The impact of stocking density on the performance, health and welfare of turkey hens

A Thesis Submitted to
the College of Graduate and Postdoctoral Studies
in Partial Fulfillment of the Requirements
for the Degree of Master of Science in
the Department of Animal and Poultry Science,
University of Saskatchewan
Saskatoon, SK, Canada

Submitted by
Sameeha Jhetam
M.Sc. Candidate

Permission to Use Statement

In presenting this thesis in partial fulfillment of the requirements for a postgraduate degree from the University of Saskatchewan, I agree that the Libraries of this University may make it freely available for inspection. I further agree that permission for copying of this thesis in any manner, in whole or in part, for scholarly purposes may be granted by the professor or professors who supervised my thesis work, or, in their absence, by the Head of the Department or the Dean of the College in which my thesis work was done. It is understood that any copying or publication or use of this thesis or parts thereof for financial gain shall not be allowed without my written permission. It is also understood that due recognition shall be given to me and to the University of Saskatchewan in any scholarly use which may be made of any material in my thesis.

Requests for permission to copy or make of material in this thesis in whole or in part should be addressed to:

Head of the Department of Animal and Poultry Science
University of Saskatchewan,
6D34 Agriculture Building, 51 Campus Drive
Saskatoon, Saskatchewan, Canada S7N 5A8

OR

Dean
College of Graduate and Postdoctoral Studies
University of Saskatchewan
107 Administration Place
Saskatoon, Saskatchewan, Canada S7N 5A2

Disclaimer

Reference in this thesis to any specific commercial products, process, or service by trade name, trademark, manufacturer, or otherwise, does not constitute or imply its endorsement, recommendation, or favoring by the University of Saskatchewan. The views and opinions of the author expressed herein do not state or reflect those of the University of Saskatchewan and shall not be used for advertising or product endorsement purposes.

Overall Abstract

The impact of graded levels of stocking density (SD) on the performance, health, and welfare, of Nicholas Select turkey hens raised from day 0 to 11 weeks of age were evaluated in this study. Poults (n=3550/block; randomized complete design with block as trial) were randomly placed into one of four final estimated SD treatments of 30, 40, 50, or 60 kg/m^2 based on final predicted body weight at 11 weeks of age. Birds were housed in open rooms (67.5m^2) with a total of four replications per treatment. Feeder and drinker space were equalized on a per bird basis. Based on CO_2 and ammonia measures, ventilation was adjusted to equalize air quality across all treatments. Productivity was evaluated by measuring group body weight and feed consumption on day 0 and in weeks 3, 5, 8, and 11. Body weight gain and feed-to-gain ratio were calculated. At week 8 and 11, flock uniformity was evaluated on 30 birds/replicate. A brief economic analysis utilizing 2019 and 2020 poult and feed costs was performed. At week 8 and 11, footpad lesions, mobility (subjective gait scores) and feather cover and cleanliness were evaluated on 30 birds/replicate and litter samples were collected to determine moisture content (block 1 only). Incidences of aggressive pecking were recorded daily. Heterophil/lymphocyte (H/L) ratios were evaluated at 3, 5, 8, and 11 weeks of age (20 birds/replicate). Behavioural activity was recorded, and scan sampled (field of view observations) at 8 and 11 weeks of age. Data were analyzed using regression analysis in SAS 9.4 (Proc Reg and Proc RSReg; SD as independent variable). An analysis of variance was performed for all data (Proc Mixed; SAS 9.4) and a Tukey's range test was used to separate means. Differences were considered significant when $P \leq 0.05$, and trends were noted when $P \leq 0.10$. Ammonia and CO_2 were consistent across treatments for both blocks. Body weight (week 11) decreased linearly as SD increased. Feed consumption was lowest at high SD (linear for week 8-11; quadratic for week 0-11). Overall body weight gain tended to decrease as SD increased. Feed-to-gain ratio, percent mortality, and uniformity were not affected by SD. Total aggression related mortality and culls linearly increased with decreasing SD. Net room income linearly increased as SD increased. Average footpad scores worsened at week 8 as SD increased. Mobility was poorer in the 60 kg/m^2 treatment at week 11. Feather cover and cleanliness scores were poorer in the high SD treatment at week 8 and 11. Incidence of aggressive damage was highest at low SD (30 kg/m^2). The H/L ratios increased linearly with increasing SD at 5 weeks of age. At 8 weeks, H/L ratios were highest in the 40 kg/m^2 treatment (quadratic) and at 11 weeks, were highest in the 50 and 60 kg/m^2 treatments (quadratic).

Behaviour was impacted with increasing SD as the percentage of birds resting, feather pecking, and total disturbances increased. The percentage of birds standing, walking, litter pecking, environmental pecking, and performing aggressive behaviours increased with decreasing SD. Litter moisture tended to increase with increasing SD at week 11. Overall, turkey hen performance, health, and welfare were negatively affected at higher SD although economic returns were greater. Conversely, the lowest SD (30 kg/m^2) also negatively affected welfare as more aggression occurred.

Acknowledgements

I would like to begin by thanking my supervisor, Dr. Karen Schwean-Lardner, for all her support, knowledge, endless guidance and advice. Her passion for poultry research, welfare, and teaching was my biggest inspiration in pursuing this endeavor. Her encouragement pushed me to work harder and learn more each day. Thank you to my committee members, Dr. Jennifer Brown and Dr. Denise Beaulieu, for all of their suggestions and input throughout the course of my master's program. Thank you to Dr. Tim Mutsvangwa for chairing my committee meetings. I would like to thank Dr. Marisa Erasmus, my external examiner, for being a valuable part of my thesis defense.

I would also like to thank all of the staff at the U of S Poultry Center for their help with my trials. Jason Marshall, Mark Meier and Marlow Thue, I am extremely grateful for all your hard work during data collections, late nights spent building boxes, and caring for the turkeys. Your knowledge, experience, and help played a large part in making my trials successful. Thank you to Rachel Savary for the late nights helping me monitor barn ventilation. I would like to specially thank Kailyn Buchynski who shared bounds of knowledge about turkeys and her experiences and spent many hours helping me through my trials. Your guidance is greatly appreciated. I would also like to thank Tory Shynkaruk, my fellow graduate students, and the volunteers for their help with weigh backs and data collections, especially during Christmas.

Last but not least, I would like to thank my family. Jen, thank you for all your support during this journey. I am truly grateful to you for accompanying me to the barn late at night (at -30º and below) to check on the turkeys, for taking care of things when I was exhausted from long days at the barn and encouraging me on the hard days. To my parents, Shireen and Aqil, thank you for your sacrifices that allowed me the opportunity to pursue my dreams and for all the love and support you have given me. Thank you for coming to Saskatoon to help with my weigh back over Christmas. To my sisters and brothers-in-law, thank you for listening to and putting up with all my turkey stories and pictures!

I would like to acknowledge the funding agencies, Canadian Poultry Research Council, Agriculture and Agri-Food Canada and Aviagen Turkeys Inc. for their financial support. Thank you to Charison's Turkey Hatchery Ltd. for their poults.

Table of Contents

List of Tables

List of Figures

List of Abbreviations

SD	stocking density
kg/m^2	kilograms per square meter
BW	body weight
FPD	footpad dermatitis
H/L	heterophil/lymphocyte ratio
HPA	hypothalamic-pituitary-adrenal
CRH	corticotropin releasing hormone
ACTH	adrenal corticotropin hormone
F:G	feed-to-gain ratio
$F{:}G^m$	mortality corrected feed-to-gain ratio
CV	coefficient of variation
CO_2	carbon dioxide
ppm	parts per million

1.0 Chapter 1. Literature Review: the impact of stocking density on the performance, health, and welfare of turkeys and broiler chickens

1.1 Introduction

Turkey production is highly intensified, as large numbers of birds are reared in commercial production units (Coleman and Leighton, 1969; Buchwalder and Huber-Eicher, 2004). In Canada, the total number of registered turkey producers in 2019 was 522 and the total turkey production was 164.8 million kg (Turkey Farmers of Canada, 2019). More specifically, in the 37-year period between 1982 and 2019, the total production of light turkey hens (5-7 kg) has increased from 24.2 million kg to 31.5 million kg. Similarly, the total production of heavy turkey hens (7-9 kg) has increased from 12.7 million kg in 2004 to 16.1 million kg in 2019 (Turkey Farmers of Canada, 2019). However, the largest increase in poultry production is seen in the broiler chicken industry with over 1.3 billion kg of product produced in 2019; a 2.5% increase in production from 2018 (Chicken Farmers of Canada, 2019). Therefore, the amount of space that livestock animals require in intensive production systems has become increasingly important as the space allowance provided per bird can impact individual productivity, health, and welfare

Stocking density (SD) is a vital parameter that affects both economics (Coleman and Leighton, 1969; Gill and Leighton, 1984; Beaulac and Schwean-Lardner, 2018) and bird welfare (Martrenchar et al., 1999; Erasmus, 2017; Beaulac and Schwean-Lardner, 2018). Stocking density is a complicated parameter of poultry management that can have varying effects on the production, health, and wellbeing of turkeys. High SD levels and/or large group sizes can negatively influence laying hen and turkey activity or behavioural time-budgets, thereby, negatively impacting their welfare (Nicol, 1989; Keeling, 1994; Sherwin and Kelland, 1998). For example, if performing a specific behaviour will benefit a bird's health and performance, and reduced expression of this behaviour occurs because of reduced space availability, there may be physiological impairment and reduced psychological wellbeing (affective state) as the bird is unable to perform the motivated behaviour (Sherwin and Kelland, 1998). However, space allowance is only one of many factors that contribute to the wellbeing of poultry and therefore, a holistic approach is needed when evaluating the welfare of commercial turkeys.

Welfare implications of how livestock are raised have become increasingly important to consumers, however, it is often difficult to quantify. Welfare is commonly assessed by breaking down the evaluation into categories of biological functioning, affective states, and 'natural' living (Fraser, 2008). Measures of biological function includes the health, productivity, and disease status of the animal (Fraser, 2008), affective states refer to the animal's experience and emotions (Duncan, 1998; Brown and Vosloo, 2017), and natural living encompasses environmental factors that allow an animal to perform naturally motivated behaviours (Fraser, 2008).

Few studies have assessed the effects of SD on turkey hen health, welfare, and performance, and many previous studies focus on just one or two of these factors. The majority of studies have focused on turkey toms as they are reared for longer periods and reach a heavier final body weight before they are marketed. More recent studies on SD in turkeys have focused on male turkeys (Jankowski et al., 2015; Hafez et al., 2016; Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019; Bartz et al., 2020), however, the most recent research on the effects of SD in turkey hens was published in 1999 (Martrenchar et al., 1999). Though these studies have provided important information on performance, health, and welfare parameters, it is important to note the change in commercial lines from the mid-1900s to today. In a study from 1969, the mean body weight for turkey hens at 10-weeks of age was 2.01 kg (Coleman and Leighton, 1969) and in a study from 1979, the mean body weight of turkey hens at 14 weeks of age was 4.37 kg (Proudfoot et al., 1979a). In comparison, the current final predicted body weight of turkey hens at 10 and 14 weeks is 5.43 kg and 9.42 kg, respectively (Aviagen, 2015a). Another confounding factor that affects the interpretation of results from previous studies is the different levels of SD used, as some studies define high SD to be 42 kg/m^2 (Leighton et al., 1985) and others define high SD as 60-62 kg/m^2 (Martrenchar et al., 1999; Beaulac and Schwean-Lardner, 2018). Another complication is that the units used to describe SD and space requirements are often different between various scientific studies as well as between different industry requirements resulting in little consistency. This makes it more difficult to make comparisons between studies. Therefore, when comparing results of previous studies in this thesis, approximate conversions have been made to kilograms per square meter (kg/m^2) when possible.

In addition to varying SD levels and units used in previous studies, there are confounding factors in the methodology that creates challenges when evaluating the effects of SD. When evaluating SD, some studies have altered group size and maintained floor space (Moran, 1985; Noll et al., 1991; Martrenchar et al., 1999; Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019) while others have maintained group size but altered pen size/floor space (Coleman and Leighton, 1969; Denbow et al., 1984; Leighton et al., 1985; Buchwalder and Huber-Eicher, 2004; Bartz et al., 2020). For the practical application of SD guidelines, altering group size instead of changing rearing facilities is more easily implemented on farm. There is also a significant variation in group sizes, age of turkeys, bird sex, and environmental conditions utilized in the various published studies, making comparisons amongst studies difficult.

More research has focused on the effects of SD in broiler chickens than in turkeys, as broiler production is the largest poultry industry. However, it is imperative to evaluate SD effects in current turkey production systems, and more specifically turkey hens, as current information is lacking. This chapter will review SD effects in turkeys by examining previous literature findings on performance parameters, health parameters, welfare, and behaviour as well as external environmental stressors for turkeys. In addition, the effects of SD on the performance, health, and behaviour of broiler chickens previously studied will be reviewed.

