nation’s hens were being housed in such systems by 2002. Though the cages were originally designed to house a smaller number of hens, modern commercial furnished cages can house more than 30 hens, and often house upwards of 80 hens or more.

The EU directive (1999/74/EC) outlines specifics in regards to cage design. It requires that furnished cages must include: increased space allowance per hen, designated nesting areas, perches, and scratch/dustbathing mats. The EU directive stipulates a minimum of 750 cm² floor space per hen, 12 cm/hen at the feeder, 15 cm/hen of perch length as well as available litter or substrate for scratching and foraging. There is no minimum requirement for the amount of nest or
foraging space per hen. The extent to which each cage is designed to incorporate these furnishings varies from company to company, and individual farms may modify the cages post-installation as long as they still meet the legal requirements. For example, some companies produce furnished cages that incorporate a plastic mat in the middle of the cage where feed particles are delivered via a central auger tube at varying intervals (Big Dutchman, 2016), whereas other manufacturers have textured pads near the feed trough, onto which feed is sprinkled at each feeding (Tecno Poultry Equipment S.p.A., 2016).

Although furnished cages may still not be ideal for hen welfare (Lay et al., 2011) or completely satisfy the consumer’s demand for high welfare products, non-cage egg production may not be an economically viable (or sustainable) option in terms of supplying the entire egg market. Furnished cages, then, may well serve as the best compromise between animal welfare and commercial profitability and may become the standard for housing in the EU and Canada (Fröhlich et al., 2012). In general, furnished cages, in comparison to conventional cages, do a better job at meeting the behavioural requirements of laying hens. However, cages of any type still limit the extent of “naturalness” a hen can experience. Nesting and foraging areas that have been designed to maintain hygiene standards and to limit the financial input may not be ideal from the perspective of the hen and therefore a maximal level of animal welfare has not yet been reached by any current furnished cage model.

2.3 ABNORMAL REPETITIVE BEHAVIOUR

Within the literature pertaining to animal behaviour and welfare, the term stereotypic behaviour has been used to encompass a wide range of abnormal, unvarying, repetitive and
functionless behaviours (Mason et al., 2007). These types of behaviours are often observed in sub-
aequate environments or when goal-oriented behaviour is restricted, and are thought to relate to
frustration and stress. For example, highly feed-restricted sows often chew on the bars of their
crate, on chains, or vacuum chew (Rushen, 1985). These behaviours decrease dramatically when
sows are fed ad-libitum (Bergeron et al., 2000). Horses housed long-term in isolation perform
abnormal locomotory behaviours such as weaving or head-bobbing, which are reduced when
horses are housed with mirrors to simulate companionship (Mcafee et al., 2002). Captive giraffes
tongue-roll and repeatedly lick objects, which is thought to reflect a lack of adequate foraging
material (Baxter and Plowman, 2001). Route tracing and sham chewing decreases when
budgerigars are housed in larger cages (Polverino et al., 2015). Additionally, feather pecking in
laying hens, which is the focus of this section, is also thought to be abnormal and repetitive in
nature, and may or may not be stereotypic (Kjaer and Sørensen, 1997; Dixon, 2008). However,
though abnormal repetitive behaviours are most often observed in stress-inducing environments,
there may be some benefit gained by the individuals performing these behaviours (i.e. these
behaviour have a function). The performance of these behaviours may allow the individual to
better cope with their environment (van Hierden et al., 2002a; Mason et al., 2007). For example,
two divergent lines of laying hens, with either a high or low propensity for feather pecking, have
been shown to adopt different coping strategies (i.e. proactive or reactive) (van Hierden et al.,
2002b). Low feather pecking chicks had higher baseline and post-treatment (5 minutes of manual
restraint) corticosterone levels than high feather pecking chicks, suggesting that the high feather
pecking strain responds to their environment differently than the low feather pecking line and are
potentially less reactive to environmental stress (van Hierden et al., 2002b). Mason et al. (2007)
has suggested another definition that may more broadly incorporate most types of stereotypic
behaviour: “repetitive behaviour induced by frustration, repeated attempts to cope and/or C.N.S. (central nervous system of the brain) dysfunction”. Therefore, Mason et al. (2007) also suggested the use of “abnormal repetitive behaviour” in cases where exact causation may be hard to ascertain, and when the definition of a stereotypic behaviour may not apply. This definition incorporates all behaviours, regardless of cause, that may indicate an animal is experiencing poor welfare and thus feather pecking could be classed as this type of behaviour.

2.3.1 Injurious Pecking

For laying hens, the most common abnormal behaviours are feather pecking and cannibalism, behaviours which are both considered a part of IP (Lambton et al., 2013). Feather pecking can be classified as ‘gentle’ (GFP) or ‘severe’ (SFP). The distinction between GFP and SFP has been scientifically documented (Savory, 1995; Bilčík and Keeling, 1999) and may represent two motivational systems (Dixon et al., 2008). GFPs are directed just at the tips of feathers, generally do not cause feather removal, are usually ignored by the recipient hen, and can be repetitive in nature (Savory, 1995; McAdie and Keeling, 2002; FeatherWel, 2013). On the other hand, SFPs include grasping and pulling of feathers and usually do lead to feather removal, and some sort of reaction from the recipient hen as they are most likely painful (Bilčík and Keeling, 1999; Gentle, 2011). SFP can be observed within bouts of GFP, or on their own, but are generally not performed as repetitively as GFPs. Cannibalistic pecks can either be pecks directed at the cloaca or vent region (‘vent pecking’) or at any other body part (other than the head) that causes tissue damage (Savory, 1995). Though aggression and aggressive pecks can in fact cause wounds, it is generally not grouped within the IP umbrella as it is thought to stem from an entirely different etiological basis (Savory, 1995; Rodenburg et al., 2008). Aggression or aggressive pecking, as categorised within the study of poultry behaviour, represents a behaviour that is a part of
establishing a hierarchy, dominance, or used for aid in resource guarding, and is not synonymous for SFP. Though GFP, SFP, cannibalism, and vent pecking have been grouped together within an overarching category, this does not mean to say that they have the exact same etiological basis either. Though there may be a correlation between GFPs and SFPs, the performance of GFP during rearing does not predict future performance of SFP (Newberry et al., 2007). There is also evidence to suggest that feather pecking and vent pecking are not necessarily correlated (Hughes and Duncan, 1972; Hocking et al., 2004); that is to say that a highly denuded flock may not have issues with vent pecking, yet another quite well feathered flock could. Vent pecks may stem from a different causal mechanism than cannibalistic pecks directed at other body parts, as they may be linked to hormonal changes at the onset of lay (Savory, 1995). In addition, exposure of the mucosal membrane during oviposition can stimulate pecking from conspecifics towards this region (Savory, 1995). Cannibalism to other body parts may develop as a result of SFP, as feather removal that causes bleeding can stimulate subsequent cannibalistic pecking (Savory, 1995). In contrast, there is evidence feather pecking and cannibalism are not linked (Keeling and Jensen, 1995). Hughes & Duncan (1972) did not find any differences in feather pecking behaviour or feather condition prior to an outbreak of cannibalism between groups of hens that were either later found to have high levels of cannibalism-related mortalities or those that were not.

Not only are these behaviours undesirable from an animal welfare point of view, but also from an economical stance as they can often lead to an increase in flock morbidity and mortality as skin lesions, wounds, and infections can affect hen health. Welfare is also impaired as high feather pecking flocks have been shown to display more behaviours associated with general distress. For example, Bright (2008) found that high feather pecking flocks performed more vocalisations, including squawks, than low feather pecking flocks. In addition, significant feather
loss can increase metabolic demands for heat production, leading to increased feed intake, lower egg production, and lower production efficiency (Glatz, 2001; Sedlačková et al., 2004; Su et al., 2006).

IP is generally considered a multifactorial problem and there are a number of risk factors for its development, and they may differ in their importance depending on the type of IP. In general, genetics, rearing environment, lack of foraging or dust bathing substrate, nutrition and feed type, hormonal effects, environmental disturbances, stocking density (and/or group size) and lighting have historically been considered as the main risk factors for IP and will be discussed in the following sections.

2.3.1.1 Access to Foraging or Dustbathing Substrate

There are two mainstream theories of IP’s etiology: 1) misdirected foraging (Blokhuis and Arkes, 1984; Blokhuis, 1986; Huber-Eicher and Weschler, 1997), or 2) misdirected dustbathing behaviour (Vestergaard and Lisborg, 1993). The problem with these two hypotheses is that they both have a common behavioural component: ground pecking. Therefore, it is relatively difficult to determine which of the behaviours, when frustrated or not satisfied, leads to redirected feather pecking. However, it appears that IP is probably most related to misdirected foraging. For example, devices that provided the opportunity for foraging, but not for dustbathing were more effective at reducing feather pecking than those that were designed for dustbathing or as a novel object (Dixon et al., 2010). And, Dixon et al. (2008) was able to demonstrate that SFP is most similar in morphology, based on fixed action patterns that hens show, to pecks performed during foraging. In addition, Huber-Eicher & Weschler (1997) found that the provision of straw (tested as a foraging substrate) reduced the rate of feather pecking, whereas the provision of sand (used as a dustbathing substrate) had no effect on feather pecking behaviour. Similarly, Nørgaard-Nielsen (1997) found
no differences in rates of feather pecking between hens given access to a dustbathing box that was either empty or contained sand. Additionally, Gunnarsson et al. (2000) found that although 50% of the hens they tested worked hard for access to feathers, they did not perform dustbathing while engaged with the feathers. Instead, hens performed behaviours more closely approximating foraging activity; scratching and pecking at the feathers (Gunnarsson et al., 2000). Pullets housed on wire flooring were more likely to feather peck than those housed on litter (Blokhuis and Arkes, 1984; Blokhuis and Van Der Haar, 1989). In addition, those that remained with litter into adulthood pecked less frequently than those that were reared with litter and moved into wire housing or those that were reared on wire and remained there throughout lay (Blokhuis and Arkes, 1984). Similarly, El-Lethey et al. (2000) and Huber-Eicher & Weschler (1997) found that hens without access to straw substrate performed more feather pecking than those housed with straw. In addition, there were other indications that hens without access to straw experienced some form of stress: the duration of tonic immobility was longer, egg production was reduced, and the heterophil:lymphocyte ratio was higher (El-Lethey et al., 2000) all suggesting a stress response. Although the availability of foraging substrates has a positive effect on levels of feather pecking, not all substrates are equal. For example, Huber-Eicher & Wechsler (1998) showed that long-cut straw was more effective at reducing feather pecking than shorter, shredded straw. Treatments that were effective at reducing feather pecking were also those treatments that promoted more foraging activity, thereby highlighting the inverse relationship between feather pecking and foraging (Huber-Eicher and Wechsler, 1998). In addition, hens will choose to work (e.g. forage through wood shavings) to gain access to more than 30% of their food intake even when it is otherwise freely available (Lindqvist et al., 2002). These findings, among others, corroborate a number of
other scientific works that have attributed the development of feather pecking to the lack of suitable foraging substrates.

In the wild, fowl spend the majority of their day feeding: engaging in both appetitive and consummatory phases of feeding. Pecking and scratching in search of food (i.e. foraging) is the main aspect of the appetitive phases, whereas the consummatory phase encompasses only the actual ingestion of food (Timberlake and Sliva, 1995). While observing a flock of feral domestic fowl, Savory et al. (1978) noted that the hens pecked at their environment up to 15,000 times per day. In commercial housing, the feed offered to the hens is comparatively energy dense and easily accessible. In barn (free run), free range and most organic systems, hens have the opportunity to perform foraging behaviour, even though all or almost all of their energy source will be from feed gathered at the feeders. However, in practice, the litter quality on commercial farms may not be ideal (e.g. litter is often wet or capped) and thus may not be adequate to support foraging behaviour. For caged hens, there is a severely limited opportunity to forage, and even then, it is only available to those hens housed in furnished cages. Though foraging and contra-freeloading behaviours are reduced for modern strains compared to less genetically selected strains (Schütz and Jensen, 2001; Lindqvist et al., 2002), these behaviours have not been eliminated by selection for egg production and are important for hens. In addition, cages also lack an appropriate or ideal substrate for dustbathing. The scratching mats in furnished cages are usually quite small (Figure 2.2), especially for large group sizes and generally do a poor job of retaining substrate for pecking, meaning that positive feedback for pecking, foraging, or dustbathing on the mats may be inadequate. Therefore, in an environment with a lack of appropriate substrate for foraging, as well as dustbathing, it is not entirely surprising that IP still occurs frequently.
Figure 2.2 Photos of scratch mats in two different furnished cage systems: (a) Big Dutchman Eurovent EU enriched colony systems (Big Dutchman Inc., USA), and (b) Tecno Comfort Plus enriched cage model (Tecno Poultry Equipment, S.p.A., Italy).

2.3.1.2 Fibre and Feed Form

Modern commercial diets are generally energy dense and freely available, meaning that hens are not required to spend large amounts of time searching or foraging for food. For example, energy-dense or pelleted feed can be consumed in less time than a diet high in fibre or in mash form, thereby allowing more time to be allocated to non-consummatory behaviour (i.e. feather pecking). El-Lethey et al. (2000) showed that hens feather pecked less frequently when fed a mash diet, which took longer to consume, compared to those fed a pelleted diet, suggesting that time spent feeding reduces time available for feather pecking. However, there may be inherent characteristics of a high fibre diet that reduce IP rates, independently of purely altering time budgets (reviewed by Choc and Hartini, 2003; van Krimpen et al., 2005; Rodenburg et al., 2013). When given free access to a control diet or a diet high in fibre (diluted by 8% spelt hulls), hens of a high feather pecking line were observed to eat more of the high fibre diet than hens from a low pecking line (Kalmendal and Bessei, 2012). Hens from high feather pecking lines have also been shown to consume more feathers (which are non-nutritive) than low feather pecking hens,
suggesting that feather pecking may be a method of gaining access to insoluble fibres that may be lacking in commercial diets (McKeegan and Savory, 2001; Harlander-Matauschek et al., 2007). In addition, McKeegan and Savory (1999) showed that pecking damage was positively correlated with fecal feather content. If feather eating is an attempt to increase the fibrous component of the diet, then this may be one of the main functions of SFP. Kriegseis et al. (2012) showed that a diet containing 10% feathers was able to reduce the incidence of SFP and reduced feather damage for certain body parts. The proposed mechanisms behind the beneficial effects of fibre are: (1) a shift in time budgets (i.e. reducing the amount of time hens have available to spend performing unwanted behaviours), or (2) via inherent properties of fibre, but data are equivocal. Morrissey et al. (2014) found a tendency for increased feed pecks and decreased feather pecks for broiler breeders fed a high fibre diet. Similarly, Steenfeldt et al. (2007) showed an increased in feed intake and a reduction in feather pecking bouts for laying hens fed a high fibre diet. In contrast, van Krimpen et al. (2008) reported an increase in eating time for low energy diets, but this did not affect feather pecking rates.

2.3.1.3 Genetic Predisposition

Genetic predisposition plays a large role in the development of IP behaviour. IP is generally associated with high producing commercial strains, though not exclusively performed by modern breeds (Hocking et al., 2004). Inherent differences for the propensity for IP do exist between breeds (Hocking et al., 2004), the performance of IP is a heritable trait (Rodenburg et al., 2003, 2004a), and divergent lines from a single strain have been developed based on levels of IP performance (Kjaer et al., 2001). In fact, Kjaer et al. (2001) were able demonstrate differences in tendency to feather peck between two divergent lines of laying hens: one with a high propensity to feather peck (HFP), and one with a low propensity (LFP). These differences were apparent after only two
generations of selective breeding. Not only do HFP hens feather peck more frequently, but they also have been shown to eat more feathers during choice tests in comparison to their LFP counterparts (Harlander-Matauschek et al., 2007). Gvaryahu et al. (1994) identified a strain of Lohmann laying hens as having the highest mortality rate compared to several other commercial strains when housed in battery cages with 3 hens/cage. More and more research has been devoted to understanding how to incorporate IP behaviour (or lack thereof) into breeding programmes. Traditionally, breeding companies have focussed on health and production parameters for hens housed in individual cages (Craig and Muir, 1996; Jones and Hocking, 1999). This can be useful for commercial hens that are housed in conventional battery cages, where group sizes were small, activity is limited, and feather peckers can be easily identified. However, selection based on very few traits has been criticised as one of the reasons for increased behavioural problems in modern farming (Jones and Hocking, 1999). In addition, the move toward alternative and socially sustainable housing systems means that breeding companies are looking to adapt their breeding programmes to meet the needs of the modern production system. Breeding companies have begun to house hens in family groups and using social and behavioural parameters as part of their breeding goals (Craig and Muir, 1996; Rodenburg et al., 2008).

2.3.1.4 Rearing Environment

It has been suggested that early life experiences can have significant effects on behaviour into adulthood (reviewed in Rodenburg et al. (2008)). GFP has been observed occurring as early as two days of age (Nicol et al., 2013) and can persist into adulthood, suggesting that this behavioural problem can develop into a chronic welfare issue very quickly. Nicol et al. (2001) showed that exposure to wood shavings for as little as 10 days during rearing resulted in less feather pecking than hens that had always been housed on wire flooring. Interestingly though,
adults hens that had never had access to wood shavings foraged at a similar frequency to hens that had already had previous experience with the particular substrate (Nicol et al., 2001). However, results presented by Blokhuis & Arkes (1984) do not support this safeguarding theory. These authors found that pullets housed with litter from 1 day of age but subsequently moved into no-litter environments at 17 weeks of age feather pecked more frequently than those left on litter or even those housed in no-litter environments for the duration of the experiment. In addition, pecking related mortalities were the highest in this group (litter → no-litter treatment). Another study showed that the presence of a mother hen during rearing increases foraging behaviour at 1 and 8 weeks of age (Riber et al., 2007). Though differences in GFP and SFP were not significant, chicks housed with brooding hens had fewer pecking-related mortalities than the non-brooded chicks (Riber et al., 2007) when they were 14-27 weeks of age. In the absence of a mother hen, careful use of light and dark periods with a dark brooder can to some extent mimic the effect of a mother hen on chick behaviour. SFP was greatly reduced for chicks that were reared with a dark brooder from 0-5 weeks of age compared to those reared with just a heat lamp (Jensen et al., 2006). These behavioural differences persisted up to 23 weeks of age. Even just a 2-hour burst of light (~750 lux) on day 19 of incubation resulted in chicks that performed more feather pecking than chicks that had been incubated in complete darkness (Riedstra and Groothuis, 2004).

2.3.1.5 Physiological Factors

At the individual level, physiological factors have also been implicated in the development of IP behaviour. The onset of egg laying is linked with increased hormone circulation and has also been linked to a spike in cannibalistic pecking and mortality (Savory, 1995). Another hypothesis not related to hormone circulation is that the exposure of the cloaca during egg laying can be attractive to young hens and become a pecking target (Savory, 1995). However, hormonal
fluctuations have been shown to increase IP as large exogenous doses of oestrogen and testosterone did increase feather pecking (Wysocki et al., 2010).

In addition, increased locomotor behaviour (or hyperactivity) has been suggested to be linked with increased IP (Kjaer, 2009), with the hypothesis that modern selection for high levels of production has altered this behavioural trait. Support of a hyperactivity model has been reported elsewhere (Kjaer et al., 2015), where high feather pecking hens were observed to be less perseverant in sequences of pecking and were quicker to switch between stimuli in a given task that measured behavioural perseveration.

Though feather pecking is generally considered an undesirable behaviour and an indicator of poor welfare, there is some evidence to suggest that it is used by some hens as a proactive coping mechanism to deal with stressful environments (reviewed in Rodenburg et al. (2004)). Jensen et al. (2005) showed that hens that performed more feather pecking were also more active in an open field test, a restraint test, as well as a novel object test, compared to hens that were classified as feather pecking recipients. However, the specific cause-and-effect between fearfulness and feather pecking is unclear in that particular study. In addition, high feather pecking hens showed lower serotonin and dopamine turnover than their low feather pecking conspecifics (van Hierden et al., 2002b). In humans, serotonergic system is responsible for regulating mood, sleep, and appetite, and has even been implicated in disorders such as schizophrenia and obsessive compulsive disorders (Wysocki et al., 2010). It is hypothesised that serotonin may have similar effects in poultry, in terms of the role it plays in psychological diseases in humans. After identifying a dose for a serotonin agonist that effectively reduced serotonin turnover, van Hierden et al. (2004) showed that dosing hens with this agonist significantly increased the performance of IP, in particular SFP behaviour.
2.4 REDUCING INJURIOUS PECKING

Currently, the two main methods used commercially to prevent and control the development of IP behaviour or the extent of the damage are beak alterations (by hot blade trimming or infra-red treating) and the use of dim lighting. Studies have shown that beak alterations can reduce the damage and mortality caused by and associated with IP (reviewed by Jendral and Robinson (2004)). Significantly more SFP and a higher rate of mortality was observed for hens housed with a high light intensity (30 lux) compared to low light intensity (3 lux) (Kjaer and Vestergaard, 1999). However, these management tools have been criticised for not actually addressing underlying motivation for these behaviours. In addition, though infra-red beak treatment and beak trimming at a young age (e.g. <10 days of age) is considerably more humane than hot blade trimming at an older age (e.g. 5-10 weeks) in terms of acute and chronic pain as well as neuroma formation (Jendral and Robinson, 2004), it is still considered a mutilation and some European countries have already banned or are moving towards a ban on all types of beak treatments (Jendral and Robinson, 2004). Less invasive methods of beak maintenance may include the use of beak blunting devices that would be voluntarily used by hens. Van der Weerd et al. (2006) showed promising results with two types of abrasive materials on the bottom of the feed trough for non-beak altered hens. From 45 to 61 weeks of age, there was on average 1 mm reduction in beak length for those hens with the abrasive material throughout the rearing and lay periods compared to hens without. Further studies will need to address its effects on feather damage and mortality associated with IP. However, interventions that address the underlying issues would be more effective at reducing rates of IP and its associated damage, but are harder to implement in commercial production and do not always work across a large array of situations.
2.4.1 Beak Alterations

Beak alteration is a common husbandry procedure to which the majority of laying hens across the globe are subjected. The goal is to remove the distal portion of the upper and lower mandibles (Cheng, 2006). In some countries it is common to see hens with less than 50% of their beak remaining intact, whereas regulations in the UK only permit up to a third of the beak tip being removed (Defra, 2010). The main reason for beak alterations is to blunt the beak tip as a means of reducing the potential for damage caused by both feather pecking and cannibalism. Currently, beak alteration remains the most effective and reliable method of reducing damage caused by pecking and associated mortality (Blokhuis et al., 2007). Although outbreaks of IP do occur for flocks with altered beaks, the damage is much less significant and spreads at a slower rate, making them easier to manage. This procedure, therefore, has benefits for both the flock as well as the producer, as mortality and morbidity associated with feather pecking and cannibalism is greatly reduced (Jendral and Robinson, 2004). However, beak alteration is considered an assault on animal welfare, since it involves the removal of part of a hen’s major sensory organ and does not actually address the cause of IP. Consequently, some countries have banned beak alteration entirely. For example, producers in Finland, Sweden, Norway, and Switzerland house only hens with intact beaks (Prescott and Bonser, 2004; Defra, 2015b). Austrian producers are encouraged to house hens with intact beaks by having economic incentives on the egg products from these flocks (CIWF, 2009; Defra, 2015b). In the UK, at least one of the organic assurance schemes restricts the use of routine beak alteration, though permission for ‘emergency beak trimming’ is granted in special circumstances should an outbreak of IP occur (Soil Association Organic Standard, 2007).

Up until recently, hot blade trimming was the most commonly-used form of beak alteration in Canada and the UK, and it is still permitted and used in various countries such as France, Spain,
and Greece (Defra, 2015b). The procedure of hot blading requires birds to be handled individually, with their beaks being placed into a machine that uses a heated guillotine style blade to remove and cauterize the beak tip (Dennis and Cheng, 2010a). Up to 50% of the beak can be removed during this procedure and variation between birds may be high due to human error. A newer technology, which is currently used in both North America and the UK, is the infra-red beak treatment (Nova-Tech Engineering LLC, Minnesota, USA). This technology employs the use of a high intensity infra-red beam that penetrates the outer horn of the beak and destroys the underlying tissues (Dennis and Cheng, 2012). Following the procedure, the beak is still intact and can remain so for up to three weeks. After the tissue undergoes complete necrosis, the distal tip of the beak sloughs off. This type of alteration is generally done within the hatchery at one day of age. The process is more automated and efficient, limiting the time that each chick is handled.

In an observational study, Carruthers et al. (2012) found that beaks that been treated with infra-red beam were shorter at 24 and 60 weeks of age than those trimmed with a hot blade. Marchant-Forde et al. (2008) reported similar results from 5 to 9 weeks of age, although results from Dennis et al. (2009) showed the opposite; that beaks were shorter at 30 weeks of age after having been trimmed with a hot blade. However, all of the chicks used in both Carruthers et al. (2012) and Marchant-Forde et al. (2008) had their beaks altered at one day of age (regardless of type of beak alteration), whereas the chicks used in the study by Dennis et al. (2009) were trimmed either at the hatchery (infra-red) or between 7 and 10 days of age (hot blade). Therefore the discrepancies between the experiments may be a result of the age difference at the time of the procedure.

Altering beaks by any method has been criticized as causing both acute and chronic pain for billions of birds around the globe (Cheng, 2006). It appears that the infra-red technology may be
less of a welfare insult than hot blading, and it is preferred by some animal welfare advocacy
groups (FAWC, 2007). After a sudden or swift injury (e.g. hot blade trimming), there is usually a
short period of time that the afflicted area remains painless. For hot blade beak trimming, this
painless phase, where birds show no behavioural differences to intact-beak controls, can last for
up to 24 hours post procedure (Gentle et al., 1991). This phase may be attributed to a few possible
causes, most likely being that the nerves are severely injured and thus are incapable of transmitting
signals, or that the injury was so traumatic as to cause an influx of endogenous opioids acting as
analgesics. Chicks trimmed with a hot blade showed increased heart rates, reduced activity levels
(as indicated by less frequent preening, pecking, and movement, and more frequent sleep bouts),
as well as reduced feed intake when compared to intact controls (Gentle et al., 1982; Glatz, 1987;
Craig and Lee, 1990). Blokhuis & Van Der Haar (1989) showed that birds beak trimmed with a
hot blade (one third of beak tip removed at 45 days of age) performed less ground pecking and
feather pecking during the rearing period than birds with intact beaks. The effect of beak trimming
on feather pecking was much more pronounced for birds that were also housed on litter; such that
beak trimmed birds on litter pecked the least, beak trimmed birds on wire feather pecked as
frequently as intact birds on litter, but less than intact birds on wire (Blokhuis and Van Der Haar,
1989). However, the differences in ground pecking between beak trimmed and non-beak trimmed
birds were not significant during the laying period, whereas feather pecking remained less frequent
for beak trimmed birds (Blokhuis and Van Der Haar, 1989). In addition, a loss of beak function,
including mechanoreception and magnetoreception, has been documented for birds with altered
beaks. For example, Prescott and Bonser (2004) reported that at 32 weeks of age, beak trimmed
hens were significantly less successful at picking up particles of feed than hens with intact beaks,
especially when presented with only one layer of feed. Freire et al. (2011) showed differences in
number of pecks, latency to peck, and magnetoreception between trimmed and intact-beak chicks. For example, beak trimmed chicks pecked less frequently and had longer latencies to first peck than chicks with intact beaks during tests on 2–9 days of age (Freire et al., 2011). After being trained to associate a magnetic field with food, chicks with intact beaks spent more time close to the magnet that contained hidden food than beak trimmed chicks (Freire et al., 2011), indicating that chicks with trimmed beak were not as capable at detecting the magnetic field. These results suggest that beak alterations are associated with acute and chronic pain as well as changes in beak function.