1.2 Stocking Density Effects in Turkeys

A review paper by Erasmus (2017) suggested that industry guidelines have outlined the importance of SD for turkeys, with a focus on basic behavioural requirements. According to the Canadian Codes of Practice for the Care and Handling of Hatching Eggs, Breeders, Chickens, and Turkeys, birds are required to have enough space to "move freely and be able to stand normally, turn around, and stretch their wings without difficulty" (National Farm Animal Care Council, 2016). There are also additional requirements for turkeys such as daily environmental monitoring, water intake monitoring, and continuous health and/or injury data recording to ensure that SD does not negatively impacting bird welfare (National Farm Animal Care Council, 2016). Depending on the final predicted body weight, the current SD recommendations in Canada range from 40 to 65 kg/m^2 for toms and hens, with additional environmental and management requirements when housing birds at higher SD levels (National Farm Animal Care Council, 2016).

1.2.1 Performance Parameters

Stocking density effects on turkey hen performance have been examined in a small number of studies. The performance parameters studied in turkeys normally include growth, mortality, and feed efficiency. From the limited studies on SD and turkeys, it was found that as SD increases, body weight of turkey toms decreases (Coleman and Leighton, 1969; Proudfoot et al., 1979a; Leighton et al., 1985; Noll et al., 1991; Martrenchar et al., 1999; Beaulac et al., 2019;

Bartz et al., 2020). The few studies on turkey hen performance have had similar results to those on turkey tom performance and SD. In the study by Coleman and Leighton (1968), SD ranging from 6.5 to 12.1 dm^2/bird (27.5 to 48.0 kg/m^2) were examined. The average body weight of turkey hens decreased as SD increased, however, no significant effects were noted for feed efficiency. These authors suggested that body weight at a given age can be directly correlated to space allowance (Coleman and Leighton, 1969). Proudfoot et al. (1979) evaluated various SD to determine which would give the optimal economic return per bird reared. Three densities ranging from 7.4 to 14.7 dm^2/bird (30.4 to 55.1 kg/m^2) were evaluated for turkey hens and three densities ranging from 11.1 to 18.8 dm^2/bird (32.0 to 50.8 kg/m^2) for toms raised to 14-weeks of age. Body weight in both sexes was negatively affected by increasing SD, and no effect of SD was found on feed efficiency. The results of this study were similar to those found by Coleman and Leighton (1968). It was determined that monetary returns were significantly increased at higher SD as the turkeys from high SD treatments did not reach a point of diminishing returns in which the profits gained were not less than the money invested into rearing birds at the high SD level (Proudfoot et al., 1979a).

In the 1985 study by Moran, turkey hens were housed at two SD levels, 33.3 and 66.7 dm^2/bird (10.8 and 21.4 kg/m^2) to 17 weeks of age. Moran (1985) found that body weight was negatively affected by increasing SD. However, contrary to the studies by Coleman and Leighton (1968) and Proudfoot et al. (1979b), Moran (1985) found that feed efficiency was negatively impacted by increasing SD between 92 to 119 days (13 to 17 weeks) of age. A second study in the same year by Leighton et al. (1985) evaluated the effects of SD on Large White turkey hens housed at 5.6, 7.0, and 9.3 dm^2/bird (19.7 to 31.8 kg/m^2) from 0 to 8 weeks of age and 14, 18.6, and 23.2 dm^2/bird (24.1 to 40.1 kg/m^2) from 8 to 16 weeks of age. The authors found no effects of SD on body weight or feed efficiency. In comparison, Denbow et al. (1984) observed no effects of SD on production parameters in Large White turkey toms up to 8 weeks of age, however, at 12 weeks of age, body weight decreased in the highest SD treatment (42.0 dm^2/bird or 39.8 kg/m^2) and feed efficiency decreased at 12-18 and 18-20 weeks of age with increasing SD. The results showed that by 20 weeks of age, the body weight of toms grown at the intermediate SD (35.0 dm^2/bird or 31.4 kg/m^2) were not different from the lowest density treatment of 28.0 dm^2/bird (26.8 kg/m^2). Denbow et al. (1984) also examined SD effects on feed consumption. No effect was noted up to 12 weeks of age, however, from 12 to 20 weeks of age, birds consumed less feed in the highest SD treatment. The authors suggested that the age at which SD significantly affects growth performance was influenced by floor space, sex, and genetic line. Leighton et al. (1985) also proposed that female turkeys may be more tolerant of the effects of SD pressure compared to males as they observed fewer production effects in hens compared to the toms in the Denbow et al. (1984) study. It is important to note that these studies were published 30-50 years ago, and the different classifications of "high" SD levels varied from 21.4, to 40.1, and 55.1 kg/m^2 between studies (Proudfoot et al., 1979a; Leighton et al., 1985; Moran, 1985) which makes definitive comparisons between results difficult. Current advances in genetic selection for higher growth in turkey hens may result in different effects on feed efficiency and growth when high SD levels are used in today's commercial production systems.

In the latest published study researching the influence of SD on behavioural, health, and productivity traits of turkey hens, body weight was found to decrease as SD increased from 10 to 16 dm^2/bird (38.8 to 62.7 kg/m^2; Martrenchar et al., 1999). More recently in turkey toms, Beaulac et al. (2019) observed a decrease in body weight with increasing SD levels (30 kg/m^2 to 60 kg/m^2) over the duration of a 16-week trial. Feed efficiency decreased linearly with increasing SD and feed consumption initially increased from week 4 to 8 and then decreased from week 12 to 16 as SD increased. Bartz et al. (2020) observed a decrease in body weight and body weight gain, and a tendency for feed efficiency to worsen in toms reared to 20 weeks of age as SD increased (45 to 60 kg/m^2).

Few studies have reported a negative impact of increasing SD levels on mortality. Coleman and Leighton (1969) observed a numerical increase in mortality at high SD (48 kg/m^2), however, the differences were not significant (*P*-value not reported). Other studies found no effect of SD on total mortality (Proudfoot et al., 1979a; Leighton et al., 1985; Martrenchar et al., 1999). Contrary to these findings, Moran (1985) noted mortality of hens increased with increasing density, whereas the opposite was observed in toms. Noll et al. (1991) observed a tendency for tom mortality to be higher at high SD (60.9kg/m^2; P=0.06). In the study by Beaulac et al. (2019), there was a tendency for mortality of toms to increase from 4 to 8 weeks of age (P=0.08), however, total mortality was unaffected by SD. From the results of previous studies, it is evident that SD had a significant negative effect on body weight of both turkey hens and toms and feed efficiency in toms, however, the majority of studies indicate little to no effect of SD on mortality.

Carcass quality can directly affect economic return for producers and carcass lesions such as scratches, breast blisters, and breast buttons results in the downgrading of carcasses. Poultry carcasses in Canada are graded into three categories: Canada A grade, Canada Utility grade, and Canada C grade (Canadian Food Inspection Agency, 2014). Carcasses can be downgraded depending on the presence and size/length of blisters, scratches, cysts, skin tears, and discolouration (Canadian Food Inspection Agency, 2014) indicating that producers will incur economic losses from poor carcass quality. Canada A grade requires a normal skeletal structure, no meat interference, blisters 2.5cm in length or less, a moderately plump breast, and a specific maximum length and total area of skin tears and discolouration depending on the body part. Canada Utility grade allows a slightly crooked keel, some meat interference, blisters 2.5cm in length or less, sufficient flesh on the breast, skin tears that are less than the length of half the breast, and specific maximum areas of discolouration depending on the area of the body. Canada C grade allows an abnormal skeletal structure, a crooked keel, some meat interference, cysts, blisters, skin tears, and discolouration not exceeding 14.5cm^2. Coleman and Leighton (1968) did not observe differences in the percentage of hens with carcass grade A between SD treatments with the mean percentage of hens falling in that grade being between 98.3-99.4 % for all SD treatments. However, the authors observed a numerical decrease in the percentage of turkey toms graded A in the higher SD levels compared to the lower SD levels. The results from the study by Proudfoot et al. (1979) showed that the percentage of carcasses graded A decreased as the SD increased. Moran (1985) found similar results in turkey hens marketed at 119 days of age where the percentage of hens downgraded to Canada Utility grade increased at high SD (24.7 kg/m^2) compared to low SD (12.7 kg/m^2). Carcasses were downgraded to Utility grade due to skin tears, scratches, broken bones, bruising, and/or discolouration (Moran, 1985). Contrary to these earlier studies, Martrenchar et al. (1999) found no differences between SD treatments on hip lesions and breast blisters in turkey hens marketed at 12 weeks of age. Overall, the effects of SD levels on carcass quality either show an increase in downgrades turkey hen carcasses (Proudfoot et al., 1979a; Moran, 1985) or little to no effect on carcass quality (Coleman and Leighton, 1969; Martrenchar et al., 1999). It is evident that further research is needed to understand the effects of SD on the carcass quality of turkey hens.

1.2.2 Health Parameters

Stocking density can significantly impact bird health and welfare, and this has been well documented in broiler chickens. More studies evaluating SD in turkeys focus on production parameters with few studies that include health and welfare in addition to production. Studies assessing SD in turkeys have evaluated footpad lesions, mobility, feather condition, heterophil/lymphocyte (H/L) ratios, behaviour, and litter moisture, however, many focus on only one or two of these parameters.

Footpad Dermatitis. A major welfare concern that affects the health of turkeys is the presence of pododermatitis, also referred to as footpad lesions or footpad dermatitis (FPD). Footpad dermatitis occurs when skin discolouration begins, followed by inflammation, hyperkeratosis, and then erosions of the skin that can develop into ulcers or necrotic lesions depending on severity (Martland, 1984; Greene et al., 1985; Weber Wyneken et al., 2015). Ulceration usually occurs on the metatarsal (pulvinaris metatarsalis) and digital footpads (pulvinaris digitalis) of turkeys (Martland, 1984). Footpad dermatitis is often found as early as 8 weeks of age and most often occurs when there is high litter moisture, which may also be affected by high SD levels (Martland, 1984). Martland (1984) suggested FPD can be linked to pain and discomfort in birds. Laying hens were found to have mechanothermal nociceptors with specific fibres that cause pain in response to a stimulus on the foot which is likely present in all bird species (Gentle et al., 2001). Therefore, these pain receptors can be stimulated with the presence of FPD. Footpad lesions can also result in carcass condemnations at slaughter (Mayne et al., 2007). Footpad lesions are of concern for a number of reasons, including that they may result in a potential entry pathway for bacteria and this may lead to synovitis (inflammation of the synovial membrane) and lameness (Clark et al., 2002). To determine if FPD is painful to turkeys, Weber Wyneken et al. (2015) used a combination of objective gait scoring in turkeys with and without FPD and provided analgesic intervention or a control saline solution. The authors concluded that FPD can be painful to turkeys as the presence of FPD affected gait and behaviour. However, the authors suggested that more research with improved analgesics is required to more specifically identify the extent of pain for different footpad scores.

High litter moisture is considered the primary cause of FPD and is most often caused by improper ventilation and temperature control at high SD (Martland, 1984; Mayne et al., 2007;

Hocking et al., 2008; Rudolph, 2008). Therefore, good litter management is imperative to maintain animal welfare. Wet litter has been directly linked to the presence of FPD in turkeys (Weber Wyneken et al., 2015). Martrenchar et al. (1999) found that with increasing SD, there was increased litter moisture, which led to increased incidence of FPD. Similarly, at 16 weeks of age, turkey toms had a higher presence of footpad lesions with increasing SD and between 12-16 weeks, litter moisture demonstrated a quadratic relationship with increasing SD (Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019). It is evident that the presence of FPD in turkeys is a welfare concern and many factors such as wet litter and high SD levels can increase the prevalence of this health challenge.

Mobility. Bird mobility is an important factor that affects the health and wellbeing of turkeys (Beaulac and Schwean-Lardner, 2018). If mobility is affected, it can impact the bird's ability to escape aggressive pen mates, access feed and water, and could cause pain resulting in decreased activity which causes poor skeletal health in turkeys housed at high SD (Duncan et al., 1991; Classen et al., 1994; McGeown et al., 1999; Jankowski et al., 2015; Beaulac and Schwean-Lardner, 2018). Poor gait scores, which can be an indicator of bird mobility, have also been linked to the presence of footpad lesions (Martrenchar et al., 1999; Weber Wyneken et al., 2015). Martrenchar et al. (1999) evaluated the effects of SD on the mobility of turkey hens and toms using a subjective gait score procedure. The results of this study demonstrated that turkey hens housed at higher SD levels, within the range of 38.8 to 62.7 kg/m^2, showed poorer gait at 12 weeks of age, and turkey toms had poorer gait scores at high SD at 16 weeks of age (Martrenchar et al., 1999). More recently, it was found that turkey toms housed at up to 60 kg/m^2 demonstrated decreased mobility at 16 weeks of age (Beaulac and Schwean-Lardner, 2018).

Feather cover. Feather cover may be an indicator of bird welfare when poor feather cover is caused by feather pecking that damages or removes feathers or when increased wear occurs (Beaulac and Schwean-Lardner, 2018). Feather cover may also be an important indicator of live market bird quality and is important for thermoregulation and as protection from scratches and skin lesions. Though feather cover is not indicative of the overall health of the bird, Coleman and Leighton (1968) found that feather cover decreased under higher SD levels for 14-week old turkey hens. Beaulac and Schwean-Lardner (2018) found that increasing SD levels negatively impacted feather cover and cleanliness when turkeys were housed between 30 and 60 kg/m^2. Contrary to these findings, studies on the effects of SD and beak trimming on the behaviour and growth of turkey hens and toms found no effects of SD on feather scores or carcass quality (Denbow et al., 1984; Leighton et al., 1985). It has been suggested that the presence of breast blisters and buttons have been associated with poor feather cover and increased contact with wet litter (Newberry, 1995), however, Martrenchar et al. (1999) found that this did not occur at the highest density tested in their experiment despite increased contact with wet litter resulting from reduced mobility of the birds.

Heterophil/lymphocyte ratio. Heterophil/lymphocyte ratios have been verified as a quantitative and reliable method of evaluating bird wellbeing, specifically stress related to social and environmental stressors (Gross and Siegel, 1983; McFarlane and Curtis, 1989). When birds are exposed to a stressor, the hypothalamic-pituitary-adrenal (HPA) axis is activated which causes the hypothalamus and pituitary to secrete corticotropin-releasing hormone (CRH) and adrenocorticotropic hormone (ACTH), respectively. The release of ACTH stimulates the synthesis and release of the glucocorticoid hormone, corticosterone, which results in increased plasma corticosterone concentrations (He et al., 2018). Corticosterone concentration in the blood is often used as a measure of acute stress, and when plasma corticosterone increased in laying hens, the number of lymphocytes in the blood decreased and the number of heterophils increased (Gross and Siegel, 1983). Heterophils, comparable to mammalian neutrophils, respond and recognize pathogens and are effective in guiding the avian innate immune response through anti-microbial functions such as phagocytosis, degranulation, and oxidative burst. (Genovese et al., 2013). Lymphocytes are white blood cells that produce antibodies, cytokines, and macrophages in an immune response. Heterophil/lymphocyte ratios are a more reliable indicator of stress levels in response to chronic and social stress as it indicates a shift in immune function compared to measuring plasma corticosteroid levels (Gross and Siegel, 1983; McFarlane and Curtis, 1989).

According to Genovese et al. (2013), specific heterophil functions and immune responses can be related to commercial poultry genetic selection and differences were noted in broiler chicken lines. Heterophil function was found to be more efficient in wild-type turkeys compared to commercial heavy turkeys in its anti-microbial function which may indicate that selection for specific performance parameters in commercial turkeys could adversely affect immune response (Genovese et al., 2006). Thus, using H/L ratios as an indicator of stress may differ between poultry species and within different commercial lines of a poultry species. Different stressors may then affect how the H/L immune response acts; thus, H/L ratios should not be the only measurement of stress. In this study, both H/L ratios and behaviour will be used to determine stress responses to SD.

To date, only two studies have assessed the effects of increasing SD levels on H/L ratios in turkeys. Hafez et al. (2016) evaluated the effects of three SD levels (25, 48, or 58 kg/m^2) on the H/L ratio in turkeys, however, no differences were observed. More recently, Beaulac and Schwean-Lardner (2018) reported that H/L ratios increased with increasing SD (30 to 60 kg/m^2) in toms at 4 weeks of age and a linear trend was observed at 12 weeks of age. However, no differences were observed at 16 weeks of age. The increase in H/L ratios corresponded with numerical increases in aggressive damage between day 0 and week 4 and increases in aggressive behaviour from 4 to 8 weeks of age with the highest incidence of aggressive damage occurring in the 30 and 60 kg/m^2 treatments. Mortality and culls linked to aggression also increased with higher SD and the authors suggested that the increasing H/L ratio was likely an indicator of stress associated with high SD levels (Beaulac and Schwean-Lardner, 2018). A previous study reported that with increasing SD levels in broiler chickens, there were linear increases in H/L ratios, however, the authors argued that the increase in H/L ratios were slight and if caused by stress, other parameters measured would have been indicative of this (Thaxton et al., 2006). The authors also stated that the increase in H/L ratios may have been due to exposure to various microbes, therefore, they concluded that increasing SD did not cause physiological adaptive changes due to stress in broiler chickens (Thaxton et al., 2006).

1.2.3 Behaviour

Bird behaviour is an essential parameter to assess when evaluating the welfare of turkeys housed at varying SD levels in commercial production. Domestic turkeys raised in intensive production systems often express negative behaviours that result in or arise from poor bird welfare. Disturbances of resting birds, feather pecking, and aggression in turkeys are some of the most commonly evaluated activities in current literature (Erasmus, 2017). Other behaviours that have been studied in relation to stocking density include nutritive, mobility, and comfort behaviours such as preening and dustbathing.

Aggression or aggressive pecking behaviour is a significant welfare and economic concern for commercially raised turkeys, especially when evaluating density. Aggression can lead to mortality as aggressive behaviour is often directed to the head of the turkey. Aggressive pecking can be defined as misplaced pecking activity directed toward conspecifics and occurs when there is repeated pecking at the head or body of a bird which leads to the removal of feathers or skin (Sherwin et al., 1999; Duggan et al., 2014). Production can be compromised if aggression occurs as affected birds will have reduced feather cover resulting in increased heat loss, higher rates of mortality and culls, and additional feed intake to maintain thermoregulation (Duggan et al., 2014). Thus, this problematic behaviour can lead to economic losses. Aggression that causes skin damage and flesh exposure gives rise to other welfare concerning behaviours such as cannibalism in which birds will repeatedly peck at the exposed skin of another bird resulting in the removal of blood and tissue which is then consumed by the bird (Savory, 1995; Dalton et al., 2013). Aggressive pecking that is directed toward the head, neck, or snood of another turkey may be caused by a social disturbance and can be a manner of asserting dominance (Moinard et al., 2001; Buchwalder and Huber-Eicher, 2004; Dalton et al., 2013). This can be increasingly problematic for turkeys raised at high SD with large group sizes as they have competitive social systems where dominance is established through aggressive encounters (Buchwalder and Huber-Eicher, 2004). It has been suggested that in large group sizes of birds raised at high SD in commercial production, birds may continually attempt to establish dominance (resulting in more aggression) but are unsuccessful because individual recognition of conspecifics is not possible in large group sizes (Hughes et al., 1997; Buchwalder and Huber-Eicher, 2003).

Buchwalder and Huber-Eicher (2004) examined the effects of floor space on aggressive behaviour in turkey toms housed in groups of five birds in two different pen sizes (small pen size of 2x3 m and large pen size of 6x13 m). The toms' behaviour toward an introduced unfamiliar conspecific was examined in each pen size. The results showed that the more floor space the turkeys had, the less aggressive pecks resulted. This may be due to the target bird's ability to escape from aggressive pen mates. The authors of this study also found that there may be a critical distance at which aggressive behaviour is avoided. They could not determine exactly how much space birds needed to retreat and suggested for 13-week old turkey toms it could be between 1 bird/m^2 to 0.08 bird/m^2 (Buchwalder and Huber-Eicher, 2004). Beaulac and Schwean-Lardner (2018) assessed aggressive behaviour in turkey toms housed at increasing SD (30 to 60 kg/m^2) and found that the total incidence of aggression was closely related to the increase in aggression related mortality for birds housed at higher SD. At 16 weeks of age, aggressive pecking linearly increased with increasing SD in toms. During weeks 4 to 8 of the study, the total cases of aggressive damage was highest in the 30 kg/m^2 and 60 kg/m^2 SD treatments. In contrast to these results, Denbow et al. (1984) and Martrenchar et al., (1999) observed no increase in aggression with increasing SD levels in turkey toms.

Nutritive behaviours, such as feeding and drinking, are highly motivated and controlled internally through neurons and hormones that influence the bird's behaviour (Duncan, 1998). All husbandry systems allow feeding and drinking as they are essential to growth and wellbeing, however, if a decrease in these behaviours occur, it may indicate reduced welfare caused by environmental or social factors (Dawkins, 1990; Duncan, 1998). Broiler chickens exhibit synchronized feeding from social facilitation in which they will move to a feeder and stay for longer bouts when conspecifics are present at the feeder (Collins and Sumpter, 2007). In a review by Dawkins (2018), it was suggested that sufficient space at feeders and drinkers is essential to ensure birds are feeding without disruption and are not avoiding feeding near dominant conspecifics. However, reduced feeding behaviour may not be indicative of poor welfare as it may decline at specific stages of the birds' life or be dependent on diet changes in the rearing cycle (Dawkins, 1990; Hughes and Grigor, 1996). Drinking behaviour is thought to be closely related to its natural function of satisfying thirst and only performed when needed (Duncan, 1998). Ross and Hurnik (1983) observed broiler drinking behaviour and noted that it changed with age as many shorter bouts became fewer and for a longer duration.

Mobility behaviours (standing or walking) or locomotion can be indicative of skeletal health, therefore, they are important behaviours to evaluate when determining bird health and wellbeing. It has been suggested that broiler leg health worsened at high SD as a result of decreased activity and high SD can negatively influence skeletal development observed through increased leg culls (Hall, 2001). Reductions in bird activity from a lack of space have been hypothesized to cause poorer gait in turkeys raised at high SD (Martrenchar et al., 1999). Decreases in walking could be a welfare concern as birds may not access feed and water which can lead to lower body weights, dehydration, or more culling in extreme cases (McGeown et al., 1999; Beaulac and Schwean-Lardner, 2018).

Comfort behaviours, including preening, dust bathing, wing flapping, and stretching, can be associated with a positive affective state (Sherwin and Kelland, 1998). Preening, dustbathing and feather ruffling are behaviours performed to maintain feather condition and remove any parasites on their skin (Appleby et al., 2004). The frequency, form, and function of comfort behaviours may be related to space allowance as these behaviours require more space to be performed (Appleby et al., 2004), however, when these behaviours are performed at higher stocking densities, it may result in the disturbance of other birds. It is important to note that some comfort behaviours such as preening can be performed as a displacement behaviour as a result of stress or frustration (Duncan and Wood-Gush, 1972; Appleby et al., 2004). For example, displacement preening in laying hens deprived of feed were described as "frantic" and occurred in the more accessible areas of the body (Duncan and Wood-Gush, 1972). Dust bathing is also performed to maintain plumage and is defined as the fluttering movement of the bird on a loose substrate while pulling the loose substrate close to the body and into the feathers to aid in distributing and removing oily secretions (Appleby et al., 2004). There is evidence that dust bathing is a highly motivated behaviour in poultry as they will dustbathe even when a loose substrate is not provided (such as in laying hen cages with wire or slat flooring) and this is considered a vacuum behaviour (Lindberg and Nicol, 1997). However, in commercial turkey houses with litter flooring, dust bathing can be performed as a comfort behaviour with adequate space allowance. Wing flapping and stretching are comfort movements important for plumage maintenance (Appleby et al., 2004) and the maintenance of functional joints and muscles and increasing bone strength (Hurnik et al., 1985; Duncan, 1998).

With only a few studies evaluating SD effects on the behaviour of turkeys, Martrenchar et al. (1999) found that SD had little effect on turkey activity but the frequency of disturbance of resting birds by other birds was higher at higher SD levels. No differences were observed for mobility behaviours or aggressive behaviour between turkeys housed at varying SD treatments. Alternatively, Gunther and Bessei (2006) found that mobility decreased with increasing SD (2.5, 3.0, and 3.5 birds/m^2; body weight not reported) in turkey hens and toms. The authors found that sitting/laying, preening, and feather pecking frequency were influenced by SD. Sitting/laying frequency was the highest at low SD which may be because these birds were less disturbed by other birds. For preening, the highest frequency occurred at the lowest SD levels which allowed for longer resting periods. The frequency of dustbathing observed in this study was higher in hens than toms, however, no reason was provided. However, for behaviours such as feeding, drinking, dustbathing, standing, and ground pecking, there were no differences in relation to SD. In a more recent study of SD effects in turkey toms, the results indicated there was a decrease in bird activity as SD increased, likely due to lack of floor space (Beaulac and Schwean-Lardner, 2018). The greater space allowance in the low SD treatment allowed for a larger percentage of birds to be more mobile as seen in a higher percentage of birds walking. The behaviour analyses also revealed that birds housed at moderate SD rested more frequently. This suggests that birds housed at the highest SD (60 kg/m^2) did not have enough space to rest as more birds were observed standing compared to resting in that treatment (Beaulac and Schwean-Lardner, 2018). The authors also observed a decrease in the percentage of birds present at the feeder and drinker at 16-weeks of age at high SD and this correlated to the decrease in body weight observed at that age. The percentage of birds preening increased with increasing density at 14 and 16 weeks of age and comfort behaviours increased as SD increased at 16 weeks of age. With only a few studies focusing on behavioural and health traits of turkeys, and more focus placed on production parameters, there is a growing area of interest and need for behaviour in relation to SD to be further studied in turkey hens.