### 2.4.2 Additional Environmental Enrichment

Environmental enrichment has been defined by Newberry (1995) as: “modification of a barren-captive environment to improve the biological functioning of animals”. The caveat is that the enrichments must support the performance of strongly motivated behaviours, or otherwise increase the complexity of the animal’s behavioural repertoire (Newberry, 1995). However, within this thesis the term enrichment may also refer to anything that is not normally present within commercial farming conditions for caged laying hens (i.e. things other than nests, perches, or scratch mats).

Lack of environmental complexity or stimulation has been suggested to lead to the development of abnormal behaviour. This has led to animal keepers and scientists developing an array of devices that can be used as environmental enrichment. Because zoo animals are kept on public display, zoo keepers have historically been the most keen to develop methods of increasing environmental complexity as well as giving the animal more to do. However, the idea of enrichments in farming systems is becoming more popular. For example, Compassion in World Farming has advocated the use of enrichment substrates for growing pigs, specifically those that
encourage and support natural rooting behaviour for pigs (CIWF, 2011). Bell et al. (1998) suggested that animal welfare organizations are interested in increasing environmental complexity and reducing negative affective states within commercial animal husbandry. The agricultural industry may be hesitant to take on the added cost of supplying enrichments, but there are financial benefits that may negate the costs, for example by reducing mortality (Bell et al., 1998).

A number of enrichment devices, or “toys”, have been tested for their effects on bird-to-bird interactions (specifically feather pecking and cannibalism), mortality (mainly due to cannibalism or vent pecking), and productivity. In a series of six individual experiments, Gvaryahu et al. (1994) showed that enrichments, in the form of coloured beads hanging from plastic key rings that were incorporated into 3-hen battery cages were effective at reducing pecks directed towards the head of conspecifics. In addition, the hens housed without these plastic toys performed more pecks in total and had higher mortality rates (Gvaryahu et al., 1994). When beak trimmed and non-beak trimmed hens were used, the presence of a plastic key ring did not appear to have any overall effects on productivity, egg weight, feed consumption, or mortality (Bell et al., 1998). However, enriched hens did perform better than non-enriched hens in early lay, but did not maintain this advantage into the later stages of the laying period (Bell et al., 1998), suggesting some degree of habituation to the enrichments. Interestingly, when analysed separately, the enrichments had a positive effect (although not statistically significant) for the beak trimmed hens, but not for the hens with intact beaks (Bell et al., 1998). Gao et al. (1994) found that a small metal bell was 2.5 times more frequently used than a plastic key ring by 33-week old hens (Agrotoy, AgroTop Ltd., Israel). In addition, habituation to the enrichments was not found. In fact, the opposite was shown by an 18% increase in usage over the duration of the 2-week study (Gao et
al., 1994). Gao et al. (1994) proposed that hens may need more than 2 weeks to respond to environmental complexity, and that hens learn to interact with novel object gradually.

Stocking density may have an impact on the use of enrichments. Increased activity at an enrichment may elicit more attention from surrounding hens. On the other hand, having only a few enrichments to satisfy a large number of hens may increase demand for a limited resource, inducing frustration and potentially more bird-to-bird pecking. In one study, Gao et al. (1994) determined that an Agrotoy (Agrotop LTD, Israel) was used more frequently in battery cages housing 5 hens (420 cm²/bird), compared to those housing only 1 or 2 hens (2100 cm² and 1050 cm²/hen, respectively). However, the use of another enrichment device (metal bell) was not affected by the same stocking densities as previously mentioned (Gao et al., 1994). In a different study with larger group sizes (5, 6, or 7 hens/battery cage), Bell et al. (1998) found that mortality within enriched (plastic key ring, Gallus Ltd., Israel) cages of 5 hens was significantly lower than their non-enriched counterparts, as well as the enriched and non-enriched cages housing 6 or 7 hens.

To capitalize on biological relevance with a minimal amount of economic input, Sherwin (1995) tested the effect of round balls in the feed trough of caged hens. This made feeding more difficult, which utilizes the desire to work for food, even when the same food is readily available elsewhere (i.e. “contra-freeloading”). In caged systems, the ability for hens to work for food, or forage, is severely limited. When balls of different sizes were placed in the feed trough of modified cages, hens pecked readily at the balls and were more likely to have their head outside the cage (Sherwin, 1995). Sherwin (1995) suggested that this was beneficial as hens are less likely to perform feather pecking or cannibalistic pecking, at least to body parts below the neck, while their heads are outside the cage. Although the data suggested that hens were interested in the balls when there was an intermediate number of them (12 or 24 balls, compared to none), there was actually
a reduced tendency for the hens to have their heads over the trough when there were 36 balls (highest density tested). Interest in the balls was maintained over a period of time (Sherwin, 1995), and interest was especially piqued following human manipulation of the balls (i.e. stock workers moving balls to redistribute them along the feed trough).

In an attempt to identify what behavioural motivation an enrichment would have to fulfil before benefiting the rate of feather pecking or mortality within a flock of hens, Dixon et al. (2010) tested three categories of enrichments (i.e. substrates used exclusively for foraging, dustbathing, and novel objects), not just different models or makes of enrichment devices. Foraging type devices included wire bird feeders that were filled with one of three substrates: (1) peanut butter in suet, (2) sunflower seeds in suet, or (3) cabbage leaves. As the substrates were contained within the wire feeders, they were not able to be used for dustbathing. Dustbathing material consisted of two different colours of sand or peat moss. Though these materials could be used for foraging, previous work showed that they are preferred for dustbathing (Sanotra et al., 1995; Olsson and Keeling, 2005). Finally, the novel objects were wooden blocks covered in either tin foil, tissue paper, or felt. Feather pecking was observed at the highest frequency within the treatment without any type of enrichment and lowest in the treatments with the foraging devices (Dixon et al., 2010). Although both the dustbathing and novel object devices reduced feather pecking in comparison to the control, they were not as effective at doing so as the foraging devices (Dixon et al., 2010). Not surprisingly, the foraging and dustbathing devices were pecked at more frequently than the novel object, suggesting that biological relevance can affect the attractiveness of an enrichment (Dixon et al., 2010), which is an important aspect that Newberry (1995) discussed.

Polypropylene rope or string has been tested in a number of research experiments (discussed in this paragraph), mainly due to its potential cost effectiveness. In addition, ropes are
relatively easy to incorporate into a variety of housing environments, and because they can be secured overhead in a hanging fashion, they remain free of excreta and usually do not harbour disease. In addition to the definition listed above as given by Newberry (1995), Jones et al. (2004) suggested that it must be practical, so as to encourage use on-farm. Therefore, there are a few requirements that must be added to the definition of an enrichment, in this case for laying hens: (1) it must be attractive, (2) it must maintain its level of attraction, (3) it must be safe, durable and easy to implement, and (4) it must reduce pecks to conspecifics rather than purely stimulate more pecking to the enrichment (McAdie et al., 2005). One study in particular found that the inclusion of enrichments simply increased the amount of overall pecking performed by the hens, rather than redirecting pecks away from conspecifics (Lindberg and Nicol, 1994). However, enrichments on the whole have been found to produce positive results, including reduced bird-to-bird pecking and associated mortality. In a series of studies, Jones et al. (1998-2006) identified characteristics of rope that may increase its ability to satisfy the requirements of an enrichment device. Jones and Carmichael (1998) found that white and yellow rope was preferred over other colours for mature hens (80-86 weeks of age). The white and yellow ropes were most readily approached and elicited the greatest number of pecks and pecking bouts (Jones and Carmichael, 1998). The blue rope was least preferred in their first experiment, and even upon comparing just the white and blue rope in a second study, the white rope remained the more attractive colour (Jones and Carmichael, 1998). Colton and Fraley (2014) reported reduced feather pecking and improved feather cover for Pekin ducks housed with ball enrichments, however, they notes that the ducks pecked more frequently at blue or green balls than red or white ones. Additionally, plain rope was more attractive and elicited more pecks than ropes incorporating shiny beads along its length (Jones et al., 2000), which may reflect a preference for a rope’s ability to be frayed. Another aspect, namely movement, of
the rope device was tested and it was found that stationary ropes attracted more pecks than those that were periodically jiggled (Jones, 2001). Pecking directed towards the ropes was still observed for up to 17 weeks after initial presentation, even when access to the enrichment was uninterrupted (Jones et al., 2000). Based on the results of their previous studies, Jones et al. (2002) concluded that rope devices had satisfied at least one of the criteria for successful enrichments – to attract pecks and maintain attraction. Another criterion was tested in a subsequent study looking at differences in feather pecking while a rope device was present or not (Jones et al., 2002). Although the data did not reveal statistically significant differences, there were numerically lower pecks directed towards the feathers of a pen-mate when the rope device was present than when a rope device was absent (Jones et al., 2002). However, the previous listed studies were all conducted using hens that were housed in small groups in barren wire cages, and therefore may have behaved differently than hens housed in larger groups, or those in furnished cages. Pecking at rope devices was not shown to be as frequent in a study by Hocking and Jones (2006) when they were used on a commercial broiler breeder farm. However, this could be a result of different motivation behind enrichment use between birds from broiler and laying strains.

2.5 FEATHER CONDITION ASSESSMENTS

As a secondary measure of feather pecking frequency, plumage condition assessment can be used. This technique makes it much easier to capture the outcome of feather pecking, and requires much less time and effort from the observer (Bilčík and Keeling, 1999; Bright et al., 2006). There are already a number of different scoring systems, some requiring close inspection of a number of different body parts (Bilčík and Keeling, 1999; Savory et al., 1999) and some giving one overall score (Hughes and Duncan, 1972) or observing the hen without handling it (Bright et
Remote feather scoring can be most applicable to research on commercial farms, or where large numbers of hens need to be assessed, as it requires less time and causes fewer disruptions and stressors for the hens themselves (Bright et al., 2006; Kjaer et al., 2011). Differences in the number of body parts scored and the scale of the scoring system itself have been apparent throughout the literature, sometimes making it difficult to compare feather condition between experiments (Kjaer et al., 2011). In addition, a high level of inter-observer reliability is much harder to attain when using a more detailed scoring system (i.e. a greater number of points with which each body part is scored, or having more body parts to score) (Bright et al., 2006). Bright et al. (2006) reported high correlations between a scoring system that handled hens and one that remotely assessed them, even though remote scoring tended to result in lower (or better) scores. Conversely, Kjaer et al. (2011) showed that remote feather scoring resulted in higher scores for the wings and tail than when the hens were individually handled. In addition, Kjaer et al. (2011) reported low levels of inter-observer reliability between two teams that were responsible for scoring the hens. They therefore suggested that this needs to be taken into consideration, balanced, and accounted for within the statistical model for experiments where multiple observers are collecting data.

### 2.6 SUMMARY

IP is a multifactorial behavioural problem affecting the egg industry, which has not lessened with the move away from conventional cages to other housing methods. To remain sustainable, the industry must strive to find effective preventative and therapeutic measures that are accepted by the general public. Beak alterations have come a long way in the last few decades,
but other less invasive methods of addressing the underlying causes for IP need to be explored and implemented in commercial systems.
Chapter 3 Can non-beak treated hens be kept in commercial furnished cages? Exploring the effects of strain and extra environmental enrichment on behaviour, feather cover, and mortality

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3.1 ABSTRACT

Commercial laying hens are prone to injurious pecking (IP), a common multifactorial problem. A 2×2×2 factorial design assessed the effects of breed (Lohmann Brown Classic (L) or Hyline Brown (H)), beak treatment (infra-red treated (T) or not (NT)), and environment (extra enrichment (EE) or no extra enrichment (NE)) on mortality, behaviour, feather cover, and beak shape. Hens were allocated to treatments at 16 weeks of age and data were collected every four weeks from age 19 to 71 weeks. Data were analysed in Genstat using mixed models. L hens had higher all and IP-related mortality than H hens (P<0.003), whilst NT hens had higher mortality than T hens but only due to culling of whole cages (P<0.001). Feather cover for L hens deteriorated more quickly with age at most body sites than H hens (age×breed×body site P<0.001). For NT hens, feather cover was worse at most body sites (beak treatment×body site P<0.001), and worsened more quickly with age (age×beak treatment P=0.014) than T hens. L and NE hens performed more bird-to-bird pecking than H and EE hens, respectively (breed P=0.015, enrichment P=0.032). More damage to mats and ropes was caused by L and NT hens than by H and T hens, respectively (age×breed P<0.005, beak treatment P<0.001). Though H hens had fewer mortalities and better feather cover, breed effects may have been influenced by farm management practices.

3.2 INTRODUCTION

IP is currently the most problematic behavioural issue facing the poultry industry as it impacts on bird welfare as well as production economics (Blokhuis and Wiepkema, 1998; Glatz, 2001). IP is categorised as bird-to-bird pecking that results in plumage damage, feather loss, and tissue damage, and includes GFP and SFP as well as cannibalism (Lambton et al., 2013). Aggressive pecking is not generally considered within the IP umbrella as it has a different
etiological basis (Savory, 1995; Rodenburg et al., 2013), even though high levels of aggression can also cause tissue damage and reduce welfare. In general, the prevalence of IP in a flock is thought to reflect unfulfilled behavioural needs, so it is not just a welfare problem for the victims (Rodenburg et al., 2013). Although IP is not a new problem (Hughes and Duncan, 1972; Blokhuis and Arkes, 1984), it is still notoriously difficult to prevent and treat.

IP is a multifactorial problem, with a wide range of risk factors, including genetics and environment (Savory, 1995). Genetic differences in the propensity to perform IP are evident between breeds (Hughes and Duncan, 1972; Hocking et al., 2004) as well as between two divergently selected lines of a single strain (i.e. high and low-feather peckers) (Kjaer et al., 2001). The heritability of IP behaviour ranges from 0.20 to 0.65 (Craig and Muir, 1993; Kjaer et al., 2001), meaning that selection against this behaviour could be introduced into commercial breeding programmes to reduce IP. However, this type of commercial selection may only prove beneficial as a long-term goal as balances between behavioural traits and egg production are perfected. There has been some indication that selection against IP increases egg production, but reduces egg quality (Su et al., 2006). Modern selection methods are beginning to incorporate group performance into breeding programs which could include some way to identify groups of high feather peckers (Rodenburg et al., 2008; Bolhuis et al., 2009). Identifying differences between breeds or strains that are currently available will help producers chose the most suitable breed for the housing system in use.

Providing birds with the opportunity to forage (e.g. by providing straw, whole grain in litter, etc.) reduces the risk of feather pecking (Blokhuis and Arkes, 1984; Huber-Eicher and Weschler, 1997; Aerni et al., 2000). On commercial free range and barn systems, maintaining good litter quality can reduce the risk of GFP (Nicol et al., 2013). However, for caged systems, providing
suitable foraging substrates can be challenging even though it might be of more importance given the relatively barren environment of a cage. Various types of plastic toys can reduce aggressive pecks (Gvaryahu et al., 1994; Bubier, 1996) and providing string enrichments to hens can reduce feather pecking, thereby improving feather cover (McAdie et al., 2005). In particular, white string enrichments have been shown to be attractive to chicks (Jones and Carmichael, 1999) and hens pecked more frequently at white or yellow string than chains and beads or blue or orange string (Jones and Carmichael, 1998; Jones et al., 2000).

Currently, using some method of beak alteration remains the most commonly used and most effective and reliable method of reducing the damage caused by IP (Hughes and Duncan, 1972; Jendral and Robinson, 2004; Guesdon et al., 2006). This is not to say that pecking is not an issue for flocks with altered beaks, but that the damage they are able to cause is lessened. There are two main methods of treating beaks for poultry: hot blade trimming and infra-red treatment. Infra-red beak treatment has fewer adverse effects on chicks than hot blade trimming with regards to neuroma formation and symptoms of chronic pain (Gentle and McKeegan, 2007; Kuenzel, 2007; Dennis and Cheng, 2010a). Despite these benefits, infra-red beak treatment is still deemed a mutilation in the EU and some European countries have altogether banned any type of beak treatment (Van Horne and Achterbosch, 2008). Other governmental agencies, like the Department for Environment, Food, and Rural Affairs (Defra) in England, aimed to ban the practice but then withdrew due to advisors indicating that subsequent damage caused by pecking in intact-beak birds would be too great a risk to welfare (Defra, 2015a). Although infrared beak treatment will continue in England and the rest of the UK for the foreseeable future, the issue may rise again, whereby the practice could stop voluntarily or be prevented through legislation. Furthermore, IP remains a problem even for beak treated flocks.
Though there is a wealth of information pertaining to the etiology, risk factors, and prevention of IP, there is not a great deal of information pertaining to its performance in furnished cage systems. The ban on conventional cages within the European Union in 2012 with Council Directive 1999/74/EC (European Commission, 1999) created a need for more housing and management information specific to this type of housing. Furnished cages were initially designed to house relatively small groups of hens (e.g. 20), however the trend now is for larger group sizes, which may have implications for social behaviour and development of IP. Therefore, understanding how to manage non-beak altered hens in these systems is important, especially if legal or voluntary bans on beak alterations take effect across the EU. Therefore, the goal of this study was to measure the effect of breed and extra environmental enrichment on behaviour and welfare in both beak treated and non-beak treated hens.

3.3 MATERIALS AND METHODS

3.3.1 Ethical Considerations

This study was conducted under a Home Office licence according to the Animals (Scientific Procedures) Act 1986 and was approved by Scotland’s Rural College’s animal ethics committee. Due to the high risk nature of housing non-beak treated hens, an overall cage threshold for intervention was defined as: “two or more birds from one cage die or are culled due to pecking related damage”. Appropriate intervention included hot blade beak trimming or culling the remaining hens within the affected cage.
3.3.2 Animals

A total of 5120 laying hens of either Hyline Brown (2560) or Lohmann Brown Classic (2560) were used. Half of the chicks of each strain were beak treated at day-old at the hatchery using an infrared technique (Nova-Tech Engineering, USA). From 0 to 7 weeks of age, pullets were reared in groups of 1280 in deep-littered floor pens at a commercial farm in the UK, according to breed and beak treatment. From 7 to 16 weeks of age, the pullets were housed in small colony cages designed for pullets. Pullets had *ad libitum* access to commercial rearing diets (Appendix I) and water, and were reared following standard commercial schedules for temperature, lighting, and vaccinations.

3.3.3 General Husbandry

For the duration of this study (16 to 71 weeks of age) all hens were housed in 80-bird furnished cages (4.81×1.26 m or 758 cm²/bird, Tecno Poultry Equipment S.p.A., Italy, see Appendix II) at a commercial farm in the UK. The shed used in this experiment contained seven banks of cages, each ten cages high and 22 cages long (a total of 1540 cages). The shed was split into two levels, with a suspended floor separating the fifth and sixth tiers. Lights were suspended from the ceiling (or the suspended floor) and bobbed between three vertical positions every 20 minutes. The bottom tiers on each level were excluded from the experiment as they were not illuminated as brightly as the others (tier 1 and 6, mean: 1.5 lux vs. remaining tiers mean: 8.8 lux). To mimic dawn and dusk, lights at both ends of the shed were switched on 30 min prior to lights-on and 30 min after lights-off. Sixty-four cages (eight cages long×eight tiers) were used in this experiment and they were all located in the exact centre of the shed. Each cage contained the furnishings required by law (European Council Directive 1999/74/EC), including two nesting areas (each measuring 60×60 cm), perches (15 cm/bird, at two different heights) and two
scratching areas (each measuring 38×24 cm). For both preventive and therapeutic reasons, hens were treated for red mite infestations throughout the lay period (Milben Ex, applied topically). All hens had *ad libitum* access to water and a standard commercial layer’s mash, delivered by automatic feed hoppers eight times per day.

### 3.3.4 Experimental Treatments

The experiment was designed as a 2×2×2 factorial, with breed (Hyline Brown (H, which was the breed used in the rest of the shed) or Lohmann Classic (L)), beak treatment (infra-red treated (T) or not treated (NT)), and extra enrichments (no extra enrichment (NE) or extra enrichment (EE)) as the three main factors. Upon arrival to the laying farm at 16 weeks of age, birds were allocated to the cages in groups of 80 hens, with eight cages (640 birds) per treatment. The treatments were systematically allocated to cages so that the eight treatments and the two levels of each of the three factors balanced spatially, with each column and each tier containing each treatment. Half of the cages had been fitted with extra environmental enrichments, including eight polypropylene ropes (8 mm diameter, 40 cm long), two pecking mats (each measuring 30×10 cm) and two beak blunting boards (each measuring 30×10 cm), evenly distributed throughout the cage. The pecking mats comprised of a combination of compressed wood chips and biodegradable glue on a plastic mesh backing (ROWA, Melle, Germany). The beak blunting boards were made up of an abrasive paste (S N Supplies, Lincoln, UK) previously used in beak blunting trials (Fiks-Van Niekerk and Elson, 2005; Van der Weerd et al., 2006) which was painted onto a Perspex® backing. Four bolts were drilled into each blunting board to act as a shiny attractant for the birds. The pecking mats and blunting boards were fixed vertically to the fronts of the cages. The ends of each rope were lightly heat-sealed to slow destruction and prevent birds from swallowing rope
fibres. The ropes were doubled over and secured to the cage top so that they hung from the cage ceiling to 20 cm below it.

3.3.5 Data Collection

Every four weeks, from 19 to 71 weeks of age, two observers visited the farm over two consecutive days to collect data pertaining to bird behaviour, feather cover and extra enrichment wear and tear, resulting in 14 farm visits in total. On the first day, scan sampling for injurious pecking behaviour, as well as assessment of feather cover and extra enrichment damage were collected. Both observers were present and each observed either the top four or bottom four tiers, alternating at consecutive visits. The second day was entirely devoted to live focal bird sampling for all types of oral behaviour (with only one observer present).

3.3.5.1 Mortality

Birds were examined by stock workers on a daily basis and mortality (deaths or culls) was recorded as necessary. Hens were assessed for cause of death (or reason for cull) and whether it was related to IP. To prevent misclassification, from age 48 weeks, the carcasses were chilled and taken to a veterinarian for post-mortem examination, from which the cause of death was established.

3.3.5.2 Behavioural Measures

One scan sample per cage was performed by two observers on the first day of each farm visit to record the number of hens performing all types of bird-to-bird pecks (Table 3.1). Only the hens in the west half of the cage (i.e. hens between the front and centre line of the cage) were observed due to limited visibility. Within each tier, the cages were always observed in order from south to north. However, the order with which the tiers were observed was balanced using four
Latin Squares on the basis of a maximum of 16 visits (and eight tiers), though the last two visits were dropped as the birds were depleted just after the fourteenth visit (approximately 73 weeks of age). Scan sample data were gathered from 09:00 to 11:00.

For focal behaviour sampling performed by one observer on the second day of each farm visit sampling cages in the same order as described above, a handheld machine (Psion Workabout Pro³, Motorola Solutions, USA) was used to collect data via Pocket Observer software (Observer XT v11, Noldus Information Technology, Netherlands). One hen per cage was observed during each visit between 09:00 and 16:00 for a maximum of 5 minutes (which was shortened to four minutes from the second visit onwards to accommodate the farm’s working hours). All pecking related behaviours were recorded (Table 3.1). To start an observation, one focal hen was systematically chosen from each cage based on its proximity to one of four pre-determined locations. To reduce selection bias, these locations or their equivalent in NE cages (nearest south nest box, second rope, blunting board or pecking mat) were distributed as evenly as possible along the length of the visible side of the cage and starting locations and observation order were balanced as evenly as possible across visits. If the observer could not see the original focal hen for the entire observation, another hen was chosen from the same original starting location and the observation was continued. This change in bird was recorded. The bottom tiers on each level (tiers 2 and 7) were observed from the ground, whereas it was necessary to use a mobile trolley to view all of the other tiers.
Table 3.1 Ethogram for behavioural observations during scan and focal sampling. All incidences of any birds observed performing each of the oral behaviours listed below were recorded.

<table>
<thead>
<tr>
<th>Behaviour</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gentle feather peck&lt;sup&gt;IB&lt;/sup&gt;</td>
<td>Furtive, deliberate, often repeated pecks at another bird’s feather where the recipient usually does not react.</td>
</tr>
<tr>
<td>Severe feather peck&lt;sup&gt;IB&lt;/sup&gt;</td>
<td>Similar to GFP, however, includes grasping and pulling of the feather and may include feather removal. Recipient often reacts, squawks or withdraws.</td>
</tr>
<tr>
<td>Cannibalistic peck&lt;sup&gt;IB&lt;/sup&gt;</td>
<td>Peck directed at (or causing) bloody wounds, tissue damage or removal. Directed towards ANY part of the body EXCEPT for the vent (see vent peck) or toes (see toe peck).</td>
</tr>
<tr>
<td>Vent peck&lt;sup&gt;IB&lt;/sup&gt;</td>
<td>Peck to the vent area.</td>
</tr>
<tr>
<td>Toe peck&lt;sup&gt;B&lt;/sup&gt;</td>
<td>Peck at another bird’s toes.</td>
</tr>
<tr>
<td>Aggressive peck&lt;sup&gt;B&lt;/sup&gt;</td>
<td>Overt, rapid, forceful, usually downward directed peck, usually directed toward recipient’s head or back of neck. Usually not overly repetitive, recipient reacts immediately.</td>
</tr>
<tr>
<td>Rope peck&lt;sup&gt;FE&lt;/sup&gt;</td>
<td>Peck directed at any of the ropes.</td>
</tr>
<tr>
<td>Board peck&lt;sup&gt;FE&lt;/sup&gt;</td>
<td>Peck directed at the blunting board.</td>
</tr>
<tr>
<td>Mat peck&lt;sup&gt;FE&lt;/sup&gt;</td>
<td>Peck directed at the pecking mat.</td>
</tr>
<tr>
<td>Other peck&lt;sup&gt;F&lt;/sup&gt;</td>
<td>Any other peck not listed above, including feeding, drinking, beak pecking, self-pecks, pecks directed to cage fixtures.</td>
</tr>
<tr>
<td>Receive peck&lt;sup&gt;F&lt;/sup&gt;</td>
<td>Any type of peck that the focal bird receives.</td>
</tr>
<tr>
<td>Bird change&lt;sup&gt;F&lt;/sup&gt;</td>
<td>Change in focal hen when original hens moves out of sight.</td>
</tr>
</tbody>
</table>

<sup>F</sup> Behaviours recorded during focal sampling only, and performed by the focal hen.
<sup>IB</sup> Behaviours grouped as a part of injurious pecking (IP).
<sup>B</sup> Bird-to-bird behaviours analysed together as a group (focal sampling only).
<sup>FE</sup> Behaviours directed at extra enrichments analysed as a group (focal sampling only).
3.3.5.3 Feather Cover

Following behavioural scan sampling, feather cover was assessed for four hens per cage, travelling along the tiers from north to south, following the same pattern as the order for scan sampling, although cage order within tier was reversed. Observed hens were selected from predetermined locations within the cage (nearest north perch, north side of scratch mat, south side of scratch mat, south perch). Hens were observed from outside the cage and given a feather cover score from 0 to 5 (where 0 was no damage; see (Savory et al., 1999)) for each visible body site (head, comb, neck, breast, back, both wings, rump, thigh, belly, and tail). The comb was given a score based on the number of scratches present up to a maximum of 5 (i.e. a comb with more than five scratches would still be given a score of 5) and was included in the analyses even though damage here most likely reflects aggression and not feather pecking.