1.3 Stocking Density Effects in Broiler Chickens

As broiler chicken production is the largest poultry meat industry, the effects of SD on performance, health, and behaviour have been studied to a greater extent. Because turkey information regarding stocking density is limited, it is important to review the effects of SD on broiler production and welfare. However, there may be differences in broiler and turkey responses to the effects of SD as broilers are reared for shorter periods and there may be species or sex differences. To maximize profits from meat yield, producers increase the number of broilers within a specific space to reduce costs associated with housing, labour, fuel, and equipment (Cravener et al., 1992). Therefore, up to a specific point, profitability can increase with increases in SD (Puron et al., 1995). However, previous studies have aided in developing industry guidelines that allow for sufficient economic return without compromising the welfare of the birds. The Canadian Codes of Practice for the care and handling of hatching eggs, breeders, chickens, and turkeys require broilers to be housed at a density that does not exceed 31 kg/m^2 and may only be increased to a maximum of 38 kg/m^2 when strict environmental management practices are in place (National Farm Animal Care Council, 2016). Therefore, effective performance and welfare are dependent on the multifaceted interactions between SD, temperature, ventilation, litter quality, and other management practices.

1.3.1 Broiler Performance Parameters

Performance parameters used to measure the effects of SD in broilers are the same as those for turkeys and include body weight, feed efficiency, mortality, and carcass quality. Across the majority of studies, body weight decreases as SD increases, with SD being defined as high within the range of 37.9 to 56.0 kg/m^2 (Proudfoot et al., 1979b; Cravener et al., 1992; Martrenchar et al., 1997; Sørensen et al., 2000; Heckert et al., 2002; Dawkins et al., 2004; Dozier et al., 2005; Goo et al., 2019; Li et al., 2019). In a more recent study on the effect of SD and sex on performance in broilers, no interactions between sex and SD for growth performance were found (Goo et al., 2019). Dozier et al. (2005), observed an improvement in feed efficiency when SD increased from 25 to 38 kg/m^2 from 0-17 days of age, however, reduced growth, feed consumption, and poorer feed efficiency was observed from 0-49 days of age. Similar results have been noted in later studies where decreased body weight gain and feed intake were observed with increasing density (Cengiz et al., 2015; Goo et al., 2019). When birds were raised at higher SD, there may be a behavioural element that affects performance parameters in which birds have more difficulty accessing feeders and drinkers because of reduced activity (Sørensen et al., 2000; Cengiz et al., 2015). Although not mentioned, another factor affecting the reduction in performance at high SD may be the birds' physical ability to reach the feeders and drinkers.

Flock uniformity is inversely related to the coefficient of variation (CV) of body weight, another indicator of bird performance. Martrenchar et al. (1997) noted poorer uniformity in male body weights compared to females at high SD levels. A study by Feddes et al. (2002) found that the CV for body weight was higher in the lowest SD treatment of 11.9 birds/m^2 (22.79 kg/m^2) compared to the highest density of 23.8 birds/m^2 (45.17 kg/m^2), and no difference in feed consumption was found between treatments. The authors suggested that the variability in body weights resulted from increased floor space which allowed fast-growing birds to grow to their full potential, whereas the decreased overall body weight seen in the highest SD treatment resulted from the close proximity of birds during growth (Feddes et al., 2002).

Mortality can negatively impact economic return for producers, and high mortality can be indicative of welfare issues. However, some studies have reported no differences in mortality between treatments of varying densities (Proudfoot et al., 1979b; Dozier et al., 2005; Buijs et al., 2009). Contrary to these findings, Bilgili and Hess (1995) observed higher mortality in male broilers reared at high SD levels (35.2 kg/m^2) during day 1-21, however, no effects were observed at the end of cycle (day 42-49). Numerical increases following the same periods for mortality were observed in female broilers. The lack of statistically significant results for mortality data in relation to SD may be a result of low mortality rates or variability between SD treatments (Beaulac et al., 2019).

Another potential economic loss and welfare concern is the downgrading of carcasses at slaughter. Proudfoot et al. (1979b) found that as SD increased, the percentage of Grade A carcasses (male) decreased in an "erratic manner" as linear, quadratic, and cubic differences were noted, and the percentage of Grade A carcasses (female) decreased linearly as SD increased. The carcass downgrades in this study were caused by breast blisters, which are a type of contact dermatitis that can be caused by increased contact with wet litter. Breast lesions are composed of exudated protein and necrotic inflammatory cells that can sometimes be covered by cornified strands of keratin (Mayne, 2005). In comparison, Bilgili and Hess (1995) observed an increase in thigh sores and scabs at high SD for broiler males, however, no differences were observed for other carcass defects nor the percentage of U.S. Grade A carcasses. Conversely, the incidence of skin lesions (sores/scabs and scratches) was higher in females housed at high SD and the percentage of U.S. Grade A carcasses decreased in the highest density treatment. Although not mentioned, the higher incidence of scratches and scabs at high SD may have been a result of birds walking on top of each other when there was reduced floor space. Male broilers in this study also exhibited a decrease in breast fillet weight as a percentage of live weight at higher SD which can lead to significant economic losses (Bilgili and Hess, 1995).

1.3.2 Broiler Health Parameters

The effects of SD on the health of broilers have been studied by looking at FPD, gait scores as an indicator of mobility, leg health, and feather condition and cleanliness. These parameters can be useful indicators of bird welfare to determine if the bird is in pain or discomfort. Previous research has provided sufficient evidence that higher SD caused increased incidence of FPD and hock burns (Cravener et al., 1992; Martrenchar et al., 1997; Sørensen et al., 2000; Dozier et al., 2005; Buijs et al., 2009; Knierim, 2013; Petek et al., 2014).

Gait score is a critical display of bird welfare because it is associated with the birds' mobility and ability to access their basic needs such as feed and water. This is evident in the study by Sørensen et al. (2000) in which broiler mobility decreased with age at higher SD levels and the birds with poorer gait had a decreased body weight; the authors suggested the birds struggled to access feed. As broilers are considered juvenile animals that are still undergoing bone development through the rearing cycle, they can be more susceptible to poor bone development and quality which can lead to fractures and reduced welfare (Buijs et al., 2012). In the study by Buijs et al. (2012), the authors observed a decrease in tibia strength and an increase in tibia curvature with increasing SD. Tibia curvature may cause lameness in broilers and can lead to secondary health issues such as hock burns, hock joint dislocation, and fractures (Buijs et al., 2012). Tibia strength does not often cause problems in broilers during rearing but can increase the risk of fractures during catching and transportation which can affect the birds' welfare (Julian, 1998). Buijs et al. (2012) suggested that the decrease in tibia strength and increased curvature at high SD may have resulted from the birds' inability to rest properly thus affected skeletal development. The evidence presented in these studies provides valuable information on potential welfare problems associated with high SD in broiler production.

Feather condition and cleanliness of broilers has only been assessed in relation to SD in a few studies. When evaluating the feather cover of the breast in broilers, Thomas et al. (2004) observed poorer feather cover with increasing SD (9.64 to 35.79 kg/m^2) and this may be caused by increased contact with wet litter. In an earlier study by Proudfoot et al. (1979b), the authors examined feather cover and found that birds in the high SD treatment had "rough and tattered" feathers (not statistically analyzed) and this correlated to a high incidence of breast blisters seen in that treatment. The authors also observed damp and packed litter quality in the high SD treatment which may have exacerbated these conditions.

1.3.3 Broiler Behaviour

Various methods have been used to evaluate the behaviour of broilers in relation to SD. Behaviours that are observed include bird disturbance, resting, locomotor behaviours (walking, standing), comfort behaviours, and feeding and drinking. Disturbance of resting birds, as seen with turkeys, and birds climbing over each other have been widely recorded as a negative behavioural outcome of higher SD (Dawkins et al., 2004; Buijs et al., 2009, 2011b; Thomas et al., 2011; Knierim, 2013; Dawkins, 2018). When birds have restricted floor space and they climb over one another, there is an increased possibility for scratching and damage to the skin (Thomas et al., 2004; Estevez, 2007; Dawkins, 2018) which can lead to poor carcass quality and wellbeing. Locomotor behaviours and ground pecking decreased as SD increased (Martrenchar et al., 1997; Hall, 2001), however, no effects were observed for resting (lying bouts) in relation to density (Martrenchar et al., 1997). Broilers have a decreased probability of standing at the feeder and drinker with increased SD (28.7 compared to 14.5 kg/m^2) (Simitzis et al., 2012) which may be an additional cause of lower body weight seen in broilers at high SD. To the best of the author's knowledge, preference tests for different SD levels have not been studied in turkeys. However, one study used a preference test for broilers in which the birds could choose between three SD treatments (23.3, 31.5, and 40.0 kg/m^2) by crossing two barrier heights (high and low), thus, the birds with a moderate to high motivation will work to cross the barrier to reach their desired density (Buijs et al., 2011a). The results found that broilers preferred the low and middle SD of 23.3 and 31.5 kg/m^2, respectively, compared to the highest SD. The authors also compared the willingness of broilers to cross the high and low barriers to reach feed after being feed deprived for 6-hours. The results show that the birds were less motivated to reach feed (20-25% of the birds did not cross the high barrier to access feed after being feed deprived) compared to the birds' motivation to reach lower densities in which more birds crossed both the high and low barriers to access lower densities. This suggests that birds are willing and motivated to work for increased space allowance.

1.4 External Environmental Stressors in Turkeys

Turkey welfare can be directly affected by high levels of stress which may make them more susceptible to illness. Previous studies have examined the disease susceptibility of turkeys exposed to different stressors and how various commercial lines and different sexes respond differently to stress (Kowalski et al., 2002; Huff et al., 2007). Kowalski et al. (2002) studied the differences in stress responses between turkeys of two commercial lines (one fast-growing and one slow-growing genetic strain). Their results indicate that corticosterone levels were higher in the fast-growing strain compared to the slow-growing strain when exposed to transport stress. Huff et al. (2007) concluded that transport stress was higher in fast-growing turkeys due to a blunted HPA axis. A blunted HPA axis occurs during high levels of stress when increased corticosterone levels (following the regular HPA axis release of corticosterone) inhibits the HPA axis from producing more corticosterone (Ayer et al., 2013). A higher stress response of fast-growing male turkeys is said to cause immunosuppression leading to increased susceptibility to bacterial infection, hence, it was suggested that genetic selection for fast growth has unintentionally selected for a stress response that may be incompatible with the external stressors associated with commercial turkey production (Huff et al., 2007). In contrast, Huff et al. (2007) did not observe the same stress response in female turkeys. The altered immune response caused by stress can be influenced by environmental conditions and social stress, therefore, when studying the effects of SD on turkeys it is crucial to understand and control the effects of environmental factors.

In commercial turkey housing, air quality is considered a function of the concentration of ambient airborne contaminants; these include ammonia (NH_3), carbon dioxide (CO_2), hydrogen sulphide (H_2S), and respirable airborne dust particles (Feddes and Licsko, 1993). Ammonia, a water-soluble gas by-product, and CO_2 occur as a result of microbial decomposition of organic nitrogen compounds in poultry manure (Ritz et al., 2004), however, the majority of CO_2 occurs as a by-product of respiration when birds exhale. Ammonia affects the health of poultry as its water-soluble characteristic allows it to be dissolved in the moist mucous membranes and eyes of birds, and it is associated with respirable dust particles (Wathes et al., 1997; Ritz et al., 2004). Ammonia poisoning in poultry can cause nasal snicking (hacking sound made as birds try to clear the upper respiratory tract), tracheal irritation, air sac inflammation, conjunctivitis, and dyspnea (shortness of breath) (Carlile, 1984). Respirable dust particles can contain pathogens for disease transmission such as Marek's Disease making it easily spreadable between poultry flocks (Woźniakowski and Samorek-Salamonowicz, 2014). Respirable poultry dust particles are also an occupational health risk to workers and were found to cause asthma, allergic alveolitis, respiratory inflammation, and chronic obstructive pulmonary disease in humans (Viegas et al., 2013).

Previous literature has suggested NH_3 should not exceed 25 ppm (parts per million) (Carlile, 1984), however, it was found that bird welfare and health can be compromised at NH_3 levels as low as 10 ppm (Nagaraja et al., 1983; Schwean-Lardner et al., 2013). Therefore, the current Canadian Codes of Practice recommend that management practices – such as increased ventilation, evaluation of SD, and minimization of water spillage – should be implemented if

NH_3 levels exceed 10ppm and action must be taken if NH_3 reaches a harmful range of 20-25 ppm (National Farm Animal Care Council, 2016). In a study on the impact of ventilation rate and SD on bird health and performance, the results showed a trend for respirable and non-respirable dust concentrations to be affected by SD, and lung lesions tended to be more severe in high SD treatments (Zuidhof et al., 1993). The study also indicated that low ventilation resulted in significantly higher NH_3 and CO_2 levels. To the best of the author's knowledge, there are no specific guidelines on acceptable CO_2 levels in poultry houses. Carbon dioxide and carbon monoxide (CO) can affect poultry health and welfare as toxic CO can have lethal effects in a poorly ventilated barn (National Farm Animal Care Council, 2016). Litter quality, air quality, and ventilation are affected by SD as higher densities of birds will produce more fecal matter which will affect all three factors (Erasmus, 2018).

Dawkins et al. (2004) found that high concentrations of NH_3 and high litter moisture caused increased fecal corticosteroid concentration in broiler chickens, indicative of stress. The difference in corticosteroid levels in broilers was suggested to be affected by temperature, humidity, season, and ventilation type (Dawkins et al., 2004). The authors also observed the effects of litter moisture (a vital attribute of litter quality) and found that heater position and number of drinkers per thousand birds was directly related to variations in litter moisture. Mayne (2005) suggested that correct temperature set points and humidity levels may prevent condensation and lower SD levels can aid in reducing litter moisture. High litter moisture may be one of the primary causes of footpad dermatitis (Mayne et al., 2007), and can in turn be a factor in high NH_3 levels. Higher NH_3 levels (up to 29.8 ppm) in combination with wet litter have been correlated to dirtier footpads, increased hock lesions, and poor leg angulations in broilers, whereas better mobility and reduced footpad lesions were observed in summer due to low relative humidity and increased ventilation (Dawkins et al., 2004). The authors concluded that certain aspects of leg health, such as gait score, were negatively affected at or above a SD of 42 kg/m^2, however, the environmental conditions of broiler chicken rearing had a greater impact on bird health and welfare than SD (Dawkins et al., 2004). It is well documented that environmental stressors can negatively impact the health and welfare of poultry species, thus, when evaluating the true effects of SD, it is important to minimize any environmental stressors that can be easily controlled through management practices.

1.5 Conclusions

It is evident from previous studies that high SD can have negative effects on the productivity, health, and welfare of turkey hens. From an economic and welfare standpoint, evaluating these effects are vital with the increase in consumer demand for better welfare of livestock. However, it must be noted that that the effects presented can be impacted by confounding factors such as litter moisture, air quality, and management practices. From the current literature on SD and its effects on poultry, it is evident that there is a complex relationship between SD and welfare. Thus, identifying the most suitable density for turkeys requires a careful analysis of behavioural requirements and potential health risks caused by environmental stressors and how these factors will impact productivity and welfare.

1.6 Objectives

The primary objective of this study is to evaluate the impact of graded levels of stocking density on turkey hen performance, health, and welfare up to 11 weeks of age. The performance parameters that will be evaluated include growth, feed consumption, feed efficiency, mortality and morbidity, and flock uniformity. To determine the effects of SD on bird health, mobility (gait score), footpad lesion severity (scores), feather condition and cleanliness (scores), litter moisture, and heterophil/lymphocyte ratio will be assessed. Behavioural observations will be made to further determine SD effects on turkey hen welfare. By statistically analyzing the relationship among these parameters in relation to SD, peaks in performance, health, and welfare will be determined when analysing the effects of graded levels of SD to determine optimal SD. A second objective will be to conduct a basic economic analysis utilizing 2019 and 2020 poult costs, feed costs, number of birds shipped, and final income based on the various stocking densities. The data from this study will be valuable for developing industry guidelines as it is more current and will consider all three elements of performance, health and welfare of the birds.

1.7 Hypotheses

High SD levels in turkey hen production will negatively impact performance parameters and bird welfare due to increased chronic stress and poorer health parameters. With increased SD levels:

- There will be a negative impact on production parameters, such as growth, feed consumption, feed efficiency, and flock uniformity, due to increased stress and reduced space allowance.
- There will be a negative impact on the health of the turkey hens resulting from increased incidence of FPD, poorer mobility and gait, and poor feather coverage and cleanliness. This will occur because at higher SD there will be increased litter moisture and less available space for mobility behaviours such as walking to occur.
- Decreased activity and increased aggression will occur due to more birds being disturbed by other birds, and this will lead to an increase in aggression related mortality.

By increasing space allowance at low SD levels, there will be an alteration in the birds' behavioural expressions. Low SD levels will result in:

- The expression of comfort behaviours as there is more space for this to occur, however, there will be higher aggression seen at low SD levels due to more birds being disturbed by other birds.

2.0 Chapter 2: Evaluating the effects of stocking density on the performance of turkey hens to 11 weeks of age

The objectives of this study were to assess the factors that can be affected by increasing SD on turkey hens to 11 weeks of age. Chapter 2 encompasses the effects of SD on turkey hen performance to 11 weeks of age by evaluating body weight, feed consumption, feed efficiency, mortality, and uniformity. An economic analysis was also performed to determine the effect of SD on net income.

2.1 Abstract

Stocking density (SD) affects economic return for turkey production and can impact production parameters. Most studies have focused on the effects of SD on toms rather than hens; thus, a gap exists in the literature. This study (two blocks) evaluated the impacts of SD on turkey hen performance to 11 weeks of age. Nicholas Select hens (n=3550 poults/block) were randomly placed in one of four final estimated SD treatments (at 11 weeks of age) of 30, 40, 50, or 60 kg/m^2 in open rooms ($67.5m^2$) with four replications per treatment. Feeder and drinker space were equalized on a per bird basis. Air quality (carbon dioxide and ammonia) was measured and ventilation was adjusted to equalize ammonia and carbon dioxide (CO_2) levels across all rooms/treatments. Group body weight (BW) and feed consumption were measured on day 0 and week 3, 5, 8, and 11. Body weight gain and feed-to-gain ratio were calculated. Mortality and culls were recorded daily and necropsied for cause of death. At week 8 and 11 flock uniformity was evaluated (30 birds/replicate). Data were analyzed using regression analysis in SAS 9.4 (Proc Reg for linear regression and Proc RSReg for quadratic regression; SD as independent variable). An analysis of variance was performed for all data (Proc Mixed; SAS 9.4) and a Tukey's range test was used to separate means. Differences were considered significant when $P \leq 0.05$. All values reported are in ascending order of SD treatments 30, 40, 50, and 60 kg/m^2. Air quality parameters of CO_2 (P=0.70 and P=0.24 for block 1 and 2, respectively) and ammonia (P=0.11 and P=0.32 for block 1 and 2, respectively) were consistent across treatments for both blocks. At week 11, BW decreased linearly as SD increased (8.36, 8.35, 8.30, 8.19 kg; P=0.05). There was a tendency for overall BW gain to linearly decrease as SD increased (P=0.06). Feed consumption decreased linearly as SD increased during week 8-11 (P<0.01) and quadratically as SD increased over the 0–11-week period (15.19, 15.29, 15.08, 14.90 kg; P=0.04). SD had no impact on feed-to-gain ratio, percent mortality, or uniformity. Total aggression related mortality and culls were highest in the 30 kg/m^2 treatment (linear; P=0.02). An economic analysis was performed utilizing commercial poult and feed costs (obtained in 2019 and 2020) and income at marketing, which demonstrated that net room income increased as SD increased (linear; P<0.01). The results indicate that high SD negatively impacted turkey hen final BW and feed consumption, but no effect was observed on feed-to-gain ratio, percent mortality, or uniformity.

Keywords: body weight, feed consumption, mortality, air quality

2.2 Introduction

Stocking density (SD) has many effects on turkey production parameters, including performance, health, welfare, and producer profitability. Stocking density can have adverse effects on turkey performance and the majority of studies have examined performance parameters such as body weight (BW), feed efficiency, and mortality. Though there has been more extensive research with broiler chickens, the available literature focusing on turkeys found that as SD increased, BW of toms and hens decreased (Coleman and Leighton, 1969; Proudfoot et al., 1979a; Leighton et al., 1985; Moran, 1985; Noll et al., 1991; Martrenchar et al., 1999; Beaulac et al., 2019; Bartz et al., 2020). However, the effects on feed efficiency varied between studies. Some studies found that increasing SD caused poorer feed efficiency (Leighton et al., 1985; Noll et al., 1991; Beaulac et al., 2019; Bartz et al., 2020) and others found no effects on feed efficiency (Coleman and Leighton, 1969; Proudfoot et al., 1979a). Stocking density has shown little negative effects on mortality of turkeys in previous literature, with only numerical differences or tendencies ($P<0.06$) for higher mortality rates with increasing SD observed in the studies by Coleman and Leighton (1969) and Noll et al. (1991), respectively.

Poorer performance in birds reared at higher densities can result from higher stress or poor environmental conditions such as air quality and litter moisture. Litter and air quality are affected by higher SD as more birds produce larger quantities of fecal matter, which contribute to wet litter and poor air quality. Ammonia is released as a result of microbial decomposition of poultry manure (Ritz et al., 2004) and CO_2 levels can also be directly related to SD due to increased respiratory output (Zuidhof et al., 1993). Ammonia levels exceeding 10 ppm affect the health of poultry (Nagaraja et al., 1983; Schwean-Lardner et al., 2013) and are associated with respirable dust particles (Wathes et al., 1997; Ritz et al., 2004). Thus, poor air quality can cause increased stress and poorer health which negatively affects bird performance. The effects of litter and air quality on birds are important confounding factors to consider when evaluating the true effects of SD.

The majority of studies assessing SD in turkeys have focused on turkey toms and the most current literature on the effects of SD in turkey hens was published 22 years ago. Though these studies provided valuable information, there have been many advances in genetic selection for improved growth rate and feed efficiency since then. The gaps in the literature for assessing SD effects on the performance and health of turkey hens demonstrates the importance of SD guidelines that are based on more current literature.

The objectives of this chapter were to evaluate the impact of graded levels of SD on turkey hen performance to 11 weeks of age to assist in improving SD guidelines for commercial turkey production. To ensure that the effects of SD were measured, and not differences in air quality that resulted from varying SD, the study also focused on reducing these confounding factors by controlling air quality, and equalizing feeder and drinker space between treatments. Finally, the chapter provides an economic analysis of the graded levels of SD used in this study. It was hypothesized that high SD levels would negatively affect BW, feed efficiency, mortality, and flock uniformity due to increased stressors and reduced space allowance. It was also hypothesized that economic returns would increase with increasing SD.

2.3 Materials and Methods

The experimental procedures for this experiment were approved by the University of Saskatchewan Animal Care Committee and all birds were cared for as specified in the Guide to the Care and Use of Experimental Animals by the Canadian Council of Animal Care (2009).

2.3.1 Experimental Design

Research on the effects of graded levels of stocking density on turkey hen productivity, health, and welfare was comprised of two blocks, starting in January 2019 and November 2019, to allow for increased replication (four replications per treatment). The study was conducted at the University of Saskatchewan Poultry Centre in a floor housing facility that includes separate, and independently controlled rooms for environmental parameters, allowing for appropriate replication of this study. The four targeted final estimated stocking densities (calculated for birds at 11 weeks) tested were 30, 40, 50, and 60 kg/m^2, and production parameters were evaluated from placement to 11 weeks of age.

2.3.2 Birds and Housing

A total of 3,550 Nicholas Select turkey hens were placed in each block of this study. The birds were obtained from a commercial hatchery, where they were beak and toe treated. The birds were randomly selected and placed in one of the four stocking density treatments. An additional 5% of birds were placed to account for predicted mortality, in an effort to allow the final target stocking densities to be reached at 11 weeks of age. The number of birds placed in each treatment was calculated according to the final predicted BW of turkey hens at 11 weeks of age (Aviagen, 2015a). The number of poults placed was 295, 388, 482, and 571 per room for the final predicted SD treatments of 30, 40, 50, and 60 kg/m^2, respectively.

Birds were housed in large open rooms (6.7 x 10.0m = 67.5m^2), with each treatment being replicated twice, resulting in two room replications per treatment per block. Birds were brooded on wood shavings 7-10 cm thick, then wheat straw (10-13 cm depth) for the rearing period. Brooder rings, 7.0 m in diameter, and heat lamps were used for the first 10 days. Feed was provided *ad libitum* using aluminum tube feeders with a pan diameter of 36 cm for the first 40 d and a large pan diameter of 44 cm for the remaining time. Water was accessed through Lubing EasyLine™ pendulum turkey nipple drinkers (Lubing, Cleveland, TN). Feeder and drinker space were equalized on a per bird basis for each SD treatment (35 birds/feeder; 30 birds/nipple), thereby reducing impacts of feeder and drinker space. Birds were fed a commercial five-phase diet (Table 2.1) in specific quantities (kg/bird; Table 2.2) and supplemental feeders and drinkers were provided throughout the first ten days. Diet changes were made when the pre-determined amount of each ration was finished, and the total feed amount was adjusted at each diet change to account for mortality. Intact small square straw bales were provided as environmental enrichment devices (1 bale/90 birds). As straw bales were destroyed, the straw was spread throughout the room and the bale was replaced, thus, no additional litter management was applied.