3.3.5.4 Extra Enrichment Use

Damage to each extra enrichment was assessed and given a score at each visit. Ropes were given a score from 1 to 3 (1=no evidence of use; 2=minimal use, <50% frayed; 3=moderate to maximal use, ≥50% frayed). Mats and boards were given a score from 1 to 5 (1=no evidence of use; 2=minimal use, <25% worn; 3=minimal to moderate use, 25–50% worn; 4=moderate use, 51–75% worn; 5=maximal use, >75% worn). An extra enrichment was replaced once its individual score reached a certain threshold of damage (score≥4 for mats and boards, score=3 for ropes).

3.3.5.5 Beak Shape

At 64 weeks of age, beaks of four hens per cage were photographed from the side on a background of graph paper. Using tpsDig2 software (SUNY Stony Brook Morphometrics, USA), four landmark reference points were added (adapted from (Fahey et al., 2007)) to each of the photos and the length of the upper and lower mandibles were traced (Figure 3.1a). In addition, the
amount of upper or lower mandible overhang (negative numbers if lower mandible extended beyond upper mandible) and beak tip angles were measured (Figure 3.1b). The single observer analysing beak photos was blind to treatment.

![Figure 3.1](image)

**Figure 3.1** Screenshot of tpsDig2 software used to landmark reference points on the beak, and to perform precise measurements of upper (A) and lower mandible (B) lengths as well as overhang (C) length (a) and to measure the inner angle of the beak tip (b).

### 3.3.6 Statistical Analyses

Tests reported from linear mixed models (LMMs) and generalised mixed models (GLMMs) are approximate $F$ tests when these are available but otherwise Wald tests are reported, with threshold for statistical significance $\alpha=0.05$. Age was fitted as a covariate in the fixed effects with linear, quadratic and cubic functions of age included, when appropriate. Higher level interactions were omitted from fixed effects in GLMMs when required due to sparseness in the response variable. Negligible random effects were also omitted from GLMMs to aid computation. Means (±standard error (SE), as well as standard error of differences (SEDs)) reported are estimated from the mixed models and when data are transformed, or for GLMMs, estimates from
models of mean (mean+SE, mean-SE) are back transformed if applicable to aid interpretation. All data were analysed in Genstat (16th edition).

3.3.6.1 Mortality

Two cages were removed from the study at 48 weeks of age due to pecking related mortality exceeding the permitted threshold and all remaining hens in these cages were culled by cervical dislocation to avoid any further welfare problems. The total mortality data over the whole trial period (age 16 to 71 weeks) were analysed both excluding these healthy birds that were culled (‘minimum’ mortality) and including them (‘maximum’ mortality) and these were further divided into mortality due to all causes (i.e. ‘all’ mortality) and those that were deemed to be related to IP. Cumulative mortality to 48 weeks of age for all treatments were also analysed and the statistical significance did not differ from the ‘maximum’ mortality (week 48 results not presented here).

The resulting four types of proportions of dead birds were analysed by fitting GLMMs to the counts of dead birds per cage out of binomial total 80, with binomially distributed errors and logit link function (Table 3.2). Tier was included as a random effect in all models. Fixed effects were breed, beak status, and enrichment, including all interactions for all mortality and two way interactions for IP related mortality.
Table 3.2 Statistical models used for each of the parameters (Generalised Linear Mixed Models GLMM, and Linear Mixed Models LMM).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Test Type</th>
<th>Fixed Effects</th>
<th>Random Effects</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mortality</td>
<td>GLMM, binomial, logit link</td>
<td>Breed×beak×enrich.</td>
<td>Tier</td>
</tr>
<tr>
<td>Scan behaviour</td>
<td>N/A</td>
<td>N/A</td>
<td>N/A</td>
</tr>
<tr>
<td>Focal behaviour</td>
<td>LMM, natural log +1 or +0.1</td>
<td>Starting location+ age×breed×beak×enrich.</td>
<td>Tier/cage/age</td>
</tr>
<tr>
<td>Feather score</td>
<td>GLMM, binomial, logit link</td>
<td>Observer+body site× age×breed×beak×enrich.</td>
<td>Tier/cage/age/ bird</td>
</tr>
<tr>
<td>Enrichment damage</td>
<td>LMM, natural log+1</td>
<td>Observer+age×breed×beak</td>
<td>Tier/cage/age/ location</td>
</tr>
<tr>
<td>Beak shape</td>
<td>LMM, natural log (beak tip only)</td>
<td>Breed×beak×enrich.</td>
<td>Tier/cage/bird</td>
</tr>
</tbody>
</table>

3.3.6.2 Behaviour

Scan sampling data were too sparse for a statistical analysis, however, summary statistics are presented.

Bird-to-bird pecks during focal sampling were quite infrequent and therefore were analysed as a group as were pecks directed at extra enrichments (Table 3.1). Each type of peck (bird-to-bird, extra enrichment, other) as well as the number of bird changes were analysed by fitting LMMs to the rate (count per min) transformed as required (natural logarithm+0.1 for bird-to-bird pecks and extra enrichment pecks, natural logarithm+1 for number of bird changes, square root for ‘other’ pecks, Table 3.2). Age within cage within tier were included as random effects in all models. Fixed effects were starting location, age (as a covariate), breed, beak status, and enrichment (including up to three-way interactions between treatment factors and age). The model
for extra enrichment pecks was applied to data from cages with extra enrichments only and excluded enrichment from the fixed effects.

### 3.3.6.3 Feather Cover

The proportions of positive feather cover scores were analysed by fitting a GLMM to the binary variable of whether each score was greater than 0, or not, out of binomial total 1, with binomially distributed errors and logit link function (Table 3.2). Tier and cage, age within tier and within cage, and bird location within cage and age (i.e. the selected bird) were included as random effects. Fixed effects were observer, body site, age (as a covariate), breed, beak status, and enrichment. For the body site fixed effect, the two wing sites were combined into one level called “wing” and low scoring sites were also grouped together (head, comb, back, rump, breast and belly) and classified as “the rest”. Head and comb scores were included in these analyses (even though they may reflect aggression and not necessarily feather pecking) as significant results did not differ when they were removed. Interactions of observer with body site, and up to three way interactions of body site with age, and each of the treatment factors were included. However, interactions with beak status were limited to two way as the data were sparse.

### 3.3.6.4 Extra Enrichment Use

When any of the extra enrichments were replaced due to wear, its damage score was reset to ‘1’. Therefore, to account for changes in scores with age, the scores for each of the extra enrichments were cumulated over time. LMMs were fitted to the cumulative damage scores transformed (natural logarithm +1) for ropes and mats, but not to scores for the beak blunting boards as they typically scored ‘1’ at each visit and therefore the data were too sparse to support a statistical analysis (Table 3.2). The rope score data is only available from 19 to 51 weeks of age because one rope dislodged from the cage at 51 weeks, giving rise to concern about damage to the
egg belt mechanism, and thus all ropes were removed. Age within cage within tier, and location within cage (i.e. each individual rope or mat) were included as random effects in all models. Fixed effects were observer together with age (as a factor), breed, beak status, and their interactions.

### 3.3.6.5 Beak Shape

LMMs were fitted to beak measurements of lower and upper mandible lengths, beak tip angle (natural logarithm transformed), and overhang length (not transformed). Bird within cage within tier were included as random effects in all models (Table 3.2). Fixed effects were breed, beak status, enrichment, and their interactions. For upper and lower mandible measurements, 19% of photos were not useable (i.e. too blurry) and were thus excluded from analysis. For beak tip angle, a subset of the data was analysed that included only clear photos of NT hens (77%) as beaks of T hens were too blunt to measure an angle, and beak status was removed from the fixed effects. For the overhang length, a subset of the data (73%) was analysed that included only clear photos in which the hens had their beaks fully closed.

### 3.4 RESULTS

#### 3.4.1 Mortality

By 48 weeks of age, two cages (L-NT-NE and L-NT-EE) had surpassed the pecking related mortality threshold and required remedial action. The first cage (L-NT-NE) had a total of ten hens found dead (12.50%), seven (8.75%) of which had been identified by the stock worker as being related to IP (mostly vent pecking). The IP-related deaths occurred between 43 to 48 weeks of age. The second cage (L-NT-EE) had a total of eight hens found dead (10.00%), five (6.25%) of which had been identified by the stock worker as being related to IP. These IP-related deaths occurred
between 28 to 44 weeks of age. All remaining hens from both cages were removed from the study and culled.

L hens had significantly higher proportions of both minimum and maximum total mortality than H hens for both all (P<0.002) and IP-related (P<0.003) deaths (Table 3.3). There was a significant beak status effect for maximum mortality data only, as NT hens had higher proportions of mortality than the T hens for both all (P<0.001) and IP-related (P<0.001) mortality. There were no differences between NE and EE hens for all or IP-related mortality. There were marginally significant breed$x$beak status interactions for minimum all (P=0.039), maximum all (P=0.013), and maximum IP (P=0.025) mortality. H-T hens had fewer minimum all mortalities than H-NT hens, but no such pattern was observed for L hens, whilst for maximum all and IP mortality L-NT hens had substantially higher mortality than the other three groups. There was a marginally significant beak status$x$enrichment interaction for maximum all mortality (P=0.037), with the increase in mortality in NT-EE compared to T-EE slightly larger than the increase in mortality in NT-NE compared to T-NE.
### Table 3.3

Raw mean percentage (%) total mortality (16-71 weeks) and estimated means±SE on the logit scale together with P values (SED) for statistically significant fixed effects from the GLMMs. Maximum data includes all hens from the two cages that were removed from the study due to surpassing the pecking related threshold, whilst minimum data only includes hens from these cages that died before the these cages were removed. Breeds were Hyline Brown (H) and Lohmann Classic Brown (L), beak statuses included trimmed (T) or intact (NT), hens were either housed with extra enrichment (EE) or not (NE).

<table>
<thead>
<tr>
<th>Breed</th>
<th>Minimum All Deaths</th>
<th>P-value (SED)</th>
<th>Maximum All Deaths</th>
<th>P-value (SED)</th>
<th>Minimum IP-Related Deaths</th>
<th>P-value (SED)</th>
<th>Maximum IP-Related Deaths</th>
<th>P-value (SED)</th>
</tr>
</thead>
<tbody>
<tr>
<td>H</td>
<td>1.52% (-4.26±0.20)</td>
<td>0.002</td>
<td>1.52% (-5.24±0.44)</td>
<td>&lt;0.001</td>
<td>0.12% (-7.84±0.71)</td>
<td>0.003</td>
<td>0.12% (-8.88±1.07)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>L</td>
<td>2.97% (-3.34±0.16)</td>
<td>0.569</td>
<td>8.52% (-3.34±0.41)</td>
<td>&lt;0.001</td>
<td>0.35% (-6.34±0.63)</td>
<td>0.021</td>
<td>0.35% (-8.34±1.10)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Beak Status</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T</td>
<td>2.07% (-4.06±0.20)</td>
<td>0.001</td>
<td>2.07% (-4.60±0.44)</td>
<td>&lt;0.001</td>
<td>0.35% (-6.34±0.63)</td>
<td>0.210</td>
<td>0.35% (-8.34±1.10)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>NT</td>
<td>2.42% (-3.74±0.17)</td>
<td>0.569</td>
<td>7.97% (-3.33±0.42)</td>
<td>&lt;0.001</td>
<td>0.63% (-5.64±0.48)</td>
<td>0.076</td>
<td>0.63% (-6.04±0.92)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Breed×Beak</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>H-T</td>
<td>1.02% (-4.61±0.30)</td>
<td>0.210</td>
<td>1.02% (-5.15±0.49)</td>
<td>&lt;0.001</td>
<td>0.08% (-7.41±0.10)</td>
<td>0.08%</td>
<td>0.08% (-9.29±1.10)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>H-NT</td>
<td>2.03% (-3.91±0.23)</td>
<td>0.039</td>
<td>2.03% (-4.45±0.45)</td>
<td>&lt;0.001</td>
<td>0.16% (-6.64±0.78)</td>
<td>0.042</td>
<td>0.16% (-8.47±1.12)</td>
<td>0.025</td>
</tr>
<tr>
<td>L-T</td>
<td>3.13% (-3.52±0.20)</td>
<td>0.076</td>
<td>3.13% (-4.04±0.44)</td>
<td>&lt;0.001</td>
<td>0.63% (-5.26±0.46)</td>
<td>0.037</td>
<td>0.63% (-7.09±0.93)</td>
<td>0.076</td>
</tr>
<tr>
<td>L-NT</td>
<td>2.81% (-3.57±0.20)</td>
<td>0.076</td>
<td>13.91% (-2.21±0.41)</td>
<td>&lt;0.001</td>
<td>1.09% (-4.65±0.38)</td>
<td>0.076</td>
<td>12.19% (-3.52±0.85)</td>
<td>0.076</td>
</tr>
<tr>
<td>Beak×Enr.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T-NE</td>
<td>2.66% (-3.84±0.24)</td>
<td>0.104</td>
<td>2.66% (-4.36±0.46)</td>
<td>&lt;0.001</td>
<td>0.47% (-5.92±0.65)</td>
<td>0.047</td>
<td>0.47% (-7.74±1.03)</td>
<td>0.047</td>
</tr>
<tr>
<td>T-EE</td>
<td>1.48% (-4.29±0.27)</td>
<td>0.037</td>
<td>1.48% (-4.83±0.48)</td>
<td>&lt;0.001</td>
<td>0.23% (-6.75±0.87)</td>
<td>0.069</td>
<td>0.23% (-8.64±1.19)</td>
<td>0.375</td>
</tr>
<tr>
<td>NT-NE</td>
<td>2.34% (-3.79±0.22)</td>
<td>0.013</td>
<td>7.81% (-3.41±0.43)</td>
<td>&lt;0.001</td>
<td>0.70% (-5.45±0.54)</td>
<td>0.085</td>
<td>6.17% (-5.88±0.95)</td>
<td>0.82</td>
</tr>
<tr>
<td>NT-EE</td>
<td>2.50% (-3.69±0.21)</td>
<td>0.076</td>
<td>8.13% (-3.25±0.43)</td>
<td>&lt;0.001</td>
<td>0.55% (-5.83±0.64)</td>
<td>0.076</td>
<td>6.17% (-6.12±1.00)</td>
<td>0.076</td>
</tr>
</tbody>
</table>
3.4.2 Behaviour

Out of a possible 35,360 occurrences during scan sampling, only 43 observations (0.12%) were made of hens performing some type of bird-to-bird pecking. Of these 43, 74.4% were GFP, 14% were SFP, and 11.6% were aggressive pecks. The differences appeared to be minimal between treatments, but were too sparse to statistically analyse.

The total number of pecks observed during focal sampling was 30,038 over all 14 visits. Relatively few occurrences of bird-to-bird pecking were observed during focal sampling, including no observations of cannibalistic or vent pecks (Table 3.4). Other pecks were the most commonly observed, and GFP, pecks received by the focal hen, and rope pecks were the only other behaviours observed more than 1% of pecks (Table 3.4).

Table 3.4. Percentages (%) of each type of peck out of the total number of observed pecks during focal behaviour sampling.

<table>
<thead>
<tr>
<th>Peck Type</th>
<th>Percentage of Observed Pecks (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gentle feather peck</td>
<td>5.29%</td>
</tr>
<tr>
<td>Severe feather peck</td>
<td>0.94%</td>
</tr>
<tr>
<td>Cannibalistic peck</td>
<td>0%</td>
</tr>
<tr>
<td>Vent peck</td>
<td>0%</td>
</tr>
<tr>
<td>Toe peck</td>
<td>0.15%</td>
</tr>
<tr>
<td>Aggressive peck</td>
<td>0.48%</td>
</tr>
<tr>
<td>Rope peck</td>
<td>1.73%</td>
</tr>
<tr>
<td>Board peck</td>
<td>0.31%</td>
</tr>
<tr>
<td>Mat peck</td>
<td>0.97%</td>
</tr>
<tr>
<td>Other peck</td>
<td>84.27%</td>
</tr>
<tr>
<td>Receive peck</td>
<td>5.86%</td>
</tr>
</tbody>
</table>
In general, bird-to-bird pecks increased with age (P<0.001), and peaked around 47 weeks of age. Observations that began with hens near the nest and mat resulted in fewer bird-to-bird pecks than for hens beginning near the rope, with the board intermediate (starting location effect P<0.001; Table 3.5). L hens performed more bird-to-bird pecking than H hens (P=0.015) and NE hens performed more bird-to-bird pecking than EE hens (P=0.033). No significant differences due to beak status were observed (P=0.248). When analysed without aggressive pecks, the breed effect was reduced (P=0.058).

For pecks directed at the extra enrichments, there was an effect of age (P<0.001) and starting location as pecks directed at the extra enrichments generally decreased over time and were more often observed near the rope and mat than the nest and board (starting location effect P<0.001). There were no statistically significant effects of other treatment factors (P>0.207).

For other pecks, there was an effect of age (P<0.001) as they generally increased over time. NT appeared to have fewer other pecks for L hens but not for H hens (breed×beak status P=0.036 (SED=0.161): means estimated from LMM back transformed onto rate scale and estimated means±SE on transformed scale: H-T 3.873 (1.968±0.188), H-NT 4.256 (2.063±0.188), L-T 5.258 (2.293±0.188), L-NT 3.595 (1.896±0.190) pecks per minute).

Finally, the rate of bird changes per observation was higher for L hens than for H hens (P<0.001; Table 3.5) and slightly higher for NT hens than for T hens (P=0.048).
Table 3.5. Estimated overall means (mean-SE, mean+SE) back transformed on to rate (pecks/changes per minute) scale together with P values for statistically significant fixed main effects from the LMMs.

<table>
<thead>
<tr>
<th></th>
<th>Bird-to-Bird Pecks</th>
<th>P-value</th>
<th>Bird Changes</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breed</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>0.042 (0.016,0.073)</td>
<td>0.015</td>
<td>1.291 (1.221,1.364)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>L</td>
<td>0.163 (0.115,0.223)</td>
<td></td>
<td>1.919 (1.830,2.012)</td>
<td></td>
</tr>
<tr>
<td>Beak Status</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T</td>
<td>0.124 (0.083,0.173)</td>
<td>0.253</td>
<td>1.460 (1.385,1.538)</td>
<td>0.048</td>
</tr>
<tr>
<td>NT</td>
<td>0.067 (0.036,0.105)</td>
<td></td>
<td>1.719 (1.635,1.806)</td>
<td></td>
</tr>
<tr>
<td>Enrichment</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>NE</td>
<td>0.153 (0.106,0.209)</td>
<td>0.033</td>
<td>1.562 (1.483,1.643)</td>
<td>0.694</td>
</tr>
<tr>
<td>EE</td>
<td>0.048 (0.021,0.081)</td>
<td></td>
<td>1.612 (1.531,1.695)</td>
<td></td>
</tr>
<tr>
<td>Start Location</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nest</td>
<td>0.030 (0.002,0.065)</td>
<td></td>
<td>1.553 (1.455,1.654)</td>
<td></td>
</tr>
<tr>
<td>Board</td>
<td>0.105 (0.061,0.160)</td>
<td>&lt;0.001</td>
<td>1.814 (1.707,1.926)</td>
<td>0.046</td>
</tr>
<tr>
<td>Mat</td>
<td>0.022 (-0.004,0.054)</td>
<td></td>
<td>1.586 (1.488,1.688)</td>
<td></td>
</tr>
<tr>
<td>Rope</td>
<td>0.330 (0.239,0.447)</td>
<td></td>
<td>1.408 (1.316,1.504)</td>
<td></td>
</tr>
</tbody>
</table>

3.4.3 Feather Cover

As would be expected, the proportion of hens with feather damage scores >0 increased with age (P=0.001). Feather cover for L hens deteriorated more quickly than for H hens at all body sites except neck (breed P<0.001, age×breed×body site P<0.001; Figure 3.2a). For the other sites, tail scores appear to differ the most between the two breeds, with the rest having the smallest difference. The proportion of scores >0 was analysed, so this reached the maximum, 1, for all body sites (except “the rest”) at about 60 weeks of age. Therefore, the differences in feather cover for most sites occurred in the middle portion of this experiment although for the rest the difference between the two breeds was still apparent at the end of the study. Feather cover in most sites worsened more quickly with age for NT hens than for T hens (beak status P<0.001, age×beak
status $P=0.014$) and NT scored higher than T for each of the body sites except tail (beak status×body site $P<0.001$) from 25 to 37 weeks up to approximately 60 weeks of age (Figure 3.2b). As with breed, apparent differences for most sites occurred in the middle portion of this experiment, although for the rest the difference between the two breeds was still apparent at the end of the study. Though there was no overall effect of enrichment ($P=0.831$), EE hens had better wing feather quality, but poorer thigh feather quality, with no differences for neck, tail, or the rest (body site×enrichment $P=0.005$) and extra enrichments did not affect feather scores for T hens, but appear to have had a negative impact on feather scores for NT hens (beak status×enrichment $P=0.033$ ($\text{SED}=0.170$): means estimated from GLMM back transformed on to proportion scale and estimated means±SE on logit scale, T-NE 0.622 (0.499±0.148), T-EE 0.608 (0.438±0.154), NT-NE 0.774 (1.233±0.151), NT-EE 0.833 (1.604±0.159)).
Figure 3.2. Mean proportion feather cover score>0 (back transformed means±SE from logit scale) estimated from GLMM by a) breed (H: Hyline Brown, L: Lohmann Brown Classic) and b) beak status (T: Infra-red treated, NT: Not treated), and by body site and age.
3.4.4 Extra Enrichment Use

The cumulative damage scores for the pecking mats increased with age (P<0.001), which is to be expected, and L hens accumulated higher scores over time than H hens (age×breed P<0.001; Figure 3.3a). NT hens caused more damage to the mats than T hens (beak status P<0.001 (SED=0.083); mean±SE estimated from LMM on log+1 scale: T 1.78±0.08 score, NT 2.22±0.08 score). To give some context to these scores, based on the back transformed means at the end of the study, mats for NT and L hens were replaced more often than mats for T and H hens, respectively (T 2.4-2.9, NT 3.4-4.3, H 2.1-2.6, L 3.9-4.9 total mat replacements). The cumulative damage scores for the ropes during age 19 to 51 weeks increased with age (P<0.001), which is to be expected, and L hens caused more damage to the ropes beyond 23 weeks of age than H hens (age×breed P=0.004; Figure 3.3b). NT hens caused more damage to the ropes than T hens (beak status P<0.001, (SED=0.041); mean±SE estimated from LMM on log+1 scale: T 0.61±0.03, NT 0.80±0.03).

![Figure 3.3. Mean (±SE) cumulative damage scores (log+1 scale) by age estimated from LMMs for a) mats and b) ropes by breed (H, Hyline Brown; L, Lohmann Brown Classic). Average SED between breeds across ages for mat score (a): 0.127 and rope score (b): 0.040.](image-url)
3.4.5 Beak Shape

There were no statistically significant effects of breed on the four beak measurements, and for beak tip angle (non-beak treated hens only) there was no statistically significant effect of enrichment. NT hens had longer upper mandible lengths than T hens (P<0.001; mean (mean-SE,mean+SE) estimated from LMM back transformed: NT 2.14 cm (2.12,2.16), T 1.52 cm (1.51,1.53)) but there was no statistically significant effect of enrichment. On average, T hens had longer lower mandibles than NT hens (P=0.017; mean (mean-SE,mean+SE) estimated from LMM back transformed: NT 1.31 cm (1.29,1.32), T 1.36 cm (1.34,1.37)). For NE hens, there was no difference between T and NT, but T-EE hens had longer lower mandibles than NT-EE hens (beak status×enrichment P=0.021 (SED=0.022); means estimated from LMM back transformed and estimated mean±SE on transformed scale: T-NE 1.31 cm (0.273±0.018), T-EE 1.40 cm (0.336±0.017), NT-NE 1.31 cm (0.273±0.018), NT-EE 1.30 cm (0.261±0.018)). The only significant effect for the overhang measure of the beak was due to beak status (P<0.001, SED=0.017), as NT hens had longer overhang lengths for the top mandible than T hens (mean±SE estimated from LMM: NT 0.258±0.014 cm, T -0.103±0.014 cm, with negative values representing longer lower mandibles).

3.5 DISCUSSION

Overall, H hens performed better than the L hens, regardless of beak status. H hens had fewer mortalities, performed fewer bird-to-bird pecks, and had better feather cover. Similarly, the T hens in general fared better than the NT hens. T hens had fewer mortalities (but only due to culling of whole cages due to feather pecking) and better feather cover. In addition, both the H and T hens caused less damage to the extra enrichments than their respective L and NT counterparts.
Fewer differences were noted between EE and NE hens. EE hens performed fewer bird-to-bird pecks, but this did not correlate with any overall differences in mortality or feather cover. Furthermore, the extra enrichments had no effect on upper mandible length or our measurement of beak tip sharpness.

Differences in mortality may be due to different management requirements between the breeds. This experiment was carried out on a commercial farm, where the hens in the shed that were not part of the study were Hyline Brown. The staff at this particular farm had more experience with Hyline hens and therefore had probably adopted slight modifications to suit this breed for particular management techniques. Though the diets were formulated to meet age requirements of the hens, they may have originally been formulated to suit the Hyline Brown breed. In addition, management techniques to reduce IP that work for Lohmanns (e.g. reducing light intensity) may not be practical for Hylines as this has had previous detrimental effects for productivity in this particular farm (C Kirk, personal communications). The extent to which the management affected the outcome variables is uncertain, as inherent breed differences have been reported to exist in other research, with significant differences in overall activity levels as well as feather pecking behaviour (Albentosa et al., 2003). Not only do commercially available breeds behave differently, but there is evidence to suggest that feather pecking is a heritable trait (Kjaer and Sørensen, 1997), as two divergent lines with different propensities for feather pecking have been developed in recent years (Kjaer et al., 2001; Kalmendal and Bessei, 2012). In addition, certain breeds may be more suited to certain housing environments than other breeds and in our case, the H hens appeared to be better suited to a large furnished cage environment than the L hens, for which the maximum all mortality (8.5%) exceeded the expected rate of 4.9% based on the breed guideline (Lohmann GB Limited, 2012). However, it is important to note that this breed effect was being driven by the L-
NT treatment (13.9%), as maximum all mortality for L-T hens (3.1%) was within acceptable breed standards. Although mortality was very low in the H group, the H-NT hens (2.0%) appeared to have more mortality than H-T hens (1.0%), though this was marginally significant.