The standard room temperature curve and lighting program can be seen in Tables 2.3 and 2.4, respectively. LED (light emitting diode) bulbs were used as the light source with daylength starting at 23L:1D (40 lux) and gradually reduced to 18L:6D (5 lux) by day 10.

Air quality was monitored and controlled from day 1 and humidifiers were utilized during the first 7 days to maintain relative humidity (RH) at a minimum of 50% (Aviagen, 2015b). Carbon dioxide (CO_2) was measured three times per week using a handheld CO_2 meter (CO240; Extech Instruments; Nashua, NH) and ammonia was monitored once per week until differences were noted, then twice per week using ammonia Dräger-Tubes and a handheld pump (Draeger, Inc.; Houston, TX). If CO_2 levels varied by 20% or ammonia differed by 5 ppm between rooms, ventilation was adjusted in each individual room to result in matched air quality for all density conditions (Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019).

Mortality and morbidity were monitored daily, and birds were necropsied by an independent diagnostic laboratory to identify the cause of illness or death (Table 2.5). Birds were culled when necessary due to illness and/or skeletal or growth abnormalities. During the first 3-days, all mortalities and culls were replaced with extra poults in an attempt to maintain the final predicted SD. In block 1, additional space was blocked off at week 3 to account for high mortality rates during the first 3 weeks of the trial, however, at week 8 space was opened up to maintain the estimated final stocking density for each treatment. In block 2, birds were removed from each treatment at week 9 to account for low mortality rates during block 2, thus the final predicted stocking density could be reached at week 11.

2.3.3 Data Collection

Body weight and feed consumption were measured by collecting group (room basis) body weights and feeder weights on day 0 and weeks 3, 5, 8, and 11. Feed-to-gain ratio (F:G), mortality corrected feed-to-gain ratio ($F{:}G^{m}$), and feed consumption was calculated for each of these times periods. Flock uniformity was determined from individually weighing a subsample of birds (30 birds/rep) at weeks 8 and 11. After the bird replacement period (day 1-3), all mortality and culls were recorded daily and sent for necropsy to an independent diagnostic laboratory, and all mortality and morbidity results were then categorized by cause (Table 2.5). Economic analyses were performed to determine net income from both blocks by utilizing feed costs (obtained from the feed company), meat price (obtained from processing plant), poult costs (obtained from commercial hatchery) in 2019 and 2020 and by calculating variables such as average BW, number of birds placed, and number of birds marketed for each SD treatment.

Room temperatures were monitored hourly for the duration of both blocks using iButton Hygrochron™ temperature and humidity data loggers (Maxim Integrated; San Jose, CA) and average weekly room temperatures were calculated over the course of each block.

2.4 Statistical Analyses

The experimental design was a randomized complete block design (RCBD; block as trial) with rooms as the experimental unit. Data from both blocks were analyzed together using regression analysis in SAS (SAS®9.4, Cary, NC, USA) via the Regression Procedure (Proc Reg) and Surface Response Regression Procedure (Proc RSReg) to determine if there was either a linear or quadratic relationship between SD and the performance parameters being evaluated. An analysis of variance (ANOVA) was performed for all data using the Proc Mixed Procedure (SAS®9.4, Cary, NC, USA) and Tukey's range test was used to separate means. Data were checked for normality (Proc Univariate) and log transformed if required. If $P \leq 0.05$, differences were considered significant and if $P \leq 0.10$, trends were noted.

Table 2.1. Nutrient content per kilogram of diets fed to turkey hens from 0-11 weeks of age

Nutrient	Starter 1	Starter 2	Grower 1	Grower 2	Finisher
ME[1] – poultry (kcal/kg)	3015	3089	3142	3229	3276
Crude protein (%)	28.2	26.0	25.0	21.9	20.5
Crude fat (%)	7.4	7.6	7.9	8.1	8.2
Crude fibre (%)	3.3	3.2	3.3	3.0	3.0
Chloride (%)	0.3	0.3	0.2	0.2	0.2
Calcium (%)	1.5	1.4	1.2	1.2	1.0
Phosphorus-total (%)	1.0	1.0	0.9	0.8	0.8
Sodium Chloride (%)	0.3	0.3	0.2	0.2	0.2
Sodium (%)	0.2	0.2	0.2	0.2	0.2
Lys-DP[2] (%)	1.7	1.5	1.4	1.2	1.1
Met-DP (%)	1.2	0.7	0.6	0.5	0.5
Met+Cys-DP (%)	1.5	1.0	0.9	0.8	0.7
Thr-DP (%)	1.0	0.9	0.8	0.7	0.7
Added selenium (mg/kg)	0.3	0.3	0.3	0.3	0.3
Add vitamin A (KIU/kg)	12.0	12.0	12.0	10.0	10.0
Add vitamin D_3 (KIU/kg)	4.4	4.4	4.4	4.5	4.5
Add vitamin E (IU/kg)	100.0	100.0	100.0	60.0	60.0

[1]ME – metabolizable energy
[2]DP – digestible protein

Table 2.2. Diet schedule based on kg of feed per bird

Diet name	Diet form	Age fed (weeks)	kg/bird	# of birds	Total Feed (kg)
Starter #1	Crumble	0-4	1.4	3550	4970
Starter #2	Crumble	5-6	1.8	3550	6390
Grower #1	Small Pellet	7-8	2.8	3550	9940
Grower #2	Pellet	9-10	3.8	3550	13490
Finisher	Pellet	11	2.2	3550	7810

Table 2.3. Standard room temperature curve (Aviagen, 2015b)

Age (Weeks)	Target (°C)	Maximum (°C)	Minimum (°C)
1*	28	31	27
2	27	29	25
3	25	27	23
4	23	26	22
5	22	24	20
6	21	23	19
7	20	22	18
8	19	21	17
9	18	20	16
10	17	19	15
11	16	18	14

*Heat lamps were added during the first week

Table 2.4. Lighting program

Age (Days)	Light:Dark Hours	Lights On	Lights Off	Light Intensity(lux)
0-2	23L:1D	4:00 am 15 min of dawn. Fully on at 4:15 am.	2:45 am 15 min of dusk. Fully off at 3:00 am.	40 + heat lamp
3	22L:2D	4:00 am 15 min of dawn. Fully on at 4:15 am.	1:45 am 15 min of dusk. Fully off at 2:00 am.	40
4	21L:3D	4:00 am 15 min of dawn. Fully on at 4:15 am.	12:45 am 15 min of dusk. Fully off at 1:00 am.	40
5	20L:4D	4:00 am 15 min of dawn. Fully on at 4:15 am.	11:45 pm 15 min of dusk. Fully off at 12:00 am.	30
6	20L:4D	4:00 am 15 min of dawn. Fully on at 4:15 am.	10:45 pm 15 min of dusk. Fully off at 11:00 pm.	30
7	19L:5D	4:00 am 15 min of dawn. Fully on at 4:15 am.	9:45 pm 15 min of dusk. Fully off at 10:00 pm.	20
8	19L:5D	4:00 am 15 min of dawn. Fully on at 4:15 am.	8:45 pm 15 min of dusk. Fully off at 9:00 pm.	10
9d – 11wk	18L:6D	4:00 am 15 min of dawn. Fully on at 4:15 am.	7:45 pm 15 min of dusk. Fully off at 8:00 pm.	5

Table 2.5. Mortality and culls diagnosis categories

Category	Diagnosis
Aggression	Head/neck pecked, wing pecked, and/or snood pulled
Metabolic	Ascites, chronic heart, right ventricular heart disease, round heart disease, slipped tendon, aortic rupture, peri-renal hemorrhage, hemorrhagic fatty liver syndrome
Infectious	Arthritis, synovitis, cellulitis, hepatitis, endocarditis, pericarditis, peritonitis, splenitis, keel bursitis, bursitis, enlarged hock joints
Unknown	No visible lesions
Mechanical	Broken wing, broken leg, ruptured tendon, trauma
Skeletal	Rickets, valgus varus, rotated tibia, spondylolisthesis, tibial dyschondroplasia
Other	Impaction, hepatomegaly, lateral tibial tarsal ligament rupture, enlarged kidney, enlarged spleen

2.5 Results

All results are presented in terms of the final estimated SD. The actual SD achieved at 3, 5, 8, and 11 weeks of age is shown in Table 2.6. At 11 weeks of age, the average final SD achieved was 31.70, 42.38, 52.01, and 61.33 kg/m^2. Carbon dioxide and ammonia concentrations (ppm) for block 1 and block 2 are shown in Table 2.7. The average CO_2 concentrations did not differ (*P* (ANOVA)=0.70 and 0.24 for blocks 1 and 2, respectively) across treatments for either block. Similarly, average ammonia concentrations did not differ across treatments for both blocks *P* (ANOVA) = 0.06 and 0.32 for blocks 1 and 2, respectively). It is important to note that spikes in CO_2 and ammonia occurred when outside ambient temperatures were extremely low as both blocks took place over winter in Saskatchewan, Canada. This resulted in spikes in CO_2 and ammonia as ventilation was reduced to maintain internal temperatures and prevent unwanted chilling of the birds. However, any fluctuations in air quality were corrected through ventilation. It is evident that air quality was similar across all treatments which helped to control any potential effects of CO_2 and ammonia on the parameters measured in this study. No differences were noted for average room temperature for each SD treatment from 1 to 11 weeks (Table 2.8).

Table 2.6. Actual stocking densities (kg/m^2) achieved at 3, 5, 8, and 11 weeks of age

Age (weeks)	n	Stocking density (kg/m^2)			
		30	40	50	60
3	4	3.44	4.57	5.65	6.65
5	4	8.87	12.07	14.95	17.59
8	4	21.25	28.66	35.13	41.51
11	4	31.70	42.38	52.01	61.33

Table 2.7. Average room carbon dioxide (CO_2) and ammonia concentrations (ppm) in relation to estimated stocking density over 11 weeks

Parameter (ppm)	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)
		30	40	50	60		
Block 1							
Average CO_2	2	1976	1990	2030	2010	18.50	0.70
CO_2 range	2	326-4058	356-3820	475-4211	463-3907	-	-
Average ammonia	2	4.8	6.5	6.0	6.6	0.315	0.11
Ammonia range	2	0-15	0-12	0-25	0-25	-	-
Block 2							
Average CO_2	2	1958	1998	2059	2076	26.12	0.24
CO_2 range	2	463-4561	634-4537	584-4089	661-3997	-	-
Average ammonia	2	6.8	7.5	6.4	6.7	0.211	0.32
Ammonia range	2	0-30	0-25	0-25	0-24	-	-

[1]Standard error of the mean.

[2]ANOVA considered significant if $P \leq 0.05$.

Table 2.8. Average weekly room temperature (°C) across estimated final stocking density treatments from 1 to 11 weeks

Age (weeks)	n	Stocking density (kg/m^2)				SEM[1]	P-value[2] (ANOVA)
		30	40	50	60		
1	4	28.2	28.3	28.3	28.3	0.081	0.98
2	4	27.4	27.4	27.2	27.3	0.079	0.90
3	4	26.0	26.0	25.7	25.8	0.087	0.69
4	4	24.1	23.9	23.9	23.9	0.058	0.65
5	4	22.4	22.3	22.1	22.2	0.072	0.63
6	4	20.9	20.8	20.7	20.7	0.113	0.96
7	4	19.9	19.5	19.6	19.6	0.089	0.44
8	4	19.1	19.1	18.8	19.0	0.058	0.36
9	4	18.6	18.4	18.4	18.5	0.065	0.88
10	4	17.7	17.6	17.3	17.3	0.115	0.63
11	4	16.4	16.5	16.4	16.5	0.084	0.96

[1]Standard error of the mean.
[2]ANOVA considered significant if $P \leq 0.05$.

2.5.1 Body weight

At placement, poult BW was similar across all treatments and no differences in BW were observed at week 3, 5, or 8 (Table 2.9). At 11 weeks of age, turkey hen BW demonstrated a linear decrease (*P*=0.05) as SD increased (8.36, 8.35, 8.30, 8.19 kg for SD treatments 30, 40, 50, and 60 kg/m^2, respectively). Stocking density did not affect turkey hen BW gain for 0-3, 3-5, 5-8, and 8-11 weeks (Table 2.10). Overall BW gain from 0-11 weeks of age demonstrated a linear tendency (*P*=0.06) to decrease with increasing SD.

Table 2.9. Effect of estimated final stocking density on turkey hen body weight (kg) at 0, 3, 5, 8, and 11 weeks of age

Age (weeks)	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
0	4	0.058	0.059	0.058	0.058	0.001	0.32	0.93	0.88	-
3	4	0.80	0.80	0.79	0.79	0.003	0.36	0.19	0.88	-
5	4	2.09	2.13	2.12	2.10	0.015	0.12	0.87	0.87	-
8	4	5.06	5.12	5.05	5.03	0.030	0.43	0.54	0.50	-
11	4	8.36	8.35	8.30	8.19	0.033	0.20	0.05	0.44	$Y = -0.56e^{-2}x+8.55$

[1]Standard error of the mean.
[2]Regression considered significant if $P \leq 0.05$.

Table 2.10. Effect of estimated final stocking density on turkey hen body weight gain (kg) from 0-3, 3-5, 5-8, 8-11, and 0-11 weeks of age

Age (weeks)	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
0-3	4	0.74	0.74	0.73	0.73	0.002	0.37	0.12	0.89	-
3-5	4	1.29	1.34	1.33	1.31	0.013	0.06	0.64	0.25	-
5-8	4	2.97	2.99	2.92	2.93	0.038	0.48	0.60	0.89	-
8-11	4	3.31	3.22	3.26	3.16	0.029	0.40	0.14	0.93	-
0-11	4	8.31	8.29	8.25	8.13	0.033	0.20	0.06	0.44	-

[1]Standard error of the mean.

[2]Regression considered significant if $P \leq 0.05$.

2.5.2 Feed consumption and feed efficiency

Turkey hen feed consumption from 0-3, and 5-8 weeks of age was not affected by SD (Table 2.11). From 3-5 weeks of age, there was a tendency for feed consumption to be higher in the 40 kg/m^2 treatment (ANOVA; P=0.09). From 8-11 weeks of age, feed consumption linearly decreased (P<0.01) as SD increased (7.50, 7.47, 7.38, 7.21 kg for SD treatments 30, 40, 50, and 60 kg/m^2, respectively). Overall feed consumption, from 0-11 weeks of age, demonstrated a quadratic relationship (P=0.04) with the lowest feed consumption in the 60 kg/m^2.

Feed efficiency (feed-to-gain; F:G) and mortality corrected F:G ratio did not demonstrate a significant regression for 0-3, 3-5, 5-8, 8-11, nor 0-11 weeks of age (Table 2.12). However, a difference at 3-5 weeks of age was observed for F:G (ANOVA; P=0.03) in which the poorest F:G ratio was observed in the 30 kg/m^2 treatment (1.45) and the best feed efficiency was observed in the 50 kg/m^2 treatment (1.42). This difference was also noted in mortality corrected F:G in which the 30 kg/m^2 treatment was poorer (ANOVA; P=0.05) than the 50 kg/m^2 treatment.

Table 2.11. Effect of estimated final stocking density on turkey hen feed consumption (kg per bird) from 0-3, 3-5, 5-8, 8-11, and 0-11 weeks of age

Age (weeks)	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
0-3	4	0.88	0.88	0.88	0.88	0.004	0.90	0.69	0.99	-
3-5	4	1.85	1.90	1.87	1.86	0.029	0.09	0.74	0.22	-
5-8	4	4.97	5.04	4.95	4.95	0.050	0.52	0.79	0.75	-
8-11	4	7.50[a]	7.47[ab]	7.38[ab]	7.21[b]	0.041	0.04	<0.01	0.32	$Y = -0.94e^{-2}x+7.81$
0-11	4	15.19[ab]	15.29[a]	15.08[ab]	14.90[b]	0.055	0.02	0.01	0.04	$Y = -0.88e^{-3} x^2 +0.067x+14.00$

[1]Standard error of the mean.
[2]Regression considered significant if P≤0.05.
[a,b] Means with common letter within a row do not differ significantly (P≤0.05).

Table 2.12. Effect of stocking density on turkey hen feed-to-gain ratio and mortality corrected feed-to-gain ratio from 0-3, 3-5, 5-8, 8-11, and 0-11 weeks of age

Age (weeks)	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
Feed-to-gain (F:G)										
0-3	4	1.20	1.20	1.20	1.21	0.006	0.42	0.50	0.88	-
3-5	4	1.45^{a}	1.43^{ab}	1.42^{b}	1.42^{ab}	0.017	0.03	0.17	0.46	-
5-8	4	1.68	1.70	1.71	1.71	0.006	0.28	0.13	0.61	-
8-11	4	2.78	2.84	2.81	2.80	0.109	0.91	0.96	0.88	-
0-11	4	1.94	1.96	1.94	1.94	0.021	0.68	0.92	0.76	-
Feed-to-gain mortality corrected ($F{:}G^{m}$)										
0-3	4	1.18	1.19	1.19	1.19	0.006	0.51	0.72	0.93	-
3-5	4	1.43^{a}	1.42^{ab}	1.41^{b}	1.42^{ab}	0.016	0.05	0.23	0.24	-
5-8	4	1.67	1.68	1.69	1.69	0.005	0.15	0.20	0.64	-
8-11	4	2.68	2.74	2.68	2.72	0.110	0.68	0.94	0.96	-
0-11	4	1.90	1.92	1.90	1.91	0.024	0.38	0.95	0.87	-

[1]Standard error of the mean.
[2]Regression considered significant if $P \leq 0.05$.

2.5.3 Mortality

Total mortality as a percentage of turkey hens placed was not significantly affected by SD (Table 2.13). Total mortality and culls categorized by cause showed significant results for the "other" category (foreign body, hepatomegaly, lateral tibial tarsal ligament rupture, enlarged kidney, enlarged spleen), infectious category (yolk sac infection, arthritis, synovitis, cellulitis, hepatitis, endocarditis, pericarditis, enlarged hock joints, etc.) and for aggression (Table 2.14). Total mortality and culls in the "other" category were highest in the lowest density treatment of 30 kg/m^2 (linear; P=0.03). Total aggression related mortality and culls were highest in the lowest SD of 30 kg/m^2 (linear; P=0.02) and lowest in the 60 kg/m^2 treatment. Infectious related mortality was highest in the 60 kg/m^2 treatment from 8-11 and 0-11 weeks (ANOVA; P<0.01). During week 5-8, metabolic related mortality and culls were highest in the 60 kg/m^2 treatment (linear; P<0.01; Table 2.15). From day 3 to 3 weeks of age, there was no aggression related mortality and culls, however, during week 8-11, mortality and culls caused by aggression were significantly higher in the lowest SD treatment of 30 kg/m^2 and no incidences were noted in the 60 kg/m^2 treatment (linear; P=0.03).

Table 2.13. Effect of estimated final stocking density on turkey hen percent mortality and culls (%) from day 3-3 weeks, 3-5, 5-8, 8-11, and 0-11 weeks of age

Age	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
Day 3-3 weeks	4	4.32	3.86	4.72	5.39	1.087	0.29	0.94	0.79	-
Week 3-5	4	1.53	1.42	1.14	0.83	0.278	0.27	0.56	0.63	-
Week 5-8	4	0.76	0.90	0.83	1.05	0.106	0.32	0.26	0.95	-
Week 8-11	4	1.44	1.29	1.92	1.23	0.217	0.14	0.88	0.55	-
Week 0-11	4	8.05	7.47	8.61	8.49	1.592	0.37	0.86	0.93	-

[1]Standard error of the mean.

[2]Regression considered significant if $P \leq 0.05$.

Table 2.14. Effect of estimated final stocking density on turkey hen percent mortality and culls (% of birds placed) by cause from day 3 to 11 weeks of age

Cause[3]	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
Metabolic	4	0.34	0.39	0.78	0.70	0.096	0.11	0.08	0.85	-
Skeletal	4	0.59	0.39	0.52	0.53	0.118	0.82	0.84	0.70	-
Infectious	4	4.66[ab]	3.99[b]	5.08[a]	5.56[a]	1.213	<0.01	0.80	0.87	-
Unknown	4	0.59	1.03	1.09	0.96	0.198	0.22	0.61	0.37	-
Other	4	1.02[a]	0.97[a]	0.57[b]	0.66[ab]	0.075	0.04	0.03	0.57	Y = -0.015x+1.47
Mechanical	4	0.08	0.13	0.10	0	0.043	0.79	0.48	0.44	-
Aggression	4	0.68	0.58	0.52	0.09	0.103	0.07	0.02	0.28	Y = -0.018x+1.29

[1]Standard error of the mean.
[2]Regression considered significant if $P \leq 0.05$.
[3]**Metabolic**: ascites, chronic heart, right ventricular heart disease, round heart disease, slipped tendon, aortic rupture, peri-renal hemorrhage, hemorrhagic fatty liver syndrome; **Skeletal**: rickets, valgus varus, rotated tibia, kinky back, tibial dyschondroplasia; **Infectious:** yolk sac infection, arthritis, synovitis, cellulitis, hepatitis, endocarditis, pericarditis, peritonitis, splenitis, bursitis, enlarged hock joints; **Unknown**: no visible lesion; **Other**: foreign body, hepatomegaly, lateral tibial tarsal ligament rupture, enlarged kidney, enlarged spleen; **Mechanical**: broken wing, broken leg, ruptured tendon, trauma; **Aggression**: head/neck pecked, wing pecked, snood pulled.

Table 2.15. Effect of estimated final stocking density on turkey hen percent mortality and culls (% of birds placed) by cause from day 3-3 weeks, 3-5, 5-8, and 8-11 weeks of age (n=4)

% In Category[3]	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
	30	40	50	60					
Day 3-3 weeks									
Metabolic	0.08	0.13	0	0.13	0.042	0.67	0.94	0.58	-
Skeletal	0.17	0.13	0.26	0.18	0.053	0.79	0.61	0.80	-
Infectious	3.05	2.90	3.94	4.42	0.945	0.99	0.75	0.95	-
Unknown	0.34	0.58	0.47	0.35	0.111	0.79	0.90	0.34	-
Other	0.68	0.19	0.26	0.31	0.072	0.07	0.09	0.07	-
Mechanical	0	0	0	0	0	-	-	-	-
Aggression	0	0	0	0	0	-	-	-	-
Week 3-5									
Metabolic	0.08	0.13	0.10	0.18	0.034	0.82	0.41	0.88	-
Skeletal	0.17	0	0.05	0.09	0.032	0.32	0.57	0.11	-
Infectious	1.02	0.90	0.67	0.57	0.219	0.96	0.59	0.99	-
Unknown	0.08	0.13	0.16	0	0.037	0.50	0.50	0.18	-
Other	0.08	0.13	0	0	0.029	0.31	0.15	0.69	-
Mechanical	0	0	0	0	0	-	-	-	-
Aggression	0	0.06	0	0.04	0.019	0.58	0.69	0.82	-
Week 5-8									
Metabolic	0^{b}	0^{b}	0.26^{a}	0.31^{a}	0.040	<0.01	<0.01	0.61	$Y = 0.12e^{-2}x-0.39$
Skeletal	0.08	0.13	0.05	0.18	0.038	0.74	0.59	0.66	-
Infectious	0.42	0.19	0.26	0.26	0.084	0.90	0.68	0.60	-
Unknown	0	0.06	0.05	0.09	0.028	0.78	0.35	0.79	-
Other	0.17	0.45	0.21	0.13	0.054	0.15	0.45	0.10	-
Mechanical	0	0	0	0	0	-	-	-	-
Aggression	0.08	0.06	0	0.04	0.027	0.77	0.48	0.58	-
Week 8-11									
Metabolic	0.17	0.13	0.41	0.09	0.063	0.32	0.99	0.31	-
Skeletal	0.17	0.13	0.16	0.09	0.038	0.92	0.57	0.87	-
Infectious	0.17^{ab}	0^{b}	0.21^{ab}	0.31^{a}	0.042	0.04	0.09	0.08	-
Unknown	0.17	0.26	0.41	0.53	0.122	0.80	0.32	0.85	-

Other	0.08	0.19	0.10	0.22	0.044	0.65	0.46	0.99	-
Mechanical	0.08	0.13	0.10	0	0.043	0.79	0.48	0.44	-
Aggression	0.59	0.45	0.52	0	0.101	0.08	0.03	0.28	Y = -0.017x+1.16

[1]Standard error of the mean.

[2]Regression considered significant if P≤0.05.

[3]**Metabolic**: ascites, chronic heart, right ventricular heart disease, round heart disease, slipped tendon, aortic rupture, peri-renal hemorrhage, hemorrhagic fatty liver syndrome; **Skeletal**: rickets, valgus varus, rotated tibia, kinky back, tibial dyschondroplasia; **Infectious**: yolk sac infection, arthritis, synovitis, cellulitis, hepatitis, endocarditis, pericarditis, peritonitis, splenitis, bursitis, enlarged hock joints; **Unknown**: no visible lesion; **Other**: foreign body, hepatomegaly, lateral tibial tarsal ligament rupture, enlarged kidney, enlarged spleen; **Mechanical**: broken wing, broken leg, ruptured tendon, trauma; **Aggression**: head/neck pecked, wing pecked, snood pulled.

2.5.4 Flock uniformity

Turkey hen flock uniformity, presented as the percentage of birds found within 5, 10, or 15% of the mean room body weight as seen in Table 2.16, was not affected in relation to SD at 8 or 11 weeks of age.