Though a relatively considerable amount of feather damage and mortality due to cannibalism was observed in this experiment, the overall level of observed IP does not appear to correspond to these measures. However, there is some evidence to suggest that only a small proportion of hens (i.e. <12%) in any given group actually perform the majority of IP behaviour (Bilčík and Keeling, 2000; Wysocki et al., 2010), and thus this may be difficult to observe in general. In this type of commercial system, video surveillance to collect behaviour data remotely is not easily accomplished. Therefore, direct behaviour observations were the best option, though this type of data collection presented its own problems as well (i.e. limited time available to observe, disruption to normal behaviour). Hens did not perform much pecking behaviour during observations, though differences were still detectable when all types of bird-to-bird pecks from focal observations were combined into one analysis. However, these hens had not been previously habituated to human presence, so the presence of the observer may have influenced behaviour. Though GFP may not be directly harmful to the recipient hen’s welfare, its presence in a repetitive manner may represent a welfare problem in itself, as it is generally observed occurring in a stereotyped manner (Kjaer and Vestergaard, 1999; Rodenburg et al., 2004b). In addition, some research has suggested that GFP is correlated to SFP (Rodenburg et al., 2008), however others have found no such link (Kjaer and Vestergaard, 1999). In addition, one study (Dixon et al., 2008) demonstrated a difference in morphology between GFP and SFP, with SFP being most similar to foraging pecks. L hens were observed to perform more bird-to-bird pecks which may have played a role in the differences observed for mortality and feather cover. They also appeared to be more
active, both in general, as reflected in the rate of bird changes during focal observations, as well as in regards to bird-to-bird pecking. Similarly, (Kjaer, 2009) found that hens from a high feather pecking line were more active than those from a low feather pecking line, suggesting a link between hyperactivity and the genetic basis for feather pecking.

Not surprisingly, the NT hens caused more feather damage, had more deaths (maximum total and IP mortalities), and caused more damage to the extra enrichments. It is well described in the literature that beak alterations can reduce mortality and feather damage (Jendral and Robinson, 2004) without having to affect the underlying behaviour itself (i.e. IP). NT hens were most likely more efficient with their beaks and so are likely to cause more damage with the same number of pecks as T hens (as there was no difference in IP rates in the current study). Another study (Prescott and Bonser, 2004) reported reduced feeding efficiency in beak treated hens, as measured by percent of pecks that resulted in successful acquisition of feed pellets, for beak treated hens compared to hens with intact beaks. A separate study reported a positive correlation between ground pecks and feather pecks within large groups of hens (Bilčík and Keeling, 2000). Though no direct correlations were made, our data also suggests that hens who pecked more frequently at the extra enrichments (i.e. L hens) also performed more bird-to-bird pecking (i.e. L hens). Ideally, pecking at enrichments would reduce the time spent feather pecking based on a shift in time budgets or based on a reduction in foraging frustration, and improve feather cover. When presented with enrichments, hens from other studies were observed to perform less feather pecking behaviour (Dixon et al., 2010) and aggression (Gvaryahu et al., 1994; Bubier, 1996). However, this did not appear to be the case in this current experiment, given the lack of an overall enrichment effect on feather cover, despite evidence to suggest that hens were in fact using the extra enrichments to some degree. Though there was a significant effect of extra enrichment presence for NT hens, it
did not have the expected effect. In fact, NT-EE hens appeared to have worse feather cover than NT-NE hens, though this effect was marginally significant and may not reflect true biological differences. In this experiment, the ends of the ropes had been sealed, which differed from other studies that used similar ropes (Jones and Carmichael, 1998; Jones et al., 2002). This may have impacted on their overall use and subsequent effect on the outcomes we measured.

Though the hens were using the ropes and mats (evident during behavioural observations as well as indicated by the damage to the extra enrichments), it did not appear that the blunting boards attracted much attention. Therefore, it is unlikely that the board would have had the chance to affect beak morphology, and this was reflected in the results. Generally, this type of device would be better suited on the bottom of the feed trough, where the hens would have to come in contact with the abrasive surface on a more regular basis (Fiks-Van Niekerk and Elson, 2005; Van der Weerd et al., 2006). It was not feasible to permanently alter the cages in any way as this experiment was carried out on a commercial farm. In addition, from a practical point of view, placing this type of enrichment on the bottom of the feed trough would not easily suit a trough with a chain feeder, limiting its potential application to barns with hopper feeders. A surprising result was the longer bottom mandibles observed in the T-EE hens compared to the NT-EE hens (without the same difference noted in the NE hens). This result was still evident even after removing some outlying data points, yet we are not certain as to why this result would exist.

3.6 CONCLUSIONS

In conclusion, we were able to show that breed choice plays an important role in regards to successful housing of laying hens. H hens had fewer mortalities overall and the increase in IP-
related mortality for NT hens was not as evident for H hens as it was for L hens. The levels of IP
related mortality and feather damage observed with the L hens were not within acceptable levels
according to the farm staff (C Kirk, personal communication) as well as the standards set out in
the breed guidelines (Lohmann GB Limited, 2012), and represent a high degree of insult to animal
welfare. However, these results appear to be mainly driven by the L-NT treatment combination,
though L-T mortality was still higher than H-T mortality. Not surprisingly, beak treatment was
effective at reducing (maximum) mortality and improving feather cover, which are important
welfare parameters for commercial production. Though the mat and rope enrichments were used
by the hens and reduced bird-to-bird pecking, overall they did not affect any of the main welfare
outcomes (i.e. mortality and feather cover). Presumably an increase in behavioural repertoire
would inherently improve welfare, but some modifications would be required to improve the extra
enrichments’ benefits prior to commercial applicability.
Chapter 4 Can animal protein, extra dietary fibre, or extra environmental enrichment reduce injurious pecking in caged laying hens?

A version of this chapter will be submitted for publication.
4.1 ABSTRACT

Laying hens with intact beaks are at risk of outbreaks of IP with more serious consequences, thus beak alteration is routinely used to reduce damage caused by IP, though it does not alleviate the underlying causes. This study assessed the effects of dietary protein source (control plant based (P) or inclusion of 50 g kg\(^{-1}\) pork meat and bone meal (A)), dietary fibre inclusion (control (C) or inclusion of 100 g kg\(^{-1}\) oat hulls (F)), and environment (control (N) or extra enrichment (E)) on mortality, behaviour, feather condition, and enrichment damage. Hyline Brown laying hens were housed in furnished cages from 16-35 weeks of age. Each of the eight treatments were replicated with eight cages of 21 hens/cage. Cages with extra enrichment contained ropes bunches, pecking mats, and beak blunting material at the bottom of the feed trough (the ropes and mats were inspected at 3-4 week intervals for damage). Behaviour data were collected over four days every two weeks (focal observations over three days, and scan observations on the fourth). Feather condition was assessed every four weeks based on remote feather scores of four hens/cage. Data were analysed in Genstat using generalised mixed models and linear mixed models with significance at \(\alpha=0.05\). There were no overall treatment effects on IP, but E hens performed less spot pecking (P<0.014), as did the F hens (P<0.002). Pecks directed at the enrichments increased with age (P<0.001), but were not affected by any treatment factors. However, the cumulative rope score increased more quickly over time for hens fed the A diet (P=0.003). Feather condition deteriorated over time (P<0.001), and the N hens had slightly more feather damage than the E hens (P=0.022). Overall, some minor welfare improvements were observed with extra fibre and extra enrichments.
4.2 INTRODUCTION

IP is an important multifactorial problem facing the poultry industry, affecting both production economics as well as animal welfare (Huber-Eicher and Sebo, 2001). This behaviour has been well documented and numerous attempts have been made to understand its etiology and to identify methods of reducing or eliminating its occurrence (Rodenburg et al., 2013). IP occurs in all housing types although it manifests differently in each housing type.

Though beak treatment is effective at reducing (but not completely eliminating) the feather damage and mortality related to IP, it does not resolve the underlying cause of IP itself. It is believed that feather pecking is a result of redirected ground pecks due to insufficient or inadequate foraging substrate (Blokhuis and Arkes, 1984; Blokhuis, 1986; Rodenburg et al., 2013), which is supported, amongst other things, by data that shows that the fixed action patterns for feather pecking are similar to those for foraging pecks (Dixon et al., 2008). In addition, hens kept on littered flooring perform less feather pecking than those housed on wire floors (Nicol et al., 2001). Furthermore, the same study showed that hens housed exclusively on wire flooring performed more feather pecking than hens given as little as 10 days access to litter. One advantage of furnished cages is that they reduce the spread of IP (or the number of affected hens) due to smaller group sizes (in comparison to non-cage systems), though the major disadvantage is that it is harder to provide foraging opportunities than in barn, aviary, or free range systems. An alternative solution to litter on the ground is to provide environmental enrichment to elicit foraging and exploratory pecks. For example, the presence of a plastic toy was associated with reduced aggression and mortality in caged laying hens (Bell et al., 1998). Additonally, McAdie et al. (2005) found a reduction in feather pecking and subsequent feather damage when hens were given access to white polypropylene rope, which had previously been shown to be attractive to hens (reviewed
in Jones et al. (2004)). This improvement was most evident when the ropes were presented from one day of age, but also effective when presented at 16 weeks of age. Some commercial free range farms have implemented the use of large sheets of crushed wood fibre glued to a mesh backing as a type of interactive enrichment. However, to the authors’ knowledge, its effects on behaviour in caged hens has not been studied. Beak blunting enrichments have also been used and some of the preliminary work shows promising results. Fiks-Van Niekerk and Elson (2005) reported shorter beak tips and reduced peck-related mortality when hens were housed with an abrasive material in the feed trough.

Incorporating biologically-relevant foraging substitutes can be difficult to manage in cages. However, because foraging makes up most of the appetitive phase of feeding behaviour, it is reasonable to expect that dietary factors requiring more time spent in the consummatory phase (i.e. feeding) would also have positive effects on the prevalence of feather pecking. There has been some evidence to suggest that diets high in fibre can reduce feather pecking. For example, Hartini et al. (2002) found that high fibre diets were associated with reduced mortality for ISA Brown laying hens, though this difference was only apparent for non-beak trimmed hens, but probably because mortality was already low for beak-altered hens fed control diets. Hens from high feather pecking lines ate more feathers than low feather pecking lines and subsequently had a higher rate of feed passage as a result of feather consumption (Harlander-Matauschek et al., 2006), which is similar to the effect of insoluble fibre (reviewed in Van Krimpen et al. (2007)). Finally, providing dietary fibre may be of more importance for caged hens as Harlander-Matauschek et al. (2007) found that they consumed more wood shavings in a test than loose-housed hens, suggesting that access to foraging substrate (e.g. litter for loose-housed hens) was better at satisfying the motivation to forage, whereas caged hens needed extra dietary fibre. It is hypothesised that dietary
fibre acts via multiple different mechanisms: 1. by altering hens’ time budgets toward feeding and away from IP (Rodenburg et al., 2013); 2. by aiding in the modulation of gizzard function (Hetland et al., 2004); and 3. by acting on the gut-brain feedback loop, where increasing satiety or satisfying foraging motivation can reduce IP (Rodenburg et al., 2013). In addition, in humans, increasing dietary fibre has been shown to improve mood and even cognitive function (Kaczmarczyk et al., 2012).

Other dietary elements can also impact behaviour. The quality and quantity of protein in the diet can have profound effects on overall health as well as performance of feather pecking (Kjaer and Bessei, 2013). Feather damage and mortality due to cannibalism increased in hens with decreasing levels of crude protein (from 19.3% to 11.1%) in the diet (Ambrosen and Petersen, 1997). In addition, purely plant-based diets have been criticised for not matching the naturally omnivorous diet of wild fowl (Kjaer and Bessei, 2013) though the effect of protein source on behaviour has not been widely studied. McKeegan et al. (2001) compared the development of feather pecking in laying pullets up to 24 weeks of age fed either a control plant-based diet or a diet containing fish meal. The control-fed pullets performed more feather pecking between 13-16 weeks of age, though feather damage scores did not reflect this difference.

The objective of this experiment was to determine the effects of dietary alterations, including protein source and extra fibre, and the addition of extra environmental enrichments on the behaviour and welfare of hens with intact beaks housed in furnished cages.
4.3 MATERIALS AND METHODS

4.3.1 Ethical Considerations

This study was conducted under a Home Office licence (Animals (Scientific Procedures) Act 1986) and was approved by the animal ethics committee at Scotland’s Rural College (SRUC). In addition, special permission to use animal by-products in poultry diets was obtained from the Animal Health and Veterinary Laboratories Agency, UK (Appendix III). Within the Home Office licence an intervention threshold was set at two or more birds from one cage dying or needing to be culled due to pecking related damage, due to the high risk nature of housing non-beak treated hens.

4.3.2 Animals

A total of 1344 non-beak treated Hyline Brown laying hens were used for this experiment (and were the same hens used in Chapter 4). From 0 to 7 weeks of age, all chicks were reared in deep-littered floor pens at a commercial farm in Scotland, UK. At 7 weeks of age, the pullets were moved into pullet colony cages for the remainder of the rearing period (to 15 weeks). Pullets had access to feed (Appendix I) and water on an *ad libitum* basis and were reared following standard commercial schedules for temperature and lighting regimens, and vaccination schedules.

4.3.2.1 General Husbandry

From 15 to 34 weeks of age, hens were housed in 21-hen furnished cages (2.40 m × 0.66 m or 754.3 cm²/hen, Valli S.p.A, Italy, see Appendix IV) at SRUC. Cages were arranged in two banks (eight cages long, two cages wide), each bank with two tiers, for a total of 64 cages. Every cage contained the furnishings required by law (European Council Directive 1999/74/EC), including a nesting area (35 cm × 66 cm), perches (15.2 cm/hen), and a scratching mat (28 × 28 cm, with a 58 cm long scratch feed trough), which was supplied with feed from feed auger above
it. Feeder trough space met the minimum European requirements (12 cm/hen), but only when the scratch trough was included (9.8 cm/hen without). The cages were illuminated by vertically hanging strips of LED lights outside the scratch areas. From 16 to 30 weeks of age, light intensity was set at 10 lux, but ranged from 3 lux (just outside the nest box) to 27 lux (opposite the scratch mat) (mean 10.9 ± 0.4 lux). At 30 weeks of age, the light intensity was increased slightly to encourage pecking and ranged from 6 lux to 39 lux (mean 17.9 ± 1.0 lux) for the remainder of the study. Pullets were photostimulated at 17 weeks of age (from 10L:14D to 11L:13D), and light duration increased incrementally from 17 – 30 weeks until 16L:8D was reached. A preventive red mite treatment was applied topically to the cages and hens at 19 weeks of age (Elector, Elanco Animal Health, Eli Lilly and Company Ltd., UK). All hens had ad libitum access to feed and water.

4.3.2.2 Experimental Treatments

This study was designed as a 2 × 2 × 2 factorial with eight treatments in total, based on the following factors: dietary protein source (control plant-based (P) or animal-based (A)), dietary fibre inclusion (control (C) or extra fibre (F)), and environment (control (N) or extra enrichment (E)). The 8 treatments were thus PCN, PCE, PFN, PFE, ACN, ACE, AFN, AFE. Treatments with A contained 50 g kg⁻¹ pork meat and bone meal (Omega Proteins Ltd., LeoGroup Ltd., Penrith, UK) and treatments with F included 100 g kg⁻¹ oat hulls (Morning Foods Ltd., Crewe, UK). Each E cage included three types of enrichments: eight polypropylene ropes (8 mm diameter), two pecking mats (ROWA, Melle, Germany), and a beak blunting board (S. N. Supplies, Lincoln, UK). The ropes (60 cm) were hung from the cage top in four bunches of two, folded in half such that each bundle was made up of four rope ends. The rope ends were left unsealed as there was no evidence in previous work (Chapter 3) that stray rope fibres were a health risk. The pecking mats (45 cm × 14 cm), which are designed to go on the floor in loose-housed systems, were fixed to the
cage fronts at two of the vertical struts. The beak blunting paste was adhered to a wooden board that was fixed to the bottom of the feed trough. In this way, the blunting board did not act as an extra pecking enrichment *per se*, but the aim was to blunt the tips of the beak as hens were pecking at the feed and hitting the bottom of the trough. Treatments were systematically allocated to cages prior to pullet placement and were balanced by blocks of eight cages on a single tier. Each treatment was represented once within each block.

When pullets arrived at the research centre at 15 weeks of age, they were weighed and wing tagged (Ketchum Mfg. Co, Inc., NY, USA) prior to placement. Pullets of similar weights were grouped together within cages to reduce variability within the cage and competition for feed and other resources. Each treatment was allocated two of the lightest groups, four medium-weight groups, and two of the heaviest groups to ensure overall means were similar between treatments.

### 4.3.3 Diet Formulation

Pullet diets were formulated by the rearing farm based on age and were fed *ad libitum* (Appendix I). During rearing, all pullets were fed the same diet.

Treatment diets were formulated by S. Pritchard from Premier Nutrition (Staffordshire, UK). The hens were fed one diet for the entirety of the experiment and the diets were formulated to meet the breed guidelines (Hy-Line International, 2012) for nutrient requirements (Table 4.1). Diets containing the same level of fibre were formulated to have similar energy and individual amino acid contents. The addition of extra fibre to certain treatments (PFN, PFE, AFN, and AFE) resulted in nutrient dilution (10%). For treatments that included the A diet, the inclusion of the meat and bone meal resulted in a reduction of soybean meal inclusion, as well as a slight adjustment of other ingredients to maintain similar nutrient levels. The diets were manufactured at
a commercial feed mill (Humphrey Feeds Ltd., Winchester, UK) (Table 4.2). Due to restrictions regarding commercial use of animal by-products, the meat and bone meal was added to treatments using the A diet in batches of 25 kg using a cement mixer at the research facility. As the use of animal by-products in poultry feed is strictly prohibited in the UK, special permission had to be attained from the Animal Health and Veterinary Laboratory Agency (AHVLA) division of the Scottish Government (Appendix III) to allow the addition of meat and bone meal to the treatment diets.
Table 4.1 Calculated nutritional components for diets based on formulation (16-34 weeks of age).

<table>
<thead>
<tr>
<th>Nutrient (%)</th>
<th>Hy-Line Guide</th>
<th>PC&lt;sup&gt;1&lt;/sup&gt;</th>
<th>PF&lt;sup&gt;2&lt;/sup&gt;</th>
<th>AC&lt;sup&gt;3&lt;/sup&gt;</th>
<th>AF&lt;sup&gt;4&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>DM</td>
<td>-</td>
<td>89.14</td>
<td>88.93</td>
<td>89.12</td>
<td>88.91</td>
</tr>
<tr>
<td>Crude protein</td>
<td>15.04-18.28</td>
<td>17.27</td>
<td>15.89</td>
<td>18.14</td>
<td>16.68</td>
</tr>
<tr>
<td>Oil B</td>
<td>-</td>
<td>5.11</td>
<td>4.78</td>
<td>4.20</td>
<td>3.96</td>
</tr>
<tr>
<td>Starch</td>
<td>-</td>
<td>34.58</td>
<td>31.52</td>
<td>36.02</td>
<td>32.82</td>
</tr>
<tr>
<td>Sugar</td>
<td>-</td>
<td>3.73</td>
<td>3.51</td>
<td>3.41</td>
<td>3.22</td>
</tr>
<tr>
<td>Crude Fibre</td>
<td>-</td>
<td>3.10</td>
<td>5.79</td>
<td>3.11</td>
<td>5.80</td>
</tr>
<tr>
<td>Ash</td>
<td>-</td>
<td>13.63</td>
<td>12.77</td>
<td>12.78</td>
<td>12.00</td>
</tr>
<tr>
<td>ME (MJ/kg)</td>
<td>11.63-12.18</td>
<td>11.50</td>
<td>10.50</td>
<td>11.50</td>
<td>10.50</td>
</tr>
<tr>
<td>Linoleic acid</td>
<td>0.88-1.08</td>
<td>2.21</td>
<td>2.04</td>
<td>1.56</td>
<td>1.46</td>
</tr>
<tr>
<td>Dig. lysine</td>
<td>0.75-0.91</td>
<td>0.77</td>
<td>0.69</td>
<td>0.77</td>
<td>0.69</td>
</tr>
<tr>
<td>Dig. M+C</td>
<td>0.63-0.77</td>
<td>0.67</td>
<td>0.60</td>
<td>0.67</td>
<td>0.60</td>
</tr>
<tr>
<td>Dig. threonine</td>
<td>0.53-0.64</td>
<td>0.53</td>
<td>0.47</td>
<td>0.54</td>
<td>0.48</td>
</tr>
<tr>
<td>Dig. tryptophan</td>
<td>0.16-0.19</td>
<td>0.20</td>
<td>0.18</td>
<td>0.19</td>
<td>0.17</td>
</tr>
<tr>
<td>Dig. isoleucine</td>
<td>0.59-0.72</td>
<td>0.60</td>
<td>0.54</td>
<td>0.60</td>
<td>0.54</td>
</tr>
<tr>
<td>Dig. valine</td>
<td>0.68-0.82</td>
<td>0.70</td>
<td>0.63</td>
<td>0.72</td>
<td>0.65</td>
</tr>
<tr>
<td>Calcium</td>
<td>3.63-4.41</td>
<td>4.00</td>
<td>3.61</td>
<td>4.00</td>
<td>3.61</td>
</tr>
<tr>
<td>Av. phosphorus</td>
<td>0.41-0.49</td>
<td>0.39</td>
<td>0.35</td>
<td>0.39</td>
<td>0.36</td>
</tr>
<tr>
<td>Sodium</td>
<td>0.16-0.19</td>
<td>0.16</td>
<td>0.15</td>
<td>0.16</td>
<td>0.15</td>
</tr>
<tr>
<td>Salt</td>
<td>-</td>
<td>0.40</td>
<td>0.37</td>
<td>0.35</td>
<td>0.33</td>
</tr>
</tbody>
</table>

<sup>1</sup> PC, plant based diet, with no extra fibre (control diet)
<sup>2</sup> PF, plant based diet, with extra fibre (100 g kg<sup>-1</sup> oat hulls)
<sup>3</sup> AC, animal based protein (50 g kg<sup>-1</sup> pork meat and bone meal), with no extra fibre
<sup>4</sup> AF, animal based protein, with extra fibre
Table 4.2 Treatment diet ingredients (16 to 34 weeks of age).

<table>
<thead>
<tr>
<th>Ingredient (% inclusion)</th>
<th>PC$^1$</th>
<th>PF$^2$</th>
<th>AC$^3$</th>
<th>AF$^4$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wheat</td>
<td>60.18</td>
<td>54.17</td>
<td>63.31</td>
<td>56.98</td>
</tr>
<tr>
<td>Oat Hulls</td>
<td>-</td>
<td>10.00</td>
<td>-</td>
<td>10.00</td>
</tr>
<tr>
<td>Hipro Sunflower Ext.</td>
<td>5.00</td>
<td>4.50</td>
<td>5.00</td>
<td>4.50</td>
</tr>
<tr>
<td>Hipro Soya</td>
<td>19.84</td>
<td>17.85</td>
<td>15.63</td>
<td>14.07</td>
</tr>
<tr>
<td>Pork Meat and Bone Meal</td>
<td>-</td>
<td>-</td>
<td>5.00</td>
<td>4.50</td>
</tr>
<tr>
<td>Soya Acid Oil</td>
<td>3.19</td>
<td>2.87</td>
<td>1.78</td>
<td>1.60</td>
</tr>
<tr>
<td>Limestone</td>
<td>9.66</td>
<td>8.70</td>
<td>8.53</td>
<td>7.68</td>
</tr>
<tr>
<td>Salt</td>
<td>0.30</td>
<td>0.27</td>
<td>0.22</td>
<td>0.19</td>
</tr>
<tr>
<td>Sodium Bicarbonate</td>
<td>0.03</td>
<td>0.03</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>MCP</td>
<td>1.21</td>
<td>1.09</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Lysine HCl</td>
<td>0.07</td>
<td>0.07</td>
<td>0.04</td>
<td>0.04</td>
</tr>
<tr>
<td>Methionine (liquid)</td>
<td>0.21</td>
<td>0.19</td>
<td>0.20</td>
<td>0.18</td>
</tr>
<tr>
<td>HF Organic Layer 751</td>
<td>0.30</td>
<td>0.27</td>
<td>0.30</td>
<td>0.27</td>
</tr>
</tbody>
</table>

1 PC, plant based diet, with no extra fibre (control diet)
2 PF, plant based diet, with extra fibre (100 g kg$^{-1}$ oat hulls)
3 AC, animal based protein (50 g kg$^{-1}$ pork meat and bone meal), with no extra fibre
4 AF, animal based protein, with extra fibre

4.3.4 Data Collection

Data collection commenced three days after arrival when pullets were 16 weeks of age and continued until they were 34 weeks of age. The hens were checked on a daily basis and mortalities and culls were recorded as and when they occurred. Dead hens were sent for post-mortem analyses to ascertain cause of death (especially in relation to IP).

4.3.4.1 Behavioural Measures

4.3.4.1.1 Focal Sampling

A single observer performed focal behaviour sampling on three consecutive days every two weeks from 16 to 34 weeks of age in order to catch brief behavioural events (e.g. bird-to-bird
pecking) that were of interest to the study which might be missed in scan sampling. To facilitate live observations, a handheld machine (Psion Workabout Pro\textsuperscript{3}, Motorola Solutions, Schaumburg, IL, USA) was used to collect data via Pocket Observer software (Observer XT v11, Noldus Information Technology, Wageningen, The Netherlands). On each observation day, one hen per cage was selected based on proximity to one of three predetermined locations (nearest to the nest, nearest to the front rope farthest from the nest box, and nearest to the south mat) and observed for five minutes. All pecking related behaviours performed by and directed toward the focal hen were recorded (Table 4.3). If the original focal hen moved out of sight, the observer chose another hen from the original starting position to resume the observation and this change in hens was recorded. One hen from each cage was observed at one location per day, and all three starting locations were represented for each cage during every observation week. All observations were done as quietly as possible so as to cause the least disturbance to the hens. To spread the observations out over the day, but within reasonable observable hours, scans started at 18\% of the total light period from the time lights came on in the morning and finished by 18\% of the light period before lights went out (i.e. if light period was 10 h, observations would start 1 h 48 min after lights on and end 1 h 48 min prior to lights out). Pullets were photostimulated at 17 weeks of age (from 10L:14D to 11L:13D), and light duration increased incrementally from 17–30 weeks until 16L:8D was reached.
Table 4.3 Ethogram for behaviours observed during focal and scan sampling. During focal sampling all incidences of any of the behaviours were recorded if performed by the focal hen or if any of the first seven behaviours were directed toward the focal hen. During scan sampling, the number of hens performing any of the behaviours was recorded.