Table 2.16. Effect of estimated final stocking density on turkey hen uniformity presented as the percentage of birds found within 5, 10, and 15% of the mean body weight at 8 and 11 weeks of age.

% within x of the mean	n	Stocking density (kg/m^2)				SEM[1]	CV[2]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[3]
		30	40	50	60						
Week 8											
5	4	43.33	49.17	47.50	48.33	2.650	22.52	0.70	0.60	0.58	-
10	4	79.17	75.83	75.00	79.17	2.419	12.52	0.90	0.94	0.52	-
15	4	90.83	94.16	93.33	88.33	2.041	8.91	0.71	0.75	0.32	-
Week 11											
5	4	47.50	43.33	51.67	45.00	2.386	20.36	0.48	0.85	0.76	-
10	4	77.50	80.83	85.83	79.17	1.984	9.82	0.40	0.63	0.23	-
15	4	93.33	95.83	93.33	93.33	1.019	4.34	0.80	0.80	0.53	-

[1]Standard error of the mean.
[2]Coefficent of variation.
[3]Regression considered significant if $P \leq 0.05$.

2.5.5 Economic analysis

The economic analysis was performed using poult cost, feed cost, number of birds shipped, and bird meat income as seen in Table 2.17. The highest SD resulted in higher profits (linear; $P<0.01$). The net income was \$1706.88, \$2283.41, \$2723.20, \$3144.22 per room for the 30, 40, 50, and 60 kg/m^2 treatments, respectively.

Table 2.17. Economic analyses of estimated final stocking density of turkey hens to 11 weeks of age

Parameter per room	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
Number placed (room)	4	295	388	482	571	-	-	-	-	-
Poult cost ($)[3]	4	604.75	795.40	988.10	1170.55	-	-	-	-	-
Number shipped	4	248.25	330.00	403.50	478.75	-	-	-	-	-
Avg. final BW (kg)	4	8.36	8.35	8.30	8.19	0.033	0.20	0.05	0.44	Y= 0.0056x+8.55
Live wt. shipped (kg)	4	2076.33	2754.83	3351.45	3921.20	-	-	-	-	-
Bird meat income[4]	4	3955.40	5247.94	6384.51	7469.89	-	-	-	-	-
Feed intake (kg)	4	3989.54	5357.84	6447.61	7535.53	-	-	-	-	-
Feed cost ($)[5]	4	1643.77	2169.13	2673.21	3155.12	-	-	-	-	-
Net income per bird ($)	4	6.87	6.90	6.74	6.56	0.109	0.19	0.28	0.63	-
Net income per room ($)[6]	4	1706.88[d]	2283.41[c]	2723.20[b]	3144.22[a]	146.264	<0.01	<0.01	0.48	Y=47.52x+326.11

[1]Standard error of the mean.
[2]Regression considered significant if P≤0.05.
[3]Poult cost was $2.05/poult
[4]Meat price per kilogram live weight used was $1.905
[5]Feed price per tonne for Block 1 (Jan-Apr 2019): starter 1- $588; starter 2- $539; grower 1- $521; grower 2- $485; finisher- $460; Feed price per tonne for Block 2 (Nov 2019-Feb 2020): starter 1- $552; starter 2- $521; grower 1- $498; grower 2- $466; finisher- $440
[6]Net income= (Number shipped*Avg. BW* meat price) - (Number placed*Poult cost) - (Feed cost)

2.6 Discussion and Conclusions

In commercial poultry production, there are many factors that can influence performance parameters and economic return, and SD is one of those influencing factors. Body weight, feed efficiency, and mortality of the poultry being raised can directly affect economic return and thus, the effect SD has on those production traits are important to evaluate. As the majority of studies on the effects of SD in turkeys focus on production parameters and more often in turkey toms, with no published studies on SD in hens in the past 22 years, this study with turkey hens is of significant importance.

Increasing SD caused a lower BW in turkey hens at 11 weeks of age in this study, with a tendency for overall BW gain to reduce with increasing density. This effect on BW at older ages has been observed in previous studies (Coleman and Leighton, 1969; Proudfoot et al., 1979a; Leighton et al., 1985; Noll et al., 1991; Martrenchar et al., 1997; Beaulac et al., 2019; Bartz et al., 2020). In these studies, as well as the current study, the number of feeders and drinkers were equalized on a per bird basis, which eliminates the effect of reduced feeder space which in itself could affect growth. In the study by Beaulac et al. (2019), the authors observed a decrease in BW and BW gain with increasing SD (30 to 60 kg/m^2) at 12 and 16 weeks of age when the final predicted SD was closer to being achieved. In comparison, the BW results from this study at 11 weeks of age are similar when the actual SD achieved was 31.70, 42,38, 52.01 and 61.33 kg/m^2 which indicates that there may be an age range (11 weeks and older) at which poor growth effects are observed at higher SD. Therefore, the reduced growth at older ages, when the estimated final SD is more closely achieved, may be related to less space and the birds reduced mobility (Chapter 3) at older ages which makes reaching the feeder more difficult. In addition to poorer mobility, older birds housed at higher SD may also experience reduced motivation to access feeders as they would need to exert more energy to move between their pen mates to access the feeder (Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019), or they may experience more competition at the feeder (Martrenchar et al., 1999) leading to reduced BW.

Few studies have examined the effect of SD on feed consumption in turkeys and all have focused on turkey toms, however, it is an important parameter to examine as it directly affects economic return and growth. Previous studies have found that feed consumption in toms decreased as SD increased between various periods from 12-20 weeks of age (Leighton et al., 1985; Noll et al., 1991; Beaulac et al., 2019) and overall feed consumption from 0-20 weeks (Noll et al., 1991). In accordance with previous literature, the results of this study found a linear decrease in feed consumption from 8-11 and overall feed consumption from 0-11 weeks of age. Similar to the effects of SD on BW at high SD, feed consumption was not influenced by feeder or drinker space as it was balanced on a per bird basis which reduced the impact of feeder space on feed consumption. Feed consumption may have decreased from poorer bird mobility (Chapter 3) and the reduced ability to reach the feeders at older ages. At 8 weeks of age, the percentage of birds at the feeder (Table 3.1) increased with increasing density and by 11 weeks of age, a numerically lower percentage of birds were observed at the feeder in the highest SD treatment (Chapter 3). This change in the percentage of birds present at the feeder could be a result of more social related feeding behaviour at 8 weeks compared to 11 weeks. When social feeding behaviour was studied in broiler chickens, it was noted that birds were more likely to approach a feeder with birds already present and would stay at that feeder for longer when the other birds were feeding (Collins and Sumpter, 2007). This may explain why birds in higher densities were able to move to feeders at 8 weeks of age despite the presence of footpad dermatitis (FPD; Chapter 3) and may have been motivated by social feeding behaviour. However, a shift away from social feeding behaviour occurred at 11 weeks of age when poorer mobility, stress, and lack of floor space inhibited birds from reaching the feeder, resulting in decreased feed consumption and lower BW.

Feed efficiency (F:G) and $F:G^m$ demonstrated no relationship with SD. Previous literature has found inconsistent results for feed efficiency in relation to SD as some studies have found that feed efficiency was poorer at high SD (60 kg/m^2) from 4 weeks of age and older (Beaulac et al., 2019) and 8 weeks of age and older (Leighton et al., 1985; Noll et al., 1991) at 40.1 and 60.9 kg/m^2, respectively. As previously mentioned, these studies equalized feeder and drinker space on a per bird basis, thus, access to feeder space was not a confounding factor. However, there may have been other stressors contributing to the results seen in previous studies as confounding factors can add stressors to the birds which may affect feed efficiency. Moran (1985) did not provide feeders and drinkers on a per birds basis and found F:G to be negatively impacted by high SD (21.4 kg/m^2), which indicates that this may have been a confounding factor. McFarlane et al. (1989) observed a negative impact on F:G with the exposure of chicks to many stressors. In the study by Beaulac et al., (2019), air quality was also equalized between treatments, however, poorer F:G^m from 4 weeks of age in toms correlated to increased stress observed at that age (H/L ratios) as well as poorer feather cover and cleanliness at older ages at higher SD (Beaulac and Schwean-Lardner, 2018). The authors suggest that more energy was directed toward thermoregulation which decreased feed efficiency. In comparison, similar stress and feather condition results were observed in this study (Chapter 3), however, no effect was seen on F:G. This correlates to other studies that found no effect of SD on feed efficiency (Coleman and Leighton, 1969; Proudfoot et al., 1979a; Zuidhof et al., 1993, 1995). The studies listed examined either both toms and hens or toms alone which eliminates sex differences as a cause. Internal room temperature and air quality were consistent across treatments and ventilation was adjusted when differences were noted, which may have contributed to the F:G results. In the study by Zuidhof et al. (1995), F:G was not affected by SD (25 and 50 kg/m^2), however, during the first 2 weeks of the study, room temperature was lower in one of the treatment rooms which the authors state may have resulted in poorer F:G and lower BW gain. Coleman and Leighton (1969) found decreased feed efficiency at high SD (48 kg/m^2) in one of two of their experiments but no effect on F:G in relation to SD was observed in their second experiment. The authors suggest that the effect seen in the first experiment at high SD was because it was conducted in winter with curtain sided barns compared to summer for the second experiment. In both of these studies, the amount of energy and feed required to thermoregulate and maintain body temperature would have resulted in poorer feed efficiency. Thus, without confounding factors of room temperature and feeder space, feed efficiency was not impacted by SD in this study.

In this study, overall mortality was unaffected by SD and this result is similar to previous literature. However, numerical differences are important to note as statistically significant results may not accurately depict the effects of SD on bird mortality, as low mortality rates and variability between treatment rooms or other confounding factors make determination of significance difficult when discussing mortality (Beaulac et al., 2019). Coleman and Leighton (1969) saw numerical increases in mortality with increasing density (27.5 to 48 kg/m^2) and similarly, Noll et al. (1991) observed a tendency for higher mortality with increasing density (29.4 to 60.9 kg/m^2). Beaulac et al. (2019) observed numerically higher mortality in the lowest (30 kg/m^2) and highest SD (60 kg/m^2) treatments and the authors suggest this may have been due to increased activity or increased frustration with large group size in the low and high SD treatments, respectively. When evaluating turkey hen percent mortality and culls by cause, the cause category of “other”, which includes death from a foreign body, hepatomegaly, lateral tibial tarsal ligament rupture, enlarged kidney, and enlarged spleen, demonstrated a linear increase with decreasing density. Statistical analysis did not show incidence of one condition within the ‘other’ category to be higher than another as these mortality causes occur at a low incidence. However, it may be important to understand why these conditions could occur when related to SD. Ligament ruptures in turkeys can be caused from trauma after physical activity, when birds move from a sitting to standing position, or stress on the hock joints and tendons from extreme weight (Crespo et al., 2002). Hepatomegaly, or enlarged liver, was linked to an increased stress response in broiler chicks given ACTH, and increased levels of corticosterone can cause an accumulation of hepatic lipid content because the liver is the major site of fatty acid synthesis in chickens (Puvadolpirod and Thaxton, 2000). Infectious related mortality and culls (8-11 and 0-11 weeks) was highest in the high SD treatment (60 kg/m^2) which was also observed in the study by Beaulac et al (2019). The authors hypothesized that presence of footpad lesions and poor feather cleanliness could potentially play a role in infectious disease levels. The higher incidence of FPD and poor feather cleanliness at high SD (Chapter 3) in the current study supports this hypothesis as footpad lesion have been associated with more secondary infections (Martrenchar et al., 2002; Mayne, 2005).

Overall aggression related mortality and culls for the duration of the trials linearly increased with decreasing density. This may be related to behavioural changes observed at 8 weeks of age such as increased walking, standing, litter pecking, and most evidentiary, the increase in aggressive behaviour at low SD (Chapter 3). This is similar to the study by Beaulac et al. (2019) in which aggression related mortality and culls in toms were numerically higher in the lowest SD during week 8-12 and a similar trend was observed from week 12-16. The authors suggest that birds at low SD may be more active compared to birds at high SD when floor space is reduced (Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019). During week 5-8, metabolic related mortality and culls (such as ascites, heart disease, slipped tendon, aortic rupture) were highest at high SD. Slipped tendons can cause lameness or present as bowed legs in turkeys with fast growing strains being more genetically susceptible to this condition which can result from trauma (Balloun, 1958; Julian, 1984). This may relate to increased disturbances observed in birds at high SD at 8 weeks of age (Chapter 3) when floor space starts to decrease, and birds would have to walk over a resting pen mate when maneuvering through the room during a significant growth period. Other metabolic causes such as aortic rupture and heart disease have been observed in fast growing strains of turkeys as they have the highest arterial blood pressure of all vertebrates and commonly increases more during the growing period of 6-20 weeks (Krista et al., 1965, 1969; Guenthner et al., 1978), which correlates