<table>
<thead>
<tr>
<th>Behaviour</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gentle feather peck&lt;sup&gt;B&lt;/sup&gt;</td>
<td>Furtive, deliberate, often repeated pecks at another hen’s plumage where the recipient usually does not react.</td>
</tr>
<tr>
<td>Severe feather peck&lt;sup&gt;B&lt;/sup&gt;</td>
<td>Peck directed at feathers that include grasping and pulling of the feather, and may include feather removal. Recipient often reacts or withdraws.</td>
</tr>
<tr>
<td>Cannibalistic peck&lt;sup&gt;B&lt;/sup&gt;</td>
<td>Peck causing tissue damage. Directed toward any part of the body except for the vent (vent peck) or toes (toe peck).</td>
</tr>
<tr>
<td>Vent peck&lt;sup&gt;B&lt;/sup&gt;</td>
<td>Peck to the vent area.</td>
</tr>
<tr>
<td>Toe peck&lt;sup&gt;B&lt;/sup&gt;</td>
<td>Peck at another hen’s toes.</td>
</tr>
<tr>
<td>Aggressive peck</td>
<td>Overt, rapid, forceful, usually downward directed peck, usually directed toward recipient’s head or back of neck. Usually not overly repetitive, recipient withdraws or reacts immediately.</td>
</tr>
<tr>
<td>Beak peck&lt;sup&gt;F&lt;/sup&gt;</td>
<td>Peck directed toward the beak of a conspecific.</td>
</tr>
<tr>
<td>Rope peck&lt;sup&gt;E&lt;/sup&gt;</td>
<td>Peck directed toward any of the ropes.</td>
</tr>
<tr>
<td>Mat peck&lt;sup&gt;E&lt;/sup&gt;</td>
<td>Peck directed toward any of the mats.</td>
</tr>
<tr>
<td>Feed peck</td>
<td>Peck directed toward the feed in the feed trough.</td>
</tr>
<tr>
<td>Spot peck</td>
<td>Peck at cage walls, flooring, feed trough, nest box walls, etc.</td>
</tr>
<tr>
<td>Drinker peck&lt;sup&gt;F&lt;/sup&gt;</td>
<td>Peck directed toward the nipple drinker, or at the drinker cup.</td>
</tr>
<tr>
<td>Other peck&lt;sup&gt;F&lt;/sup&gt;</td>
<td>Any other peck not specified above (e.g. ‘air peck’, preening peck).</td>
</tr>
</tbody>
</table>

<sup>B</sup> Bird-to-bird pecking behaviour grouped together for statistical analyses;  
<sup>E</sup> Pecking directed at the enrichments were grouped together for statistical analyses;  
<sup>F</sup> Behaviours only recorded during focal sampling and not during scan sampling
4.3.4.1.2 Scan Sampling

On the day following the three days of focal sampling, scan sampling was used to record the number of hens performing any type of oral behaviour within a cage at each scan, for a total of six scans per cage per day. All types of bird-to-bird pecks were recorded, as well as pecks directed at the enrichments (mats and ropes), the feed, or parts of the cage (see Table 4.3; drinker, beak, and other pecks were not recorded during scan sampling). To spread the observations out over the day, scans started 10% into the total light period from the time lights came on in the morning and ended with 10% of the light period remaining before lights-out.

4.3.4.2 Feather Cover

Feather cover was scored at 16, 19, 23, 27, 31, and 34 weeks of age. At each assessment, four hens were selected based on their proximity to four different predetermined locations within the cage (the three used for focal sampling plus another nearest to the opposite pecking mat) and scored on twelve body parts (comb, head, neck, back, breast, both wings, both thighs, rump, belly, and tail). Hens were observed remotely (adapted from Bright et al. (2006), using similar methods to those used in Chapter 3) and each body part was given a damage score from 0 to 5 (where 0 was no sign of damage and increasing scores corresponded with increasing damage, see Savory et al. (1999)). The comb was given a score based on the number of scratches present up to a maximum of 5 (i.e. a comb with more than five scratches would still be given a score of 5). Head and comb scores were analysed along with damage on other body parts even though damage to these areas may reflect levels of aggression rather than feather pecking.

4.3.4.3 Extra Enrichment Damage

For cages fitted with extra enrichments, the level of damage to each of the ropes and mats was assessed and scored at 19, 23, 27, 31, and 34 weeks of age. The beak blunting boards were
not scored as they were located in the feed troughs and covered with feed. Each of the ropes and mats were given an individual score on a scale of 1 to 5 (Table 4.4). Pecking mats were considered damaged when the plastic mesh backing was visible. An enrichment was replaced at a score of ≥4.

Table 4.4 Scoring system used to quantify enrichment damage.

<table>
<thead>
<tr>
<th>Score</th>
<th>Characteristics</th>
<th>Rope</th>
<th>Pecking Mat</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>No use</td>
<td>No evidence of use</td>
<td>No evidence of use</td>
</tr>
<tr>
<td>2</td>
<td>Minimal use</td>
<td>≤50% unravelled, ≤25% frayed</td>
<td>≤25% damaged</td>
</tr>
<tr>
<td>3</td>
<td>Minimal-moderate use</td>
<td>&gt;50% unravelled, ≤25% frayed</td>
<td>26–50% damaged</td>
</tr>
<tr>
<td>4</td>
<td>Moderate use</td>
<td>≤50% unravelled, &gt;25% frayed</td>
<td>51–75% damaged</td>
</tr>
<tr>
<td>5</td>
<td>Maximal use</td>
<td>&gt;50% unravelled, &gt;25% frayed</td>
<td>&gt;75% damaged</td>
</tr>
</tbody>
</table>

4.3.5 Statistical Analyses

All data were analysed using Genstat (16th edition, VSN International, Hemel Hempstead, UK) with a threshold of significance at α=0.05. Both linear mixed models (LMMs) and generalised mixed models (GLMMs) were used, and results reported are approximate $F$ tests (when available) or Wald tests. When data were sparse, higher level interactions were excluded from the fixed effects in GLMMs. In the fixed effects, hen age was fitted as a covariate, with linear, quadratic, and cubic functions of time.
4.3.5.1 Behavioural Measures

4.3.5.1.1 Focal Sampling

Certain behaviours were grouped together to facilitate analysis as data were relatively sparse. All bird-to-bird pecks (excluding beak pecks and aggressive pecks) were combined into one category (this included GFP and SFP, cannibalistic pecks, vent pecks, and toe pecks that were given or received by the focal hens). Similarly, pecks directed at the ropes and mats were combined into one category called ‘enrichment pecks’. A GLMM was fitted to the counts of each type of beak related behaviour (bird-to-bird, feed, enrichment, spot, drinker, beak, and other pecks as well as bird changes) with observation starting location, age, and the treatment factors as the fixed effects (Table 4.5). Enrichment was removed from the fixed effects when analysing pecks directed at the enrichments. A natural logarithm link function was applied and a Poisson distribution was assumed. The response data were offset by the transformed duration of each observation (natural logarithm). The random model included cage, age, and observation day.
Table 4.5 Statistical models for each variable, including fixed and random effects models. Tests used were either linear or generalised mixed models (LMM or GLMM). Age is always a factor when included in the random model. Interaction levels were restricted to three-way for all tests.

<table>
<thead>
<tr>
<th>Test</th>
<th>Fixed Model</th>
<th>Random Model</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Focal Behaviour</strong></td>
<td>GLMM</td>
<td><strong>Random Model</strong></td>
</tr>
<tr>
<td></td>
<td>Startinglocation+age(covariate)<em>protein</em>fibre*enrichment</td>
<td>Cage+cage.age+age.day</td>
</tr>
<tr>
<td><strong>Scan Behaviour</strong></td>
<td>GLMM</td>
<td>Age(covariate)<em>protein</em>fibre*enrichment</td>
</tr>
<tr>
<td><strong>Feather Cover</strong></td>
<td>GLMM</td>
<td>Bodypart+age(covariate)<em>protein</em>fibre*enrichment+bodypart.protein+bodypart.fibre+bodypart.enrichment</td>
</tr>
<tr>
<td><strong>Enrichment Damage</strong></td>
<td>LMM</td>
<td>Age(factor)<em>protein</em>fibre</td>
</tr>
</tbody>
</table>

1 Enrichment was removed as a factor from fixed effects model when pecks at enrichments were being tested.

4.3.5.1.2 Scan Sampling

Behaviours that were similar in etiology or too sparse to be analysed separately were grouped. This included GFP and SFP, cannibalistic, vent, and toe pecks, which were combined to make a category ‘bird-to-bird pecks’ and rope and mat pecks, which were combined to make a category ‘enrichment pecks’. For each behaviour grouping, a GLMM (Table 4.5) was fitted to analyse the proportion of hens performing the behaviour out of the total number of hens in the cage (i.e. 21 except for cages with deaths). A logit link function was used and binomially distributed errors were assumed. For each test, the fixed effects model included age (as a covariate) and each of the treatment factors (up to a three way interaction). Enrichment was removed as a factor from the fixed effects model when analysing extra enrichment pecks. The random model was the same for all of the analyses and it included cage and age within cage.
4.3.5.2 Feather Cover

Feather cover scores (0-5) were converted to a binary dataset (where score 0 = 0, scores 1-5 = 1), and were analysed by fitting a GLMM to the proportion of scores greater than zero. This model was fitted with a logit link function and assumed binomially distributed errors. The scores from both wings were combined into one category, as were the scores from both thighs. In addition, the scores for some of the low-scoring body parts were grouped together and called “the rest” (head, comb, back, rump, breast, and belly). Though comb and head scores may reflect aggressive pecking rather than feather pecking, these data were still included in the final analysis, as the results were the same with or without them. The fixed effects model included body part, age (as a covariate), protein source, fibre inclusion, extra enrichment, and the appropriate interactions (Table 4.5). Bank, tier, cage, age (as a factor), and bird location within the cage were accounted for within the random effects model.

4.3.5.3 Extra Enrichment Damage

The damage scores for the ropes and mats were cumulated over time and LMMs were fitted to transformed data (natural logarithm +1). Both of the fixed effects models included age, protein source, fibre inclusion, and their interactions (Table 4.5). The random effects models included cage, age, and enrichment location within the cage.

4.4 RESULTS

4.4.1 Feed Analyses

Each of the diets came in two or more batches from the feed mill. A sample from each batch was sent for nutrient analysis (company) and these results were averaged for comparison
purposes (Table 4.6). Batch 2 of diet PC was found to be calcium deficient and thus more calcium was added to the diets after being shipped to the research farm and were sampled again to verify appropriate calcium levels before being fed to the hens.

Overall, diet analyses were similar to what was expected, however the crude protein in the PC diet was lower than the calculated value, and lower than the PF diet. In addition, the crude fibre levels were lower than expected for the PF and AF diets. Differences in the amino acids between calculated and analysed may be due to differences in digestible amino acids (calculated values) and total amino acids (analysed values).

Table 4.6 Analysed nutritional components for treatment diet formulations (averaged over two or three batches) from 16 to 34 weeks of age.

<table>
<thead>
<tr>
<th>Nutrient</th>
<th>PC1</th>
<th>PF2</th>
<th>AC3</th>
<th>AF4</th>
</tr>
</thead>
<tbody>
<tr>
<td>DM (%)</td>
<td>89.43</td>
<td>89.07</td>
<td>88.87</td>
<td>89.46</td>
</tr>
<tr>
<td>Crude protein (%)</td>
<td>15.90</td>
<td>16.35</td>
<td>17.85</td>
<td>15.87</td>
</tr>
<tr>
<td>Oil B (%)</td>
<td>5.37</td>
<td>4.60</td>
<td>4.20</td>
<td>4.23</td>
</tr>
<tr>
<td>Crude Fibre (%)</td>
<td>3.60</td>
<td>4.95</td>
<td>3.00</td>
<td>4.37</td>
</tr>
<tr>
<td>Ash (%)</td>
<td>11.17</td>
<td>11.20</td>
<td>12.50</td>
<td>12.83</td>
</tr>
<tr>
<td>GE (MJ/kg)</td>
<td>16.00</td>
<td>15.60</td>
<td>15.02</td>
<td>15.06</td>
</tr>
<tr>
<td>Tot. lysine (%)</td>
<td>0.86</td>
<td>0.86</td>
<td>0.86</td>
<td>0.83</td>
</tr>
<tr>
<td>Tot. M+C (%)</td>
<td>0.62</td>
<td>0.56</td>
<td>0.58</td>
<td>0.56</td>
</tr>
<tr>
<td>Tot. threonine (%)</td>
<td>0.60</td>
<td>0.58</td>
<td>0.62</td>
<td>0.59</td>
</tr>
<tr>
<td>Tot. isoleucine (%)</td>
<td>0.67</td>
<td>0.68</td>
<td>0.67</td>
<td>0.65</td>
</tr>
<tr>
<td>Tot. valine (%)</td>
<td>0.79</td>
<td>0.79</td>
<td>0.81</td>
<td>0.78</td>
</tr>
<tr>
<td>Calcium (%)</td>
<td>3.55</td>
<td>3.79</td>
<td>4.10</td>
<td>4.03</td>
</tr>
<tr>
<td>Tot. phosphorus (%)</td>
<td>0.50</td>
<td>0.57</td>
<td>0.63</td>
<td>0.58</td>
</tr>
</tbody>
</table>

1 PC, plant based diet, with no extra fibre (control diet)
2 PF, plant based diet, with extra fibre (10% oat hulls)
3 AC, animal based protein included (5% pork meat and bone meal), with no extra fibre
4 AF, animal based protein included, with extra fibre
4.4.2 Mortality

Overall, mortality was low (0.7%) and below the standards for hens at 34 weeks of age (1.0%) set out by the breed guidelines (Hy-line International, 2012). In total, three hens were culled and six were found dead. One hen was culled due to injuries from aggressive pecking. The remaining culls/deaths were due to: egg peritonitis (n=3); head wound due to cage furnishings (n=1); fatty liver haemorrhage (n=1); necrotic intestinal tissues (n=1); and unknown (n=2). Data were too sparse to analyse statistically, but each treatment combination had one or two deaths, except for PCN and PCE which had none.

4.4.3 Behavioural Measures

4.4.3.1 Focal Sampling

During focal sampling, the majority of observed pecks were directed at the feed (Table 4.7). Within bird-to-bird pecks, GFPs were observed most frequently, with vent and cannibalistic pecks not observed at all.
Table 4.7 Breakdown of individual behaviours during focal and scan sampling observations.

<table>
<thead>
<tr>
<th>Peck Type</th>
<th>Focal Sampling (% pecks)</th>
<th>Scan Sampling (% hens)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gentle feather peck*</td>
<td>1.02%</td>
<td>0.77%</td>
</tr>
<tr>
<td>Severe feather peck*</td>
<td>0.02%</td>
<td>0.02%</td>
</tr>
<tr>
<td>Vent peck*</td>
<td>0%</td>
<td>0%</td>
</tr>
<tr>
<td>Cannibalistic peck*</td>
<td>0%</td>
<td>0.01%</td>
</tr>
<tr>
<td>Aggressive peck*</td>
<td>0.06%</td>
<td>0.03%</td>
</tr>
<tr>
<td>Toe peck*</td>
<td>0.01%</td>
<td>0%</td>
</tr>
<tr>
<td>Mat peck</td>
<td>0.93%</td>
<td>0.98%</td>
</tr>
<tr>
<td>Rope peck</td>
<td>1.83%</td>
<td>6.15%</td>
</tr>
<tr>
<td>Feed peck</td>
<td>73.89%</td>
<td>84.34%</td>
</tr>
<tr>
<td>Spot peck</td>
<td>9.71%</td>
<td>7.71%</td>
</tr>
<tr>
<td>Drinker peck</td>
<td>7.70%</td>
<td>N/A</td>
</tr>
<tr>
<td>Beak peck</td>
<td>0.95%</td>
<td>N/A</td>
</tr>
<tr>
<td>Other peck</td>
<td>3.87%</td>
<td>N/A</td>
</tr>
<tr>
<td>Totals</td>
<td>294,018 pecks</td>
<td>19,410 observations</td>
</tr>
</tbody>
</table>

*Pecks both given and received by the focal hen in these categories were combined for focal sampling.

The results presented for focal sampling are back transformed rates of the performance of each behaviour. The starting location of the focal observation had a significant effect for all analysed behaviours (Figure 4.1, P<0.009). Feed and spot pecks occurred less frequently when observations began near the rope, while ‘other’ were more frequent near the rope. Spot pecks were more frequent when the chosen hen was near the nest, whereas bird-to-bird, beak and enrichment pecks were less frequent when observations began in this location.
Figure 4.1 Effect of starting location on the rate (at time offset = 0) of feed pecks, all other types of pecks, and number of bird changes observed during focal sampling. Average SEDs between starting locations: feed pecks=0.006, spot pecks=0.017, drinker pecks= 0.017, other pecks=0.026, beak pecks=0.049, bird-to-bird pecks=0.050, enrichment pecks=0.033, and number of bird changes=0.114.

The main effects of age, protein source, and extra fibre were not significant factors for bird-to-bird pecking (P>0.211). There was, however, a non-significant tendency for extra enrichments to reduce bird-to-bird pecking (P=0.053 (SED=0.215): means estimated from GLMM back transformed to rate scale and estimated means±SE on natural log scale, N 0.13 (-2.07±0.20), E 0.09 (-2.40±0.21)). There was a significant interaction of age × protein source × extra fibre on bird-to-bird pecking (P=0.044, Figure 4.2a), where the AC hens appeared to perform less bird-to-bird pecking than the other treatments at 28 and 30 weeks of age.

There were no treatment effects on pecks directed at the enrichments or other pecks.

Spot pecks generally decreased with age (P=0.002), extra fibre (P=0.002), and extra enrichments (P=0.013). However, there was an age × extra fibre × extra enrichment interaction associated with spot pecks (P=0.041, Figure 4.2b), as extra enrichments appeared to reduce spot
pecking for the extra fibre diet but not for the control diet. Protein source did not affect spot pecks (P=0.830).

Feed pecks increased with age (P<0.001), but neither protein source, fibre level or extra enrichments were significant factors (P>0.183). However, there was an age × fibre interaction (P=0.044, Figure 4.2c) with rate of feed pecks higher at 34 weeks of age with control fibre levels, but lower at 16 weeks of age.

Pecks at the drinker were affected by age (P=0.014, Figure 4.2d) as there appeared to be a cuboidal effect over time. The main treatment effects were not significant (P>0.122), though there was a protein source × extra enrichment effect (P=0.012, Figure 4.3a) as extra enrichment reduced drinker pecks for the plant based diet, but not for the animal protein based diet.

Beak pecks decreased significantly with age (P=0.042, Figure 4.2e) and there was a three-way interaction between the treatment factors (P=0.045, Figure 4.3b on beak pecks), though there were no overall main effects of the treatments (P>0.110).

Finally, the number of bird changes was affected by age (P=0.024), age × protein source (P=0.033), and age × protein source × extra fibre (P=0.044, Figure 4.2f), though there were no overall main effects of the treatments (P>0.122).
Figure 4.2 Rate (predicted means±SE back transformed from natural logarithm scale) over time for a) bird-to-bird pecks, b) spot pecks, c) feed pecks, d) drinker pecks, e) beak pecks, and f) bird changes during focal behaviour sampling. Treatments include plant based diet (P), diet containing animal protein (A), control level of fibre (C), increased fibre content (F), cages with extra enrichment (E) and cages without (N).
Figure 4.3 Rate (predicted means±SE back transformed from natural logarithm scale) of a) drinker pecks and b) beak pecks during focal behaviour sampling. Treatments include plant based diet (P), diet containing animal protein (A), control level of fibre (C), increased fibre content (F), cages with extra enrichment (E) and cages without (N). Average SEDs on transformed scale for a=0.352, and b=0.496.

4.4.3.2 Scan Sampling

Similar to the pattern observed during focal sampling, feed pecks constituted the majority of the observations. Pecks at the ropes were observed more frequently than pecks at the mats. For the combined bird-to-bird pecking category, GFP accounted for the majority of the data, while there were no observations of vent or toe pecks.

For bird-to-bird pecking, none of the main effects of age, protein source, fibre level, or extra enrichments were significant (P>0.130). However, bird-to-bird pecking was affected by an age × protein source × fibre level interaction (P=0.010, Figure 4.4a), where PC and AF diets were associated with more bird-to-bird pecking than the PF and AC diets until 25 weeks, and an age × fibre level × enrichment interaction (P=0.002, Figure 4.4b), where CN and FE peaked between 22 and 28 weeks of age.
Age was positively associated with an increase in pecks directed at the enrichments (P<0.001), though neither the main effects of protein source nor extra fibre were significant (P>0.731).

Spot pecking increased with age from 16 to 20 weeks of age, and decreased thereafter (P<0.001), but was not affected by the main effects of protein source or extra fibre (P>0.148). However, there was a significant age × fibre level interaction (P<0.001), as F-fed hens performed fewer spot pecks than the C-fed hens after 22 weeks of age (Figure 4.5a). There was an overall enrichment effect (P<0.001) and an age × enrichment interaction (P<0.001) on spot pecks, both which show that N hens spot pecked more than E hens, with the largest difference at 20 and 34 weeks of age (Figure 4.5b).

Age was associated with pecks at the feed (P<0.001), as there appeared to be a cubic function of age (Figure 4.6). Though there were no overall main treatment effects (P>0.193), there was a significant age × protein source × fibre level interaction (P=0.022), as differences between treatments were more apparent toward the end of the study (Figure 4.6) when AF showed greater feed pecks than all other treatments toward the end of the study.
Figure 4.4 Proportion of hens (predicted means±SE back transformed from logit scale) over time performing bird-to-bird pecking by a) protein source × extra fibre and b) extra enrichment × extra fibre during scan sampling. P: plant protein, A: animal protein, C: control fibre, F: extra fibre, N: no extra enrichments, E: extra enrichments.

Figure 4.5 Proportion of hens (predicted means±SE back transformed from logit scale) observed spot pecking over time by a) fibre level and b) extra enrichment during scan sampling. C: control fibre, F: extra fibre, N: no extra enrichments, E: extra enrichments.
Figure 4.6 Proportion of hens (predicted means±SE back transformed from logit scale) observed pecking at feed over time by protein source and fibre level. P: plant protein, A: animal protein, C: control fibre, F: extra fibre).

4.4.4 Feather Cover

Overall, plumage condition remained good for the duration of the study. Only two cages had any obvious feather damage in two different treatments (PCN and ACN). Overall, feather cover was worst on the tail and best on the head and body (body site P<0.001 (SED=0.096): means estimated from GLMM back transformed to proportion scale and estimated means±SE on logit scale, tail 0.50 (-0.01±0.34), neck 0.20 (-1.36±0.34), thighs 0.15 (-1.76±0.33), wings 0.11 (-2.08±0.34), and the rest 0.01 (-5.09±0.35)). Not surprisingly, feather cover deteriorated with age (P<0.001, Figure 4.7). In addition, E hens had better feather cover than N hens (P=0.022 (SED=0.130): means estimated from GLMM back transformed to proportion scale and estimated means±SE on logit scale, N 0.13 (-1.88±0.34), E 0.10 (-2.24±0.34)). However, neither the main effects of protein source nor extra fibre affected feather condition (P>0.535).
Figure 4.7 Mean proportion±SE of scores>0 (predicted means from GLMM back transformed from logit scale) from 16-34 weeks of age.

4.4.5 Enrichment Damage

The only factor that affected the cumulative scores for pecking mats was age (P<0.001), and as expected, the cumulative damage increased over time (Figure 4.8). The cumulative scores for the ropes were similarly affected by age (P<0.001). They were also affected by an age × protein source interaction (P=0.003), as the hens fed the A diet accumulated a higher score over time (Figure 4.8).
Figure 4.8 Mean±SE cumulative rope damage scores (natural logarithm+1 scale) estimated from the LMM by protein source (P, plant based; A, 5% inclusion of animal protein). Average SED between protein sources across ages: 0.043.

4.5 DISCUSSION

Overall, mortality was low, and fell below the breed guideline standards of 1.0% at 34 weeks of age despite the lack of beak treatment for this flock. Higher levels of mortality are generally observed for non-beak treated flocks, due mainly to increased IP and associated damage (reviewed by Hester and Shea-Moore (2003)), however the severity of an IP outbreak depends on a variety of factors. Though IP outbreaks can occur at any age, evidence of IP behaviour is usually evident before or at peak egg production (25-35 weeks of age), as suggested by McKeegan and Savory (1998). Rodenburg and Koene (2003) showed an increase in SFP from 25-34 weeks of age for hens from a high-feather pecking line. In addition, flocks of hens that exceeded an acceptable level of feather damage by the end of lay had already done so by 40 weeks of age (Drake et al., 2010). Therefore, the period of data collection for this experiment was purposefully
set up to capture mortality, behaviour, and feather damage leading up to and during this sensitive period.

The current flock showed minimal feather damage and few bird-to-bird pecks observed. No overall treatment differences for bird-to-bird pecking were detected despite the slight improvement in feather condition for hens with extra enrichment. Greater treatment differences may have been elicited with an older flock or a flock with poor performance, as mortality and feather damage generally increase with age. However, it has been suggested that vent pecking can be triggered by the hormones produced at the onset of lay (Hughes and Duncan, 1972; Sedlačková et al., 2004), and IP increases around peak lay (i.e. 25-35 weeks of age), which is within the age range used for this study. For example, Wechsler and Huber-Eicher (1998) showed an increase in feather pecking to 22-24 weeks of age and differences in performance of feather pecking was observed from 19-30 weeks of age. In addition, McAdie et al. (2005) was able to show improved feather condition in 35-week old hens housed with string enrichment. In the current study, the hens were housed for 20 weeks and data collection spanned the peak lay period, so a lack of IP up to this point may mean that IP was unlikely to develop without a particular trigger (such as sudden changes to their environment).

In contrast to the results presented here, provision of similar extra enrichments to hens in furnished cages on a commercial farm (Chapter 3) did not affect overall feather damage, but it did reduce bird-to-bird pecking. McAdie et al. (2005) showed (in two separate experiments) that rope enrichments were able to reduce feather damage (for a commercial strain) and IP (for a high feather pecking strain). The enrichments provided in the current study were observed to elicit more frequent pecks over time; an increase in attraction to ropes agrees with Jones et al. (2004). Even though the hens fed the animal protein accumulated higher damage scores for the ropes than the
hens fed the plant-based diet, there were no associated differences in IP or feather damage. Despite hens being omnivores, pork would not be a part of their natural diet, and thus the results may have been different if a more biologically relevant animal protein was used (e.g. insect protein). McKeegan et al. (2001) observed a reduction in SFP for pullets fed a diet containing fishmeal, though it was only detected from 13-16 weeks of age, and this also did not affect feather condition, similar to the findings from the current study. In contrast to our results, van Krimpen et al. (2009) found that diets diluted with oat hulls reduced feather damage, with greater improvement as dilution rates increased. We observed the tail to have the most feather damage, as did Glatz (2001) and Kriegseis et al. (2012), possibly because this is the easiest site for peckers to target or most prone to abrasion from the cage.

From both focal and scan behaviour sampling, the proportion of peck types followed similar rankings, with feed and spot pecks observed most frequently. Within bird-to-bird pecks, GFPs were observed more often than aggressive and SFPs. Though McAdie and Keeling (2000) also observed GFPs more frequently than SFPs, the inverse was shown by Lambton et al. (2010) when observing hens within large commercial flocks. In the current experiment, feather damage was minimal and therefore it makes sense that more damaging pecks like SFPs were less frequent than GFPs.

For some behaviours, the results from scan sampling mirrored those from focal sampling, whereas other behaviours differed slightly. For example, spot pecks decreased after 20 weeks for both observation methods. The combination of fibre inclusion and extra enrichments reduce spot pecks for scan and focal sampling. During scan sampling, there were overall effects on spot pecking due to fibre inclusion (where F<C) and due to extra enrichments (where E<N). Although there was an effect of fibre inclusion over time during the focal sampling for feed pecks, the
differences between diet types were small and there was no clear trend one way or the other. Hens fed the AF diet were observed to peck more at the feed than the other treatments during scan sampling toward the end of the study which may have been to maintain peak egg production as the crude protein and fat content of this diet was generally lower than the others. For bird-to-bird pecks, both focal and scan sampling revealed a three-way interaction with age, protein source, and fibre inclusion, however, the direction of the results were slightly different depending on sampling method. During focal sampling, the AC treatment performed less bird-to-bird pecking than the other treatments near 26-30 weeks of age. However, scan sampling indicated that AC and PF treatments performed less bird-to-bird pecking from approximately 20-25 weeks of age, whereas, the PC and AF treatments performed less from approximately 30-35 weeks of age.

Data from focal and scan sampling suggests that the ropes were more attractive than the mats. When presented with a rope, cane, or ring enrichment device, Jones and Carmichael (1999) found that chicks pecked most at the white rope, although the chicks most likely were not able to manipulate the cane or ring devices as easily as the ropes. The mats used in the current experiment were easy to manipulate and thus should have offered the hens some positive feedback. It may be possible to increase pecks at both the ropes and mats by changing their presentation position, as the vertical presentation (as opposed to horizontal, or on the floor) may have discouraged their use, as a natural foraging position would be to peck items on the floor. However, the fixed action patterns of pecks delivered to “chicken-shaped” or flat objects did not differ (Dixon et al., 2008). In that case, the motivation to peck a horizontal vs. vertical object may not differ either, though the ability to scratch at an object (if horizontal) may increase its viability as a foraging substrate. In another study, Moroki and Tanaka (2016) showed that singly-housed hens were interested in an enrichment (wooden board with stones) that was fixed horizontally to the floor, and actually spent
more time pecking at the enrichment than they did to the cage floor or other cage parts. However, interest in the enrichment was less marked for hens that were pair-housed.

Though starting location was balanced across observations for each cage, it was detected as a significant factor for all behaviours during focal sampling. To some extent, these results are not surprising. For example, feed pecks occurred less often when observations began near the rope; probably because this was farther from food trough compared to the nest or mat. Spot pecking was most frequent near the nest as the flaps were often the target for spot pecks. Enrichment pecks were least frequent near the nest simply because the other starting locations were in close proximity to the enrichments themselves. However, some of the other differences are less easily explained. For example, bird-to-bird and beak pecks were observed most often when the observations began near the mat and least often near the nest.

Though reductions in IP, feather damage, and mortality are usually the goals for improving animal welfare, the reduction in other potentially stereotypic behaviour may be just as important. The performance of excessive spot pecking may indicate a lack of suitable foraging opportunities. Therefore, a reduction in overall spot pecking could reflect a higher degree of satisfactory foraging opportunity permitted by the extra enrichments. However, it appeared that the extra fibre and extra enrichment (FE) treatment was most successful at reducing spot pecking (as observed during focal sampling). Some three-way interactions were reported for bird-to-bird pecking, though the differences are harder to decipher. For focal sampling, the AC treatment showed less bird-to-bird pecking than the other treatments at approximately 26-30 weeks of age.

It is conceivable that the hens fed the extra fibre may not have had the same motivation to peck at the enrichments as the control fibre hens. If both the extra fibre and extra enrichments are
expected to act as a type of substitute for foraging behaviour, in that they would both alter the hens’ time budgets, then it could be argued that extra fibre would reduce enrichment pecks. However, we did not detect a difference in frequency of pecks at the enrichments between these two diet types.

Feed pecks were observed most frequently with the AF diet during scan sampling. This follows with the feed disappearance results from Chapter 5, where the F diet resulted in greater feed disappearance rates than the C diet. Though feed wastage was definitely an issue, higher feed intake was expected for the F diet as it was energetically diluted, so increased feed pecks probably reflects increased feed intake for the AF hens.

4.6 CONCLUSIONS

In conclusion, providing extra enrichments to hens in furnished cages resulted in improved feather condition, although the biological difference was not large and this difference was not reflected in observed feather pecking behaviour. Overall feather condition was quite good, yet differences due to enrichments were still detected. Therefore, the potential to improve feather condition in flocks where feather pecking is a problem may be greater than that observed here.

There was no consistent effect of of the main factors of protein source, added fibre or extra enrichments or any treatment interactions on bird-to-bird pecking. However, spot pecking was reduced by the extra fibre diet and the extra enrichment treatment, suggesting that these treatments occupy the hens’ time and attention, though these differences may have been driven mainly by the high fibre(extra enrichment (FE) combination. Further work is needed to determine if this
treatment combination would be more effective for flocks with a greater disposition for feather pecking and if improvements could be maintained throughout the entire laying period.
Chapter 5 Effect of dietary protein source, fibre inclusion, and extra environmental enrichment on production parameters of non-beak treated laying hens

A version of this chapter will be submitted for publication.
5.1 ABSTRACT

Some methods used to reduce injurious pecking in flocks of laying hens with intact beaks include dietary alterations that may impact on production parameters. This chapter includes production data collected on the same flock of hens as in Chapter 4, where behaviour and welfare data are reported. The goal was to ensure that the treatments used to reduce injurious pecking did not negatively impact production measures that are economically important. Hens were housed in furnished cages from 16 to 34 weeks of age and data were collected throughout this period. Treatments were applied as a 2×2×2 factorial, with protein source (P: plant-based, A: 5% pork meat-and-bone meal), fibre inclusion (C: control level, F: 10% dilution with oat hulls), and extra enrichment (N: control, E: extra enrichments including ropes, pecking mats, and blunting boards) as the main factors. The F diet resulted in greater feed disappearance (feed intake plus feed wastage) than the C diet (P=0.030, indicating that the F hens either ate or wasted more feed. Though there were no treatment differences for hen-day egg production, the hens fed the P diet laid heavier eggs (P<0.001) as did hens fed the C diet, but only at the onset of lay (P=0.013). Various aspects of external eggshell quality were affected by age and treatment, with the main result being that N hens laid more eggs with ‘good quality’ eggshells than E hens (P=0.005). Excreta dry matter content was higher for P than for A hens, and was also higher for F than for C hens after the first sampling (age × protein P=0.002, age × fibre P<0.001). Overall, differences in production efficiency were mainly due to feed disappearance rates (where F>C) and some eggshell quality factors (where treatment differences varied) but not in overall egg production. There were some apparent welfare benefits with the F and E treatments (Chapter 4), though there were some negative production traits associated with these treatments as well. Therefore, a balance between welfare and production should be optimised.
5.2 INTRODUCTION

In conjunction with the data collected in Chapter 4, production data were also obtained in order to assess whether the applied treatments had an effect on parameters of importance to production efficiency. The main goal of these dietary alterations was to reduce injurious pecking, which in turn reduces morbidity and mortality within a flock, thereby reducing the associated egg loss. Despite these anticipated benefits to productivity, some dietary alterations (e.g. energetic dilution by increased fibre content) may act in other ways to reduce production efficiency (e.g. increased feed intake) or external eggshell quality (e.g. dirty eggs via reduced excreta dry matter).

Most of the diets used to improve behavioural outcomes for laying hens include altering protein level or source or increasing the fibre content. Previous research on laying hens fed alternative diets has shown some differences in feed intake, but few differences for other production parameters. For example, Hartini et al. (2002), Steenfeldt et al. (2007) and van Krimpen et al. (2009) all found that high fibre diets resulted in significantly higher feed consumption than low fibre diets. Though Hartini et al. (2002) and Steenfeldt et al. (2007) also found a reduction in mortality for hens fed high fibre diets, the hen-day egg production was not affected in comparison to the control diets. Similarly, van Krimpen et al. (2009) found no difference in egg mass or hen body weight between diets. McKeegan et al. (2001) found no differences in early egg production or egg weight between groups of hens reared on and fed a purely plant based diet or a diet containing fish meal from 0 to 24 weeks of age, however, the fish meal-fed hens were heavier from 4 weeks onward. In another study where hens were fed a standard plant-based diet, a diet containing meat and bone meal, or a diet containing just meat meal from 20 to 40 weeks of age, the proportion of wet litter was lower for the groups fed the meat and bone meal in comparison to the control and meat meal-fed hens (van Krimpen et al., 2011).
For this experiment, two dietary alterations plus the addition of extra enrichments were assessed. The dietary alterations included two levels of fibre inclusion: control and high (diet diluted by the addition of 100 g kg\(^{-1}\) oat hulls), and two protein sources: plant based (control) and processed animal protein (50 g kg\(^{-1}\) pork meat and bone meal). The hypothesis was that the dietary alterations would not affect body weight, egg production, or external eggshell quality. However, the hens fed extra fibre were expected to have higher feed intakes and lower excreta dry matter content. In addition, the extra enrichments were not expected to affect any of the production parameters. Behavioural and welfare outcomes were also measured and these data are presented in the following chapter.

5.3 MATERIALS AND METHODS

5.3.1 General Husbandry

Data presented in this chapter were collected during the same experiment presented in Chapter 5, and thus the general methodology and animal husbandry is the same.

5.3.2 Data Collection

5.3.2.1 Body Weight

Pullets were individually weighed and tagged upon arrival at the research facility (15 weeks). Hens were weighed again at the end of the study (34 weeks).

5.3.2.2 Feed Disappearance

Bags of 25 kg feed were weighed and allocated to individual cages. New weighed bags of feed were added as needed. Every four weeks (starting at 19 weeks of age), the remaining feed was weighed, which included both the weight of feed remaining in the cage’s respective feed bag
(minus the weight of the bag), plus the weight of the feed remaining in the feed trough. To calculate feed disappearance (rather than feed intake as there was evidence of feed wastage on the manure belt) over each four week period, the total weight of feed added minus the weight of the remaining feed was used and divided by the number of hen days per cage to give mean g per hen.

5.3.2.3 Egg Production and Eggshell Quality

From point of lay until the end of the study, all eggs were counted daily between 11:00 – 13:00 and recorded for each cage. Every two weeks, starting at 19 weeks of age, all of the day’s eggs were collected for an external eggshell quality check. All of the eggs from each cage were group weighed for mean egg weight. In addition, each egg was examined individually and shells were scored for a variety of defects (Table 5.1), which were not mutually exclusive (except for ‘good’). Eggshell quality data were collected in numerical order based on cage number (i.e. cage 1 through to cage 64), but the observer was blind to the treatment of each cage during data collection.
Table 5.1 Categories of eggshell defects (adapted from Wolc et al. (2012) and Alltech (2016)).

<table>
<thead>
<tr>
<th>Defect</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cracked</td>
<td>Any break in the shell surface, either very obvious (due to damage by toes or beaks) or subtle (hairline fractures)</td>
</tr>
<tr>
<td>Dirty with blood</td>
<td>Evidence of blood on shell surface</td>
</tr>
<tr>
<td>Dirty with faeces</td>
<td>Evidence of faeces on shell surface</td>
</tr>
<tr>
<td>Extra-cuticular calcium</td>
<td>White dusting or raised white bumps on the shell’s surface</td>
</tr>
<tr>
<td>Softshelled</td>
<td>Shells that are very easy to crack, thin shells, or eggs that are missing shells completely</td>
</tr>
<tr>
<td>Double yolked</td>
<td>Very large eggs with two yolks</td>
</tr>
<tr>
<td>Brown spots</td>
<td>Eggs with brown flecks or speckles</td>
</tr>
<tr>
<td>Other</td>
<td>Any other defect not listed here, i.e. misshapen, very pale</td>
</tr>
<tr>
<td>Good</td>
<td>Eggs not displaying any of the defects listed here</td>
</tr>
</tbody>
</table>

5.3.2.4 Excreta Dry Matter

From 21 weeks of age, and every four weeks onward, a ~300g sample of excreta was collected from the manure belt beneath each cage after a 24 h collection period onto clean manure belts. Each of the samples was weighed and then transferred into a large drying oven (80°C) for a period of at least 48 h. After the drying period, each of the samples was re-weighed for a dry weight. Dry matter content was calculated as a percentage of the fresh weight.

5.3.3 Statistical Analysis

All data were analysed in Genstat (16\textsuperscript{th} edition) with $\alpha=0.05$. For sparse data sets, age was fitted as a covariate (with the linear, quadratic, as well as cubic functions of time tested) within the model, but presented as the best fit for the data in the results section.
5.3.3.1 Body Weight

A linear mixed model was fitted to the body weight data (following natural logarithm transformation) at 16 and 34 weeks of age with fixed effects including age, protein source, extra fibre, and extra enrichments (Table 5.2). The random model included cage and hen within cage.

Table 5.2 Statistical models fitted to each of the datasets.

<table>
<thead>
<tr>
<th>Dataset</th>
<th>Test</th>
<th>Transformation</th>
<th>Fixed Model</th>
<th>Random Model</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body Weight</td>
<td>LMM</td>
<td>Natural logarithm</td>
<td>Age<em>protein</em>fibre*enrich</td>
<td>Cage/hen</td>
</tr>
<tr>
<td>Feed Disapp.</td>
<td>LMM</td>
<td>N/A</td>
<td>Age<em>protein</em>fibre* enrich</td>
<td>Cage/age</td>
</tr>
<tr>
<td>Egg Production</td>
<td>LMM</td>
<td>N/A</td>
<td>Age<em>protein</em>fibre*enrich</td>
<td>Cage/week/day</td>
</tr>
<tr>
<td>Egg Quality</td>
<td>GLMM</td>
<td>Binomial, log link</td>
<td>Age<em>protein</em>fibre*enrich</td>
<td>Cage/age</td>
</tr>
<tr>
<td>Egg Weight</td>
<td>LMM</td>
<td>Natural logarithm</td>
<td>Age<em>protein</em>fibre*enrich</td>
<td>Cage/week+#egg</td>
</tr>
<tr>
<td>Excreta DM</td>
<td>LMM</td>
<td>N/A</td>
<td>Age<em>protein</em>fibre*enrich</td>
<td>Cage/age</td>
</tr>
</tbody>
</table>

5.3.3.2 Feed Disappearance

Feed disappearance was measured on a cage level for net feed weight between observations. To calculate feed disappearance per hen per day, the number of hen-days per cage (accounting for cage-level mortality) was used to get a mean value on an individual hen basis. Residuals for these data approximated a normal distribution, and therefore data were not transformed. An LMM was fitted with age, protein source, fibre inclusion, and enrichment in the fixed effects model (Table 5.2). The random effects model included age within cage.
5.3.3.3 Egg Production and Eggshell Quality

Egg production data were analysed on a weekly basis for each cage. The data were calculated as a proportion of egg production on a hen-day basis so that mortality within each cage was taken into account. An LMM was used to analyse the fixed effects of age, protein source, extra fibre, enrichment, and their interactions. The residuals of the raw data approximated a normal distribution, so no transformation was necessary. The random model included cage, week, and individual day within week.

For egg quality, data were treated as binomial for each egg at each characteristic (i.e. present or not) and were analysed using a GLMM. For rare defects (other, cracked, bloody, soft shell, double yolk), the prevalence was combined into one category. Age (covariate), protein source, extra fibre, and extra enrichment were tested in a two-way interacted model. The random model included age nested within cage. Mean egg weights were transformed on the natural logarithm scale and analysed in an LMM. Age, protein source, fibre level, enrichment, and their interactions were included in the fixed effects model. The random model accounted for cage, age within cage, and the total number of eggs laid/cage per day.

5.3.3.4 Excreta Dry Matter

Raw data (proportion of dry matter) were analysed as the residual plots resembled normal distribution curves. An LMM was fitted to the data, with age, protein source, fibre level, and enrichment as the fixed effects. The random model included age within cage.
5.4 RESULTS

5.4.1 Body Weight

Not surprisingly, there was an effect of age on body weight, as the hens weighed significantly more than the pullets (P<0.001 (SED=0.002): means estimated from LMM back transformed to grams and estimated means±SE on transformed scale: 15 weeks 1269.02 g (7.15±0.008) and 34 weeks 1976.34 g (7.59±0.008)). There were no significant main treatment effects (P>0.608). However, there was a significant age × protein source × fibre level × enrichment interaction (P=0.006) as the increase in body weight differed slightly between the treatments (Table 5.3). All of the treatment means exceeded the breed standard for 34 weeks of age.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>15 weeks</th>
<th>34 weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCN</td>
<td>1269.0 (7.146)</td>
<td>2002.2 (7.602)</td>
</tr>
<tr>
<td>PCE</td>
<td>1266.5 (7.144)</td>
<td>1960.6 (7.581)</td>
</tr>
<tr>
<td>PFE</td>
<td>1270.0 (7.147)</td>
<td>1966.0 (7.584)</td>
</tr>
<tr>
<td>PCE</td>
<td>1276.7 (7.152)</td>
<td>2002.2 (7.602)</td>
</tr>
<tr>
<td>ACN</td>
<td>1271.6 (7.148)</td>
<td>2020.3 (7.611)</td>
</tr>
<tr>
<td>ACE</td>
<td>1266.5 (7.144)</td>
<td>1992.2 (7.597)</td>
</tr>
<tr>
<td>AFN</td>
<td>1261.4 (7.140)</td>
<td>1945.0 (7.573)</td>
</tr>
<tr>
<td>AFE</td>
<td>1271.6 (7.148)</td>
<td>1929.5 (7.565)</td>
</tr>
<tr>
<td>Breed Standard</td>
<td>1260</td>
<td>1910</td>
</tr>
</tbody>
</table>

Table 5.3 Mean body weights estimated from LMM back transformed to grams and estimated means on transformed scale in parentheses. Average SED= 0.029, average SE=0.022. Breed standards (in grams) are presented as well for reference purposes only. (P: plant protein, A: animal protein, C: control level of fibre, F: extra fibre, N: no extra enrichment, E: extra enrichment).
5.4.2 Feed Disappearance

In general, feed disappearance was much higher than expected based on feed intake data from Hy-Line breed guidelines after 19 weeks of age (Figure 5.1a). However, as feed disappearance is feed intake plus feed wastage, greater values were expected. There was a significant effect of age (P<0.001) as feed disappearance increased with age. There was also an overall effect of fibre level (P=0.030) as the diets with extra fibre resulted in higher rates of feed disappearance (Figure 5.1b), but no differences were detected due to protein source or extra enrichment (P>0.302).

![Figure 5.1 Feed disappearance (grams) a) by age (P<0.001) against breed standard (dotted line) and b) by fibre inclusion (P=0.030). Means±SE predicted from LMMs.](image)

5.4.3 Mortality, Hen-Day Egg Production, and Eggshell Quality

In total, 9 hens were found dead or culled over the entire experimental period and thus data were too sparse to analyse statistically, but there did not appear to be any obvious treatment effects (see Chapter 4 for more details). By 20 weeks of age at least one hen in each cage had laid an egg. Hen-day egg production was only affected by age (P<0.001), as egg production increased from 17
to 21 weeks of age and then plateaued until the end of the study (Figure 5.2). Hen-day egg production increased more quickly and peaked higher than the breed standard for all treatments.

**Figure 5.2** Egg production (hen-day) for each treatment (raw means). Graph includes the range for the Hy-Line Brown breed standards (shaded area). A: animal protein, P: plant-based protein, C: control fibre level, F: extra fibre, N: no extra enrichment, E: extra enrichment.

The largest proportion of eggs were categorised as good eggs (Figure 5.3). The most prevalent egg defect was brown spots, followed by extra calcium deposits, dirty, other, and cracked. Bloody, soft shelled, and double yolked eggs were the least prevalent.
The proportion of good eggs increased over time \((P<0.001, \text{Figure 5.4})\), and N hens laid a greater proportion of good eggs than E hens \((P=0.005, \text{SED}=0.0766\): means (proportions) estimated from GLMM back transformed and estimated means±SE on transformed scale: N 0.57 (0.29±0.05), E 0.53 (0.11±0.05)). There was a significant age × fibre interaction \((P=0.039)\), as C-fed hens had fewer good eggs at 23 weeks, but more at 31 weeks of age (Figure 5.5a). In addition, there was a protein source × fibre interaction as F increased the proportion of good eggs for hens fed the P diet, but decreased good eggs for those fed the A diet \((P=0.019, \text{SED}=0.1084\): means (proportions) estimated from GLMM back transformed and estimated means±SE on transformed scale: PC 0.52 (0.08±0.08), PF 0.58 (0.33±0.08), AC 0.57 (0.26±0.08), AF 0.53 (0.13±0.08)). There was also a three-way interaction between age, protein source and fibre level \((P=0.009, \text{Figure 5.5b})\).
Figure 5.4 Proportion of eggs of different characteristics (good, brown spots, extra cuticular calcium (ECC), dirty with faeces + blood spots, cracked, and other (soft shelled, double yolks, or other)) over time. Means±SE back transformed from predicted GLMM estimates.

Figure 5.5 Proportion of "good" eggs over time by a) fibre level and b) protein source × fibre level. Means±SE back transformed from predicted GLMM estimates. (Plant based protein (P), meat and bone meal inclusion (A), control level of fibre (C), and high fibre diet (F)).

The proportion of eggs with extra cuticular calcium (ECC) was affected by age (P=0.005) as the prevalence decreased with age (Figure 5.4). The F diet resulted in fewer ECC eggs from 21-
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27 weeks (age × fibre P=0.004, Figure 5.6a), though this difference was mainly driven by the AF diet as there was also an age × protein source × fibre interaction (P=0.028, Figure 5.6b).

**Figure 5.6** Proportion of eggs with extra cuticular calcium (ECC) over time by a) fibre level and b) protein source × fibre level. Means±SE back transformed from predicted GLMM estimates. (Plant based protein (P), meat and bone meal inclusion (A), control level fibre (C), high fibre (F)).

The presence of brown spots increased with age (P<0.001, Figure 5.4). Extra enrichment was also a significant factor (P=0.013), as N hens had fewer eggs with this defect (SED=0.099: means (proportions) estimated from GLMM back transformed and estimated means±SE on transformed scale: N 0.23 (-1.23±0.07), E 0.26 (-1.05±0.07)). There was also an age × protein source interaction (P=0.023, Figure 5.7a) as well as an age × fibre interaction (P=0.014, Figure 5.7b). In addition, there was a protein source × fibre interaction (P=0.002) as the F diets decreased this defect in the P diets, but did not in the A diets (SED=0.1400: means (proportions) estimated from GLMM back transformed and estimated means±SE on transformed scale: PC 0.25 (-1.12±0.10), PF 0.20 (-1.36±0.10), AC 0.25 (-1.10±0.10), AF 0.27 (-0.97±0.10)).
Figure 5.7 Proportion of eggs with brown spots over time by a) protein source and b) fibre level. Means±SE back transformed from predicted GLMM estimates. (Plant based protein (P), meat and bone meal inclusion (A), control level of fibre (C), high fibre diet (F)).

The proportion of dirty and/or bloody eggs decreased with age (P<0.001, Figure 5.4) and fibre level (C<F; P<0.001; SED=0.121 means (proportions) estimated from GLMM back transformed and estimated means±SE on transformed scale: C 0.03 (-3.57±0.09), F 0.04 (-3.26±0.08)). Age × extra enrichment affected the prevalence of dirty and bloody eggs (P=0.026), as E hens had significantly fewer dirty eggs from 31 weeks of age (Figure 5.8a). There was also an age × fibre level × enrichment interaction (P=0.042, Figure 5.8b).
Figure 5.8 Proportion of dirty and/or bloody eggs over time by a) enrichment inclusion and b) protein source × enrichment inclusion. Means±SE back transformed from predicted GLMM estimates. (Control fibre (C), high fibre diet (F), no extra enrichment (N), extra enrichment (E)).

Age affected the proportion of observed cracked eggs (P<0.001), as they were the highest closer to the onset of lay (Figure 5.4). Finally, the combined egg quality category (other, soft shell, and double yolk) was affected by age (P<0.001) with a cuboidal time effect (Figure 5.4). There were also two three-way interactions (age × protein × fibre P=0.044, Figure 5.9a, and age × fibre × extra enrichment P=0.039, Figure 5.9b).
Mean egg weight significantly increased with age (P<0.001, Figure 5.10). Protein source was also a significant factor as P-fed hens laid heavier eggs on average than the A-fed hens (P<0.001 (SED=0.0048): means estimated from LMM back transformed to grams and estimated means±SE on transformed scale: P 60.10g (4.08±0.003) and A 59.09g (4.10±0.003)). Age interacted with fibre level (P=0.013), as the C-fed hens laid heavier eggs closer to the onset of lay (Figure 5.10). In addition, there was a non-significant trend for protein × fibre level (P=0.063), as the AF diet tended to have lighter eggs than the other diet types (SED=0.0068: means estimated from LMM back transformed to grams and estimated means±SE on transformed scale: PC 59.92g (4.09±0.005), PF 60.22g (4.10±0.005), AC 59.50g (4.09±0.005), and AF 58.73g (4.07±0.005)).
Figure 5.10 Mean egg weight at different ages (±SE) estimated from LMM back-transformed to grams between the control diet (C) and the extra fibre diet (F). (SED=0.0074 on transformed scale.)

5.4.4 Excreta Dry Matter

Age affected DM content (P<0.001), as it was lowest at 25 weeks of age. The A and F diets resulted in increased DM content when compared to the P and C diets, respectively (both P<0.001). There were also significant interactions between age and protein (P=0.002, Figure 5.11a) as well as age and fibre (P<0.001, Figure 5.11b) as both the A and F diets resulted in an increase in DM, with the differences increasing over time.
5.5 DISCUSSION

Overall, hen performance was similar to what was expected based on breed standards, with the exception of heavier body weights and greater feed disappearance. Although all of the treatment means for body weight exceeded the breed standard at 34 weeks of age, there was still a significant four-way interaction between age and all three treatment factors. It is difficult to interpret this interaction. Again, all treatment means exceeded breed standards for feed intake, however what was actually measured during this experiment was feed disappearance as there was evidence of feed wastage when cleaning the manure belts. Regardless, the high fibre diets resulted in greater rates of feed disappearance than the control diet, which was expected. In speculation, the observed differences could be due to greater feed intake due to energetic dilution of the diet, meaning that the hens would have to consume a larger volume of feed to meet energy requirements (Hetland and Svihus, 2001; Kjaer and Bessei, 2013). However, it is also possible that the hens wasted more of the high fibre diet as the hens may have sorted through the feed more vigorously. Similar to previous research using alternative diets, there were no differences in overall egg

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**Figure 5.11** Means predicted from LMM (± SE) for proportion of excreta dry matter (DM) by age in weeks and protein source (a) or fibre inclusion (b). P: plant based protein, A: animal based protein, C: control fibre level, F: extra fibre inclusion.
production between treatments (Kalmendal et al., 2013), which is an important financial aspect when considering changes to hen diets. However, there were some differences in egg weights, which also plays an important financial role for egg producers. Eggs from plant-fed hens were heavier than those from the hens fed the diet containing meat and bone meal. This may not have been a direct impact of protein source, but rather, at least in part, due to a difference in dietary fat content (as measured by Oil B in Table 4.6). Grobas et al. (1999) showed that supplementing the diet with fat increased mean egg weight by more than a gram from 22 to 65 weeks of age. In the analysis of the diets in the current study, the plant-based diet contained slightly higher levels of fat (Oil B) than the animal protein-based diet, therefore, this may have affected egg weight. There was also an effect of an age by fibre interaction on egg weight, with the control diet having heavier eggs than the fibre diet at the beginning of the laying period. This observed difference was probably driven in part by the AF diet in particular as the hens fed this diet tended to have lighter eggs than the hens fed the other diets. Though the AF diet did contain less fat than the PC and PF diets, it contained a similar level to the AC diet, therefore suggesting that dietary fat content is not the only factor affecting egg weight in this experiment. According to Summers and Leeson (1983), body weight is the main driver of early egg weight and despite a lack of differences in body weight at the start of the current study, the hens were weighed prior to being fed the treatment diets, and thus could have differed in initial weight gain once the treatments began.

Some eggshell deformities, in particular misshapen eggs or those with extra cuticular calcium, are often associated with delayed oviposition as a result of acute stress (Reynard and Savory, 1999). The proportion of good eggs was higher for hens without extra enrichment, which was mostly due to a difference in brown spots. According to Alltech (2016), brown spots are a result of excessive calcium in the diet or a disturbance to the hens during shell formation, however
the enrichments did not contain calcium. Perhaps the assessment and replacement of the enrichments could have disturbed the hens enough to result in brown spots, although the inspections occurred only once every two weeks and based on anecdotal observations, the hens did not appear to be bothered by the procedure. However, even acute stressors can have relatively prolonged effects on egg shell quality, as Hughes et al. (1986) showed that injections of adrenaline caused an increase in proportion of eggshell defects for 10 days. Brown spots were also present more often for A-fed hens toward the beginning of the trial, which may in fact have been related to calcium levels as the average calcium level between the AC and AF diets were higher than that for the PC and PF diets (4.07% vs. 3.67%, see Table 4.6). Age, protein source and fibre inclusion also affected the proportion of good eggs. The differences were most apparent towards the end of the study, which may be a reflection of adaptation to the different diets. The PF diet had the highest proportion of good eggs and the lowest proportion of eggs with brown spots.

Reynard and Savory (1999) demonstrated that heavier calcium deposits were shown to be associated with a longer delay in oviposition (i.e. more stressed). The high fibre diets, in particular the AF diet, resulted in fewer eggs with extra cuticular calcium which may indicate that the alternative diet resulted in a reduction in stress (i.e. by having better gustatory feedback or being associated with reduced spot pecking; see Chapter 4). However Reynard and Savory (1999) suggested that lighter calcium dusting was not correlated with length of delay or lack thereof in oviposition (i.e. no symptoms of stress). In the current study, even light dustings were classified as calcium deposits and therefore may have inflated the proportion of eggs with extra cuticular calcium that would actually correspond to stress levels. In addition, the stress imposed on the hens in the former study was an acute disruption of daily activity and thus may not reflect the type of stress a diet type would have.
Though there were treatment differences in the proportion of dirty/bloody eggs, the values observed during this experiment were within the range observed by Appleby et al. (2002) of 1.3%-4.5% in conventional or furnished cages. Control fed hens had fewer dirty eggs than those fed the high fibre diet, which does not correspond to the excreta dry matter content, as the high fibre diets resulted in a higher dry matter content. In a review, Roberts (2004) indicated that wet or sticky droppings could negatively affect eggshell quality. However, Choc (1997) suggested that insoluble fibre is important in maintaining appropriate consistency of poultry excreta and both Amerah et al. (2009) and van der Hoeven-Hangoor et al. (2014) found increased excreta dry matter content for broilers fed diets containing coarse particles of non-starch polysaccharides or oat hulls. However, the high-fibre hens had higher feed disappearance (and higher feed intake, assumedly) and thus may have had a greater volume of excreta and a greater chance to contaminate their eggshells. Our excreta dry matter values were similar to those calculated in Ribeiro et al. (2014), which ranged from 27.5-29.2%. Though excreta dry matter content is less relevant to welfare outcomes for caged hens, it may be of greater importance for barn housed or free range hens (e.g. drier litter results in reduced footpad lesions, see Wang and Ekstrand (1998) and Mayne et al. (2007)). Higher moisture content of the excreta may also increase volatile ammonia levels (Maliselo and Mwaanga, 2016) and therefore may in fact directly affect bird welfare. In the current study, the diet containing the pork meat-and-bone meal increased dry matter content of the excreta when compared to the hens fed the plant-based diet. Vieira and Lima (2005) showed increased digestibility and excreta dry matter content for broiler diets containing 70 g kg\(^{-1}\) of animal by-products when compared to an all vegetarian diet. Similar results were found by Hossain et al. (2013), as the inclusion of fish meal to broiler diets resulted in increased excreta dry matter content.
In addition, the hens without extra enrichment had more dirty eggs than the hens with extra enrichment near the end of the study. Though we did not measure activity level directly, it is possible that the hens with extra enrichments were more active and thus had less manure build up on the cage floor, thereby reducing the chance for eggs to be laid in dirty areas.

5.6 CONCLUSIONS

Overall, the differences were not very large between the treatments, and they were not always in the same direction. In terms of production efficiency, the high fibre diet resulted in increased feed disappearance, no difference in egg production, more dirty eggs and lighter first laid eggs, but greater excreta dry matter content. Though the bulkier, high fibre diets may be more inexpensive than a standard diet, it would take up more storage space and require larger volumes of feed to be manufactured and shipped. These reasons would all serve to make egg production more costly. The animal protein-based diet did not affect feed disappearance or egg production, or egg weight, but resulted in greater excreta dry matter than plant-based diets. Finally, the extra enrichments also did not affect feed disappearance or egg production, but had some effects on external eggshell quality. For example, hens without extra enrichment had more good quality eggshells, fewer eggs with brown spots, but more dirty eggs towards the end of the study. However, both the high fibre diets and the extra enrichments improved some indices of good welfare (i.e. feather condition and spot pecking, see Chapter 4) and thus some of these benefits will have to be weighed against the differences in some of the parameters. In addition, future work with these dietary alterations should be explored further and flock performance should be observed throughout the entire lay period.
Chapter 6 Quantifying the effects of age, breed, cuttlebones, and an abrasive beak blunting device on beak shape and pecking force

A version of this chapter will be submitted for publication.
6.1 ABSTRACT

With potential bans on all types of beak alterations being considered throughout Europe, it is becoming imperative to find alternative methods to reduce the damage caused by injurious pecking. One approach is to provide an abrasive surface at which the hens will peck that results in a reduction of the length and sharpness of the beak. To determine the efficacy of two different types of abrasive materials in two different locations on beak morphology and peck force, 36 White Leghorn hens and 36 Columbian Rock hens with intact beaks were housed in individual cages for 10 weeks. Twenty-four hens of each breed were given access to abrasive pecking devices in two locations within the cage; 12 were given cuttlebones and 12 were given beak blunting boards. The remaining 12 hens of each breed were housed without any pecking devices, acting as controls. Data were collected prior to device installation (29 weeks of age), halfway through the experiment (35 weeks), and at the end (40 weeks). At each of the data collection periods, all hens were photographed from the side so as to digitally measure top mandible length and beak tip angle, and all hens were subjected to a peck force test in the same week. Data were analysed using Linear Mixed Models in Genstat (16th Edition). Age affected both top mandible length (P<0.001) as well as beak tip angle (P<0.001), as beaks were longest at 35 weeks and had the smallest beak tip angle (i.e. sharper) at 40 weeks of age. Columbian Rocks had longer top mandibles (P=0.011) and smaller beak tip angles (P<0.001) and there was a tendency for increased cuttlebone use to blunt beak tip angle (P=0.070) but not beak length (P=0.632). There was little indication that the blunting boards were used, though the cuttlebones showed promising results. Peck forces in all directions were strongest at 29 weeks of age (P<0.001), but were not affected by device type or location. Further studies are needed in large scale commercial settings to assess the efficacy of cuttlebone access on not only beak morphology but also injurious pecking behaviour and feather cover.
6.2 INTRODUCTION

Historically, beak alteration has been used to reduce the damage caused by bird-to-bird pecking, including feather pecking and cannibalism, behaviours which are grouped under an umbrella term known as injurious pecking (IP). Currently, the main method used in North America, and the only permitted method in the United Kingdom (UK), is infra-red beak treatment. However, some countries (e.g. Finland, Norway, Switzerland) have successfully stopped any type of beak alteration. Likewise, this practice has been under recent public scrutiny in the UK and the Department for the Environment, Food and Rural Affairs (Defra) had proposed a review of the practice with a view to ban any type of beak alteration in England in 2011. Upon advice from the Farm Animal Welfare Council (FAWC), the ban was subsequently deferred until January 2016, however, after a review of evidence by the Beak Trim Action Group, this ban was cancelled as the risk of increased morbidity and mortality associated with IP in intact-beak flocks was too great (Defra, 2015). However, with the practice of beak alteration still of concern to the public and governments, another potential ban may arise in the future, making finding alternative solutions to reducing IP still very relevant. Moreover, IP is still a problem even for beak altered flocks and thus, regardless of potential bans, reducing the damage caused by IP would still greatly improve hen welfare around the globe.

Some effort has been made to use abrasive surfaces within the housing facilities that act to blunt the beak tip when voluntarily pecked (Fiks-Van Niekerk and Elson, 2005; Van der Weerd, 2006). When a beak blunting material was pasted onto the bottom of feed troughs of non-beak trimmed pullets from 6 weeks of age onward, pullets with access to the material had beaks that were significantly shorter than pullets without the blunting material when their beaks were measured at 45, 49, 57, and 61 weeks of age (Lumb, 2006; Van der Weerd, 2006). However, the
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differences were quite small, ranging from 0.67–0.97 mm, where differences between intact (without blunting material) and conventionally hot-blade trimmed beaks ranged from 7.82–8.20 mm. In our previous study (Chapter 3) we used an abrasive paste (S N Supplies, Lincoln, UK) painted onto Perspex© boards that were mounted on the inside of the cage fronts from 16–71 weeks of age. Beaks of hens housed with the devices were not any shorter than those without the devices. However, the devices were placed such that the hens had to peck at them intentionally rather than being in the feed trough. It was suggested that the devices were not attractive or interactive enough to initiate or sustain interest, and so did not blunt the beaks. In Chapter 4, a similar device was installed in the feed trough (meaning the hens were more likely to come into contact with the device) for hens from 16–35 weeks of age, but beak morphology was not measured.

For pet birds, cuttlebones (i.e. the internal shell of the cuttlefish, *Sepia officinalis*) are generally used as calcium supplements as well as beak blunting devices to maintain good beak health (Arnall, 1965; O’Brien and Villm, 1988). Cuttlebones are readily used and offer an interactive surface as it wears down as birds peck at them. In addition, cuttlebones are mostly comprised of calcium and other minerals and thus can provide some positive nutritional feedback for both pet birds as well as egg laying hens, which may sustain pecking interest directed toward them. For example, Taylor (1996) reported greater use of cuttlebones when canaries were denied access to other soluble grit sources (i.e. oyster shells).

In a recent study, Dennis and Cheng (2010) showed that hot blade beak-trimmed chicks used less force to peck at feed when measured by a force transducer at 3 weeks of age. This result was most likely due to pain, as hot blade beak trimming has been reported to cause both acute and chronic pain (Breward and Gentle, 1985; Hester and Shea-Moore, 2003). However, it is possible
that the reduction in pecking force is caused by an adaptation of pecking style due to morphological changes in beak shape (i.e. shorter, blunter beak tip). Prescott and Bonser (2004) showed that beak trimmed hens were less successful at picking up feed particles than their intact-beak flockmates. In addition, wild birds with beak deformities have been observed to alter their pecking style (i.e. turning their head to one side) in order to gain access to food sources (Pomeroy, 1962). Regardless of beak deformities, longer, wider, and deeper beaks were correlated with higher bite forces in several species of finches (Herrel et al., 2005) and even though this is a different type of beak use in comparison to poultry (i.e. bite vs. peck), this relationship may also exist in poultry. In another study, bite force and time spent husking seeds were negatively correlated (van der Meij and Bout, 2006). The data from the research with finches suggest that changes in beak use, bite force, or beak efficacy can be modulated by alterations in the biomechanical structure of the beak and head (Podos and Nowicki, 2004), which are separate from the modulations caused by acute or chronic pain associated with beak trimming in poultry.

The objectives of this study were to determine the effect of two abrasive pecking devices as well as placement location of the devices in the cage on beak shape (length and beak tip angle) and peck force in two different breeds of laying hens. We hypothesised that use of both devices would result in shorter beaks and beak tips with larger angles (i.e. blunter) and that these beaks would result in less force applied to the force plate.
6.3 MATERIALS AND METHODS

6.3.1 Ethical Considerations

All procedures were approved by Scotland’s Rural College’s animal ethics committee and the University of Guelph’s Animal Care Committee and were in accordance with the Canadian Council for Animal Care’s guidelines.

6.3.2 Animals

Thirty-six intact beak Columbian Rocks and 36 intact beak White Leghorns (both Shaver strain) were used for this experiment. The breeds were used for breeding purposes at the research station, one of which (Columbian Rocks) had historically been observed to cause substantial feather damage to their conspecifics and the other (White Leghorns) was most similar to commercial breeds used in North American egg production. However, for this particular flock, the White Leghorn strain had higher mortality during rearing than the Columbian Rocks (9.8% vs. 3.5% for White Leghorns and Columbian Rocks respectively from 0-14 weeks of age (see Habinski et al. (2016)). Both breeds were housed in Farmer Automatic Combi Pullet cages (Clark Ag Systems, Ontario, Canada) at the Arkell Research Station in Guelph in breed-specific groups from 1 day to 16 weeks of age, where they were used for another experiment (Habinski et al., 2016). Hens of both breeds were not beak treated because as adults, the hens in the breeding program were routinely housed individually (which limits bird-to-bird pecking) for selection of egg laying traits. At 16 weeks of age, pullets were returned to the selection flock and thus moved into individual cages. All birds were supplied with *ad libitum* feed (standard diets according to age) and water via nipple drinkers. Birds were reared and housed according to the research farm’s standard operating procedures. During the experimental period, the lights were on a 16L:8D schedule, with a 20 minute dawn and dusk setting.
6.3.2.1 Experimental Treatments

With a total of ten treatments, the experiment was set up as a $2 \times 2 \times 2$ factorial (+2 controls) with breed, beak blunting device type and beak blunting device location as the main three factors (Table 6.1).

Table 6.1 Experimental treatments. All hens were housed individually in conventional cages.

<table>
<thead>
<tr>
<th>Breed</th>
<th>Device Type</th>
<th>Device Location</th>
<th>No. Cages</th>
</tr>
</thead>
<tbody>
<tr>
<td>White Leghorn (WL)</td>
<td>None (NA)*</td>
<td>N/A (NA)*</td>
<td>12</td>
</tr>
<tr>
<td>WL</td>
<td>Cuttlebone (CB)</td>
<td>Vertical In Cage (VC)</td>
<td>6</td>
</tr>
<tr>
<td>WL</td>
<td>CB</td>
<td>Feed Trough (FT)</td>
<td>6</td>
</tr>
<tr>
<td>WL</td>
<td>Blunting Board (BB)</td>
<td>VC</td>
<td>6</td>
</tr>
<tr>
<td>WL</td>
<td>BB</td>
<td>FT</td>
<td>6</td>
</tr>
<tr>
<td>Columbian Rock</td>
<td>NA*</td>
<td>NA*</td>
<td>12</td>
</tr>
<tr>
<td>CR</td>
<td>CB</td>
<td>VC</td>
<td>6</td>
</tr>
<tr>
<td>CR</td>
<td>CB</td>
<td>FT</td>
<td>6</td>
</tr>
<tr>
<td>CR</td>
<td>BB</td>
<td>VC</td>
<td>6</td>
</tr>
<tr>
<td>CR</td>
<td>BB</td>
<td>FT</td>
<td>6</td>
</tr>
</tbody>
</table>

* Denotes control groups

The beak blunting devices included large (15 cm) cuttlebones (Prevue Hendryx Pet Products, Chicago, IL, USA) and beak blunting boards (similar to those used in Chapter 3, 4, and 5). The beak blunting boards were made by coating pieces of wood (15 cm × 7 cm × 1 cm) with an abrasive paste (S N Supplies, Lincoln, UK). The devices were placed in one of two different locations within the cages: in the feed trough (Figure 6.1a and Figure 6.2a) or fixed vertically to the side of the cage (Figure 6.1b and Figure 6.2b). Cuttlebones fixed vertically in the cage were
placed against a wooden backing to prevent damage from the neighbouring hen. Devices were placed into the cages at 30 weeks of age, one week after baseline measurements for beak morphology and pecking force were recorded. Hens were checked on a daily basis and the devices were replaced as needed (i.e. when cuttlebones were depleted).

![Figure 6.1](image1.png)

**Figure 6.1** Photographs of cuttlebones in a) the feed trough and b) vertically in the cage.

![Figure 6.2](image2.png)

**Figure 6.2** Photographs of the beak blunting boards in a) the feed trough and b) vertically.
6.3.3 Data Collection

6.3.3.1 Beak Shape

At 29, 35, and 40 weeks of age, photographs of each of the hens’ beaks were taken using a digital camera (Canon Powershot ELPH160, Canon Canada Inc., Mississauga, ON, Canada) against a background which included a ruler for size reference (Figure 6.3). Upper mandibles (i.e. culmen length) were measured from the anterior edge of the comb tissue to the beak tip (Figure 6.4) as well as the angle of the beak tip using tpsDig2 software (SUNY Stony Brook Morphometrics, Stony Brook, NY, USA). The tracing technique was similar to that used in Chapter 3 (Figure 6.4), however for this study, two observers (blind to treatment), rather than just one (as in Chapter 3), measured each photograph and re-measured photographs once again if the difference between the two observers exceeded 5%.

Figure 6.3 Example of photograph used for upper mandible length and beak tip angle measurements. Bird ID number was the only identifying factor in the photographs so that the observers could remain blind to treatment during measurement.
6.3.3.2 Peck Force

Peck force tests were conducted within the two days following collection of beak shape photographs. An arena was built for testing purposes (Figure 6.5), with the force plate (Bertec Corporation, Columbus, OH, USA) fixed vertically to the back wall of the test pen. The force plate (15cm × 15cm) and its output was amplified via a digital signal converter (Bertec AM6500) that was connected to a computer during testing. Each hen was tested individually, though two neighbouring hens were present to reduce isolation anxiety. Each hen was exposed to the testing arena for approximately five minutes in the week prior to baseline testing. Each test lasted up to five minutes, and was shorter if the hen pecked more than 20 times in the first 30 seconds. Hens were encouraged to peck the force plate by restricting feed access for 2–3 hours prior to testing and by gluing feed particles, brown feathers, white feathers, and shiny stickers to each of the four
quadrants of the force plate. All tests were video recorded and time stamped to validate pecks and peck types against the force plate output. The forces applied to the plate were measured in milliNewtons (mN) in three-dimensions (Fx, Fy, and Fz, see Figure 6.6) and were automatically recorded every millisecond for the duration of the test.

Figure 6.5 Test arena for peck force. The middle hen was always the test subject with one hen on either side (pictured here with just one) to reduce isolation anxiety.
Figure 6.6 Diagram of force plate (Bertec Corporation, Columbus, Ohio, USA) and directions of force in three dimensions: anterior-posterior (Fx), medial-lateral (Fy) and vertical (Fz). During testing the plate was fixed vertically, therefore the Fz measurements represented the force applied perpendicular to the surface of the plate.

6.3.4 Statistical Analyses

All data were analysed using linear mixed models (LMMs) in Genstat (16th edition, VSN International, Hemel Hempstead, UK). Approximate F tests are reported, when available, and Wald tests otherwise, with a significance threshold of $\alpha = 0.05$.

6.3.4.1 Beak Shape

The upper mandible lengths were averaged between both observers and analysed using an LMM with age, breed, enrichment location, enrichment type, and some logical interactions in the fixed model (Table 6.2). The number of device replacements (as a continuous covariate) was also included in the fixed effects model. The random model included age within individual bird.
Residuals were normalised by a natural logarithm transformation prior to analysis. The same model and analysis were used for the angle of the beak tip.

<table>
<thead>
<tr>
<th>Table 6.2 Statistical models used for each of the observed parameters.</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Parameter</strong></td>
</tr>
<tr>
<td>Upper Mandible Length, Beak Tip Angle</td>
</tr>
<tr>
<td>Peck Force</td>
</tr>
<tr>
<td>Cuttlebone Use</td>
</tr>
</tbody>
</table>

6.3.4.2 Peck Force

For each peck force test, the force plate software (Bertec Acquire 4, Bertec Corporation, Columbus, OH, USA) recorded data at a frequency of 1000Hz and produced one Excel file (force in all directions recorded every millisecond), which could be used to produce a graph (Figure 6.7). To prepare the data for analysis, the files for each of the hens at each of the three testing periods were collated into one large file. Data were then subjected to a filter so as to isolate intervals associated with individual pecks. The filter was created so that it identified peaks greater than 2000 mN and extracted the data from 0.05 seconds prior and 0.10 seconds after this peak. The range in
time was set because there was a rebounding wave associated with each peck and, depending on the initial force, this rebound could itself exceed 2000 mN and would otherwise be automatically identified as a separate peck (Figure 6.8), which was known not to be the case based on the video footage. This dataset was even further condensed, as summary statistics (e.g. minimum and maximum forces in each of the directions) for each peck were calculated so that it included only one line of data for each individual peck. During the test, hens were expected to peck directly onto the flat surface of the force plate. However, this was not always the case, as hens sometimes pecked at the sides, the cable, or at the bolts and wood that fixed the plate in place. Therefore, peck types were categorised into two main groups: ‘true’ pecks, which were directed onto the front surface of the force plate, and ‘non-true’ pecks, which were those that still registered a force greater than 2000mN but were not directed to the front surface of the force plate. Forces that were known to be ‘non-true’ pecks were omitted either manually (based on notes of unusual occurrences, e.g. if a hen bumped into the force plate with her body) or by applying an automatic filter that eliminated pecks that had characteristics of ‘non-true’ pecks. Based on information gained from the video recordings of the tests and correlating peck types with force plate output, it was evident that ‘true’ pecks differed from ‘non-true’ pecks in the force they created (Figure 6.8). True pecks had relatively high absolute ratios between the largest positive Fz force and the lowest negative Fz force (i.e. >1.5).

An LMM was fitted to the data (with natural logarithm transformation) for each directional force with age, breed, enrichment type, and enrichment location as the fixed effects (Table 6.2). Age was interacted with each of the treatment factors as well. Individual bird and age were included as random effects. For forces in the Fx and Fy directions (which included both positive
and negative values), a constant (the smallest value within the dataset) was added to ensure all of the values were equal to or greater than zero.

![Figure 6.7 Example of force plate output in the Fz direction. Time was measured in seconds along the x axis (this trace was approximately two minutes in duration).](image)

![Figure 6.8 Example of force plate output for a) true peck with large (>1.5) absolute ratio between maximum and minimum values (i.e. absolute value (6000:-2000)=3) and b) non-true peck with small absolute ratio (i.e. absolute value (6000:-5000)=1.2). Each graphic represents approximately 0.5 seconds of data and only one peck at the force plate or surrounding area.](image)
### 6.3.4.3 Cuttlebone use

Because there was some evidence that the cuttlebones were being used often by some hens more than others, the effect of cuttlebone use (measured by number of cuttlebone replacements) on changes in beak length or beak tip angle from 29-40 weeks of age was assessed using LMMs. Breed was included in the fixed effects to account for the differences in cuttlebone use between breeds. Individual bird was used as the random effect. For these tests, data from hens housed with blunting boards or without any devices were excluded.

### 6.4 RESULTS

Overall, cuttlebones were well-used (Figure 6.9). Of the 24 hens with cuttlebones, 12 of them had their cuttlebones replaced at least once over the 10-week study period (number of replacements mean (SED): 6.42 (4.25), range: 0–14). Of these 12 hens, nine were Columbian Rocks, three were White Leghorns, eight were located in the feed trough, and four were fixed vertically in the cage.
Figure 6.9 Examples of damage to cuttlebones on right. Large (6 inch) cuttlebones were purchased from pet stores (pictured on left) and replaced in each cage as needed.

6.4.1 Beak Shape

Upper mandible length was affected by age (P<0.001), as beaks were longest at 35 weeks of age, but there was no change between 29 and 40 weeks of age (Figure 6.10). Overall, Columbian Rocks had longer upper mandibles than the White Leghorns (P=0.011). The presence of cuttlebones only tended to decrease beak length (P=0.070). There were no differences due to device location (P=0.331) and none of the interactions were significant.
Figure 6.10 Mean upper mandible lengths (cm) predicted from LLM back transformed (±SE). Average SEDs (on transformed scale) for Age=0.012, Breed=0.015, Device Type=0.029, and Device Location=0.027. Treatment factors include: Columbian Rock (CR), White Leghorn (WL), Blunting board (BB), Cuttlebone (CB), Feed trough (FT), Vertical in cage (VC), No enrichment present (NA).

Beak tip angle was affected by age (P<0.001) and breed (P<0.001), as beak tips had smaller angles (i.e. sharper) at 40 weeks of age and for Columbian Rocks (Figure 6.11). Device type and location, as well as all interactions, were not significant.
Figure 6.11 Mean angles (°) predicted from LLM back transformed (±SE). Average SEDs (on transformed scale) for Age=0.027, Breed=0.045, Device Type=0.067, and Device Location=0.077. Treatment factors include: Columbian Rock (CR), White Leghorn (WL), Blunting board (BB), Cuttlebone (CB), Feed trough (FT), Vertical in cage (VC), No enrichment present (NA).

6.4.2 Peck Force

Maximum peck forces in all directions were affected by age (P<0.001), including the sum of all the forces, Ftotal (Figure 6.12). For all directions, the mean maximum peck force was strongest at 29 weeks of age, then decreased for 35 and 40 weeks of age. Age x breed was significant in the Fx direction (P=0.024), as the White Leghorns pecked more to one side than the Columbian Rocks at 29 weeks of age (Table 6.3). All other factors and interactions were not significant.
Figure 6.12 Mean maximum peck forces (back transformed from LMM predicted means±SE) in all directions by time (P<0.001 for Fx, Fy, Fz and Ftotal). Average SEDs (on transformed scale): Fx=0.032, Fy=0.035, Fz=0.050, Ftotal=0.048.

Table 6.3 Maximum peck force in the Fx direction for age × breed (P=0.018). Predicted means and SEs (from LMM) are presented, with back transformed means in parentheses. Average SED on transformed scale=0.053.

<table>
<thead>
<tr>
<th>Age (weeks)</th>
<th>Columbian Rocks</th>
<th>White Leghorns</th>
</tr>
</thead>
<tbody>
<tr>
<td>29</td>
<td>8.25±0.04 (3839.13)</td>
<td>8.44±0.04 (4619.31)</td>
</tr>
<tr>
<td>35</td>
<td>8.08±0.04 (3213.13)</td>
<td>8.12±0.04 (3350.95)</td>
</tr>
<tr>
<td>40</td>
<td>8.07±0.04 (3197.10)</td>
<td>8.09±0.04 (3268.22)</td>
</tr>
</tbody>
</table>

6.4.3 Cuttlebone use

There was no significant relationship between total cuttlebone use and changes in beak length from 29 to 40 weeks of age (P=0.632). However, there was a weak tendency for beak tip
angles to get bigger from 29 to 40 weeks of age (i.e. beak tips became blunted) as cuttlebone use increased (P=0.070, Figure 6.13).

![Figure 6.13](image)

**Figure 6.13** Changes in beak tip angle from 29 to 40 weeks of age against total cuttlebone use, with each dot representing data from one hen (n=24).

### 6.5 DISCUSSION

Cuttlebones were well used as indicated by the relatively high replacement rate for some of the hens and based on anecdotal behavioural evidence. Their attractiveness here reflects that in other bird species: in a magazine article, one author wrote about feeding cuttlebone to a naïve male Hornbill, who apparently pecked and injested the cuttlebone within 30 minutes (Martin, 1982). Taylor (1996) reported increased cuttlebone intake for canaries that were not given oyster shell (soluble grit) compared to those that were. In contrast, the blunting boards did not appear to attract the same amount of attention, based on a lack of deterioration and on anecdotal behavioural observations, which is similar to what was found in Chapter 3. The lack of interest in the blunting
boards in both studies suggests that they were inherently unattractive as they appeared to be ignored even in the barren individual cages in the current study. Similarly when individually caged hens were housed with porous concrete blocks, the hens showed very little pecking (0.12 pecks/h) directed toward the blocks (Holcman et al., 2008). Therefore, any future studies on the effectiveness of this type of blunting material would be best to place it where hens would come in contact with it when they were pecking at feed (e.g. bottom of the feed trough).

Surprisingly, beaks were observed to be the longest at 35 weeks of age, with 29 and 40 weeks of age being shorter and similar to each other. We expected the hens without pecking devices to have beaks that stayed the same length or grew over time and for the pecking devices to result in a shortening over time. Gabrush (2011) found beak length to be consistent from 38–57 weeks of age. Similarly van der Weerd et al. (2006) found consistency in beak length from 45–61 weeks of age. Potentially, there may have been an error in measuring beak length due to the angle of the photographs at 35 weeks, though it is not clear that there were any differences in the photographs obtained at the different ages. Columbian Rock hens had longer and sharper beaks. Although for this particular flock the White Leghorns had higher mortality during rearing (potentially pecking-related), this farm has had high feather pecking and cannibalism in previous flocks of Columbian Rocks. It is possible that the longer, sharper beaks observed for the Columbian Rocks may be linked to the historically higher pecking related injuries.

Provision of cuttlebones tended to reduce beak length though this difference was not statistically significant. We had expected to find a correlation between overall cuttlebone use and change in beak length (i.e. hens that used the cuttlebone more would have larger reductions in beak length), however this was not the case. These lack of differences may have been due to the small sample of hens who regularly used the devices; only 12 of 24 had the cuttlebones replaced and
even fewer hens (9) had them replaced more than once, which may have decreased the statistical power of the study. That being said, these data show promise for the use of beak blunting material that offers some positive (nutritional or sensory) feedback. For some of the hens that used the cuttlebones quite heavily, the cuttlebones were usually pecked immediately following replacement. As it appeared that some hens were quite motivated to peck the cuttlebones, any effects on hen health, egg production, and eggshell quality need to be evaluated as the cuttlebones may have been ingested and would have thus supplied some minerals such as calcium. Diets for commercial laying hens are specifically formulated to meet nutritional needs so providing extra calcium may disrupt the sensitive calcium:phosphorus ratio. There did not appear to be any effect of the blunting boards on beak length when compared to hens housed without any device, which suggests that the boards were not used to a large enough extent to wear the beak tip down. In addition, anecdotal observations for behaviour also corroborates the notion that cuttlebones were pecked much more readily than the boards.

The lengths of the beaks measured in this study were slightly longer on average than the beaks measured by Van der Weerd (2006). However, the method of measurement differed and was probably more accurate in the current study as we used digital morphometrics to precisely follow the curve of the beak from specific landmarks and the former study used metal mechanical callipers which would have measured the beak as though it were a straight line.

An unexpected result was that beak tip angle was smallest at 40 weeks of age, suggesting that the beaks were sharpened over time. While it was a concern that the ‘beak blunting’ devices may in fact act to sharpen the beaks, there was a trend for hens with high device use to have an increase in beak tip angle from 29 to 40 weeks of age, though it was not statistically significant. More precise methods to measure beak morphology and changes in overall shape have been
described by van der Meij and Bout (2008) and Dalton et al. (2015), where additional landmarks on each beak were digitised so that the whole of the beak shape could be quantified using principle components analysis. However, for the current experiment, we chose to adapt a similar method of measurement for beak tip angle as was used in Chapter 3 as it was the quickest and simplest way to gauge beak sharpness. That being said, interobserver reliability was relatively weak, especially compared to measurements in beak length and thus more subtle differences in beak shape or beak tip angle may have been elucidated had we used a more precise method.

Neither of the pecking devices affected the peck force in any of the directions. Peck force can be controlled by components of the biomechanical structure of the head and beak or by muscular architecture (Podos and Nowicki, 2004). Herrel et al. (2005) and Soons et al. (2010) suggested that longer beaks (in finches) were correlated with larger bite forces. Though hen pecks probably differ in motor patterns from ‘nut-cracking bites’ by finches, it was hypothesised that hens with shorter beaks (which the hens with the devices were expected to have) to produce smaller forces with their pecks. However, though they found positive correlations with beak length and bite force, Soons et al. (2010) found that beak width and depth were better predictors of bite force than length. There was no correlation between beak length and peck force at any age in the current study. Similarly, no significant correlation was found between beak tip angle and peck force in the current study. Rico-Guevara and Araya-Salas (2014) found that the interaction between beak curvature and sharpness explained some of the variation in peck force for hummingbirds (i.e. more curved and pointier beaks required less force to puncture a membrane). Finally, age affected peck force, as hens appeared to peck the hardest on the first testing day. This may be a result of the relative novelty of the testing arena and force plate, or the lack of feedback from the device as hens learned that the particles of feed were glued to the plate. In contrast, data reported by Jongman et
al. (2008) suggested an increase in peak peck force from 12-62 weeks of age, though age effects were not tested statistically in that study. We also observed an age × breed effect for peck force in the Fx direction. Though we were most interested in the Fz force (horizontal to the ground) of a peck, we were also curious about other components of pecks (i.e. forces in other directions), as hens with blunter beaks may have to adapt their pecking technique (e.g. peck on an angle) as gross changes in beak morphology can affect pecking style (Pomeroy, 1962). The minor changes in beak shape observed over the 10 week period were most likely not severe enough to cause a change in peck force.

6.6 CONCLUSIONS

Overall, the inclusion of cuttlebones within the cage environment showed promising results in terms of beak length reduction, although the practicality of such a device being used in commercial farms and the effect of beak blunting on injurious pecking behaviour has yet to be determined. Though the results for the blunting boards showed no indication of being effective, this was most likely due to the fact that they were not used to a large enough extent. If blunting boards are to be used, they should be first tested at the bottom of the feed trough as done by Van der Weerd (2006), as they do not appear to create or maintain pecking attraction.
Chapter 7 General Discussion

The aims of this chapter are to outline the major findings of the previous chapters in relation to the original hypotheses, how each of the experiments could be improved upon, and an opinion on the direction of future research in this area.

7.1 OVERALL THESIS OBJECTIVE

Overall, the objective of this thesis was to assess various methods of mitigating feather pecking and cannibalism in furnished cage-housed hens without the need for beak alterations. This thesis set out to investigate the effects of extra enrichments, breed, and dietary alterations on a number of behavioural and welfare-related parameters. Finding less invasive solutions (compared to any type of beak alteration) to reducing injurious pecking has huge potential to improve the welfare of many laying hens around the globe. Though this behavioural problem has been a welfare topic for the past few decades, it is still of great importance as there is still no reliable method for reducing the quantity or quality of injurious pecking (other than beak alteration). Even more pressing is the political decision surrounding the housing and treatment of farm animals. For instance, in the UK, Defra has entertained a potential ban on any type of beak alteration twice in the past decade. Although the latest proposal for a ban has been turned down (Defra, 2015a), it surely will be of importance throughout the next few years. Therefore, finding a solution to injurious pecking, without the use of beak alterations, is of great importance.
7.2 CHAPTER HYPOTHESES

In Chapter 3, the non-beak treated hens were expected to have higher mortality, worse feather cover, and display more injurious pecking behaviour, but that the addition of extra enrichments would significantly reduce these negative outcomes. Because some Lohmann strains have been used in countries where altering beaks is already prohibited, there was also an expected improvement in welfare parameters with Lohmann Classic hens in comparison to the Hyline strain, and for these differences to be most pronounced between the non-beak treated groups. The presence of extra enrichments was expected to alter time budgets and perhaps to satisfy foraging motivation as well, and in turn to reduce mortality, feather damage, and injurious pecking behaviour. The extra enrichments (mainly the beak blunting board) were also expected to reduce upper and lower mandible lengths, as well as increase beak tip angle (i.e. make the beaks blunter).

In Chapter 5 and Chapter 4, the extra enrichments again were expected to alter time budgets and perhaps to satisfy foraging motivation as well, and in turn to reduce mortality, feather damage, and injurious pecking behaviour. The aim was that differences between hens with and without extra enrichments would be more pronounced than in Chapter 3 due to the modifications imposed to make them (hopefully) more effective. It was not expected that the extra enrichments would affect any of the production parameters. For the dietary alterations, the hypothesis was that the extra fibre would act to alter time budgets (i.e. by making the hens spend longer feeding and thus have less time for feather pecking), but also to act via the gut-brain axis to reduce the motivation to perform injurious pecking and thus reduce mortality and feather damage. Some differences in feed intake were expected (as hens would need to eat larger volumes of fibre-rich diets to match the nutrient intake of control-fed hens). However, the hens were expected to fully compensate by eating more so that body weight and thus egg production or egg weight would not be affected.
Excreta dry matter was anticipated to be reduced with control diets and therefore there would be more dirty eggs (as expected with stickier excreta) compared to fibre diets. Because hens are naturally omnivorous and it has been suggested anecdotally that vegetarian diets may predispose hens to outbreaks of injurious pecking (McKeegan et al., 2001; van Krimpen et al., 2011). Therefore, it was hypothesised that the hens fed the animal protein-based diet would perform less injurious pecking, and thus have fewer mortalities and better feather cover, than the all vegetable diet. Though the digestibility of animal proteins has been observed to be greater than that of plant protein, there were no expected differences in production parameters due to dietary protein source as the diets were formulated to contain similar levels of metabolisable energy as well as digestible amino acids.

In Chapter 6, the anticipation was shorter and blunter beaks for hens housed with either the cuttlebone or beak blunting board. It was also thought that the devices in the feed trough would be more readily used and therefore result in hens with shorter and blunter beak tips. In addition, changes in peck force were expected for hens with the devices, due to changes in beak morphology.

### 7.3 CHAPTER OUTCOMES

The major results from each of the chapters are summarized and listed in the table below (Table 7.1).

The efforts to find a robust solution throughout the length of this Ph.D. for reducing injurious pecking did not come to fruition as hoped. Though there were some promising results in each of the experiments (in particular for the extra enrichments), there was not any overwhelming evidence to suggest that one treatment over the other would have a large effect on injurious pecking.
or associated mortality or feather damage. That being said, some of the predictions were, in fact, confirmed in these experiments.

For the first study, an increase in mortality for non-beak treated hens was observed, however, this was only significant when all 80 hens from each of the two culled cages were included. Though removing these cages from study was necessary to avoid further pain and suffering, and to stay within the limits outlined in the Home Office Project Licence, it would have been interesting from a scientific perspective to see how far the pecking-related mortality would spread. Originally, the mortality data was analysed by excluding the culled hens (i.e. ‘minimum’ mortality), and these results were always presented with the caveat that we did not have “true” data for two cages (i.e. they were not allowed to run their ‘natural’ course). It was a Scottish egg producer that pointed out that, to the general public, it appeared this data was misleading. From then on, the data were analysed by including a ‘maximum’ mortality variable. In addition to the mortality differences due to beak alteration, the hens with intact beaks also had longer upper mandibles (which was to be expected) and more feather damage.

Breed effects were evident in the first study as well as the third study, though they were not always in the direction that had been hypothesised. In the first study, the Lohmann Classic strain was expected to fare better than the Hyline Brown, especially for the non-beak treated group, as this strain is housed in other countries that do not alter beaks. However, the opposite was found for the majority of welfare outcomes, including increased mortality, increased bird-to-bird pecks, and increased feather damage. However, though the breed effect was not in the anticipated direction, there still appeared to be a clear-cut “winner”, as the Hyline breed had relatively low mortality (especially for the beak treated hens) and better plumage condition. As for the breed effects in the third study, the Columbian Rocks had longer and sharper beak tips than the White
Leghorns. Though no breed differences for beak shape were detected in the first study, others have indicated that beak dimensions (for finch species) are, at least in part, genetically determined, with relatively high heribilities (Grant and Grant, 2000). In addition, Hutt (1949) suggested that severe beak abnormalities in poultry are heritable. Therefore, it is possible that beak shape can be selected for; however, this would only be effective for breeding companies if beak shape affected feather pecking rates or efficacy.

In the second study, there were not many obvious trends for or against the welfare outcomes associated with the dietary alterations. The hens fed the extra fibre were observed to perform fewer spot pecks and the hens fed the animal protein caused more damage to the enrichments, but this did not correspond with overt changes in any other welfare measure. Though financial outcomes were not calculated for this study, the benefits of the dietary alterations most likely would not have outweighed the additional cost of the treatments.

The only consistent factor throughout the three studies was the presence of extra enrichments (and/or beak blunting devices). In the first study, a reduction in bird-to-bird pecking was observed, though there were no associated differences in mortality or feather damage. There was little evidence of use of the beak blunting boards (via behaviour and damage scores), and they did not have an effect on beak shape, which all together suggests that they were not used. The extra enrichments were slightly altered between the first and second studies, with the pecking mats increasing in surface area, the rope ends were not sealed and there were more rope ends per hen. In addition, the beak blunting boards were placed at the bottom of the feed troughs, however, due to an oversight, beaks were not measured. However, for this study, hens with extra enrichments spot pecked less frequently and had better feather cover. Again, there were no differences in mortality (though mortality was low overall). In contrast to the first study, there were no observed
differences due to the main effect of extra enrichments for bird-to-bird pecking. In the final study, only devices that were designed to blunt the beaks were assessed. Though the differences did not reach statistical significance, there was a tendency for one of the devices (the cuttlebones) to shorten upper mandibles and for increased cuttlebone use to dull beak tips. Because less than half of the hens used more than two cuttlebones, the data may have reached significance given a larger number of hens.

**Table 7.1** Outline of major results from each study. L, Lohmann Classic; H, Hyline Brown; T, beak treated; NT, non-beak treated; NE or N, no extra enrichment; EE or E, extra enrichment; P, plant protein; A, animal protein; C, control fibre level; F, extra fibre; WL, White Leghorn; CR, Columbian Rock; CB, cuttlebone.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Age Effects</th>
<th>Factor Effects</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Chapter 3</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mortality</td>
<td>N/A</td>
<td>L &gt; H, NT &gt; T (max.), HT &lt; HNT, TEE &lt; TNE, LNT highest</td>
</tr>
<tr>
<td>Behaviour (Focal)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bird-to-bird pecks</td>
<td>↑</td>
<td>L &gt; H, NE &gt; EE</td>
</tr>
<tr>
<td>Enrichment pecks</td>
<td>↓</td>
<td>Near rope &gt; near mat, near nest</td>
</tr>
<tr>
<td>Other pecks</td>
<td>↑</td>
<td>LT &gt; LNT, HNT &gt; HT</td>
</tr>
<tr>
<td>Bird changes</td>
<td>-</td>
<td>L &gt; H, NT &gt; T</td>
</tr>
<tr>
<td>Feather Cover</td>
<td>↓</td>
<td>L &lt; H, T &lt; NT, Tail worst</td>
</tr>
<tr>
<td>Enrichment Damage</td>
<td>↑</td>
<td>L &gt; H, NT &gt; T</td>
</tr>
<tr>
<td>Beak Shape</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Upper mandible length</td>
<td>N/A</td>
<td>NT &gt; T</td>
</tr>
<tr>
<td>Lower mandible length</td>
<td>N/A</td>
<td>T &gt; NT, TEE &gt; NTEE</td>
</tr>
<tr>
<td>Overhang</td>
<td>N/A</td>
<td>NT &gt; T</td>
</tr>
<tr>
<td>Beak tip angle</td>
<td>N/A</td>
<td>-</td>
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</tbody>
</table>
### Chapter 4

#### Mortality
- N/A

#### Behaviour (Focal)

<table>
<thead>
<tr>
<th>Category</th>
<th>Trend</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feed pecks</td>
<td>↑</td>
<td>F &gt; C at 16 weeks, C &gt; F at 34 wk, Near rope &lt; near mat, near nest</td>
</tr>
<tr>
<td>Spot pecks</td>
<td>↓</td>
<td>FE &lt; FN, but CE = CN, Near nest &gt; near rope, near mat</td>
</tr>
<tr>
<td>Drinker pecks</td>
<td>cubic</td>
<td>PN &gt; PE, but AN = AE, Near rope &lt; near mat</td>
</tr>
<tr>
<td>Other pecks</td>
<td></td>
<td>Near rope &gt; near mat, near nest</td>
</tr>
<tr>
<td>Beak pecks</td>
<td>↓</td>
<td>AFE lowest, Near nest &lt; near mat, near rope</td>
</tr>
<tr>
<td>Bird-to-bird pecks</td>
<td></td>
<td>AC lowest at middle of study, Near mat &gt; near rope &gt; near nest, Tendancy for E&lt;N</td>
</tr>
<tr>
<td>Enrichment pecks</td>
<td></td>
<td>Near nest &lt; near rope, near mat</td>
</tr>
<tr>
<td>Bird changes</td>
<td>cubic</td>
<td>P &gt; A near middle, Near mat &lt; near rope, near nest</td>
</tr>
</tbody>
</table>

#### Behaviour (Scan)

<table>
<thead>
<tr>
<th>Category</th>
<th>Trend</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Feed pecks</td>
<td>cubic</td>
<td>AF &gt; others at end</td>
</tr>
<tr>
<td>Spot pecks</td>
<td>↓</td>
<td>N &gt; E, C &gt; F after 22 weeks</td>
</tr>
<tr>
<td>Bird-to-bird pecks</td>
<td></td>
<td>CN and FE &gt; CE and FN for 22 – 28 wk, PC and AF &gt; PF and AC until 25 wk</td>
</tr>
<tr>
<td>Enrichment pecks</td>
<td>↑</td>
<td>-</td>
</tr>
<tr>
<td>Feather Cover</td>
<td>↓</td>
<td>E &gt; N</td>
</tr>
<tr>
<td>Enrichment Damage</td>
<td>↑</td>
<td>A &gt; P, Tail worst</td>
</tr>
</tbody>
</table>

### Chapter 5

#### Body Weight
- age×protein×fibre×enrichment

#### Feed Disappearance
- F > C

#### Egg Production
- -


<table>
<thead>
<tr>
<th>Good eggs</th>
<th>↑</th>
<th>N &gt; E, PF &gt; PC, AC &gt; AF, age×fibre</th>
</tr>
</thead>
<tbody>
<tr>
<td>Extra-cuticular calcium</td>
<td>↓</td>
<td>F &lt; C at start, AF &lt; others at start</td>
</tr>
<tr>
<td>Brown spots</td>
<td>↑</td>
<td>A &gt; P, E &gt; N, PF &gt; PC, but AF = AC</td>
</tr>
<tr>
<td>Dirty/bloody</td>
<td>↓</td>
<td>C &gt; F, N &gt; E at end, FE highest at start, CE lowest at end</td>
</tr>
<tr>
<td>Cracked</td>
<td>↑</td>
<td>-</td>
</tr>
<tr>
<td>Other defects</td>
<td>cubic</td>
<td>PC highest at start</td>
</tr>
<tr>
<td>Egg Weight</td>
<td>↑</td>
<td>P &gt; A, AF &lt; the rest, C &gt; F at start</td>
</tr>
<tr>
<td>Excreta DM</td>
<td>↓ at 25 weeks</td>
<td>A &gt; P, F &gt; C</td>
</tr>
</tbody>
</table>

### Chapter 6

#### Beak Shape

- **Upper mandible length**: ↑ at 35 weeks. CR > WL, CB tended to ↓ length
- **Beak tip angle**: ↓ at 40 weeks. WL > CR

#### Peck Force

- **Fx**: ↓. WL > CR at 29 weeks
- **Fy**: ↓. -
- **Fz**: ↓. -
- **Ftotal**: ↓. -

#### Cuttlebone Use

- **N/A**: ↑. device use tended to ↑ beak tip angle

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### 7.4 Future Research

Given the encouraging results from the addition of the extra enrichments, it would be interesting to pursue this further, both in an attempt to alter time budgets and to shorten and blunt beak tips. The ropes appeared to increase their attractiveness the more frayed they became, which appeared to be the opposite to the pecking mats (as they deteriorated with increased use). In addition, more pecks were observed to be directed at the ropes than at the mats (for both the first
and second studies), suggesting they were more attractive than the mats even when their ends were heat-sealed. In a commercial setting, enrichments that require minimal maintenance would be preferred over those that require multiple replacements over time (which would cost money and time). Therefore, it would be interesting to test the effect of rope inclusion only on multiple commercial flocks, especially those known for high levels of feather pecking.

In addition, there was evidence to suggest that cuttlebones could be effective at reducing beak length as well as beak sharpness, though this would need to be repeated. However, cuttlebones would not be suitable for commercial application as they are brittle and break relatively easily, and would therefore require constant replacement for large groups of hens. In addition, the effect on mineral metabolism (including aspects of bone health, egg production, and egg quality) should be assessed prior to large-scale implementation. However, if a similar, more durable material could be constructed this may solve the fragility and nutritional issues. That being said, the blunting paste was not effective at attracting hens, and therefore was ineffective at changing beak shape.

For the most part, the treatments imposed on the hens throughout this study happened prior to any major development or outbreak of injurious pecking. It would be interesting to test whether the treatments (dietary or environmental) would have improved behaviour, mortality, or feather cover for a flock with high levels of injurious pecking.

Finally, as this type of research is very applied, it would be interesting to determine the economic viability of different interventions. For example, at what point would the benefit (e.g. decreased mortality) of a treatment outweigh the costs (e.g. including animal or insect protein).
Chapter 8 Appendices

Thorton Mill
Strathore Road
Thorton
Fife
KY1 4DX

24.00 RHARB 22B1
A complete feedingstuffs for pullets.

ANALYTICAL CONSTITUENTS

<table>
<thead>
<tr>
<th>Component</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protein</td>
<td>16.5 %</td>
</tr>
<tr>
<td>Calcium</td>
<td>0.95 %</td>
</tr>
<tr>
<td>Oil A-BB</td>
<td>2.5 %</td>
</tr>
<tr>
<td>Phosphorus</td>
<td>0.67 %</td>
</tr>
<tr>
<td>Fibre</td>
<td>5.1 % (total)</td>
</tr>
<tr>
<td>Ash</td>
<td>5.6 %</td>
</tr>
<tr>
<td>Sodium</td>
<td>0.16 %</td>
</tr>
<tr>
<td>Lysine (total)</td>
<td>0.0 %</td>
</tr>
<tr>
<td>Methionine</td>
<td>0.36 %</td>
</tr>
<tr>
<td>Sodium Chloride</td>
<td>0.045 %</td>
</tr>
<tr>
<td>Total acids</td>
<td>88%</td>
</tr>
<tr>
<td>Monomer acids</td>
<td>55%</td>
</tr>
</tbody>
</table>

COMPOSITION:
WHEAT, BARLEY, WHEATFEED, SUNFLOWER DECORCITED, HIGH PRO SOY A
(produced from genetically modified), CALCIUM CARBONATE, MONOCAL
PHOSPHATE, SOYA OIL (produced from genetically modified), SODIUM
BICARBONATE, TERMIN-8, VITAMIN TRACE ELEMENT SUPPLEMENT, LYSIN
SODIUM CHLORIDE, LQQ METHIONINE

Contains the 0.045% (d2 hydroxy 4 methyl thiobutanoic acid. To
tota acids 88% monomer acids 65%)

ADDITIVES (PER KG)
Vitamins:
VITAMIN A (E672) 8000 IU, VITAMIN D3 (E671) 3000 IU, VITAMIN E (E3)
50 mg

TRACE ELEMENTS:
Cupric Sulphate Pentahydrate 35 mg, Ferrous Sulphate Monohydrat
e 129 mg, Manganese Oxide 69 mg, Calcium Iodate 3.1
mg, Sodium Selenite 0.23 mg

KNZYMKS:
Reg No (4a1600) to give myo-inositol hexaphosphate phosphohydroxyl-
a (EC 1.1.3.8) 500 FTU, Reg No 4a7 to give endo xylanase (IUB No EC
3.2.1.8) 560 TXU, Endo glucanase (IUB No EC 3.2.1.4) 250 TGU

Appendix I Pullet diet formulation.
Appendix II Schematic of furnished cage used in Chapter 3. Not pictured are the nipple drinkers, plastic coated wire flooring in nest boxes, egg roll-out tray, second feed trough on far side, and mesh barrier along midline of nest boxes.
Appendix III Letter of approval from the Scottish Government for the use of animal by-products as a feed ingredient for poultry.
Appendix IV Schematic of cage used in Chapters 4 and 5. Not pictured here are the nipple drinkers and the plastic mesh bottom of nest box.
Chapter 9 Literature Cited


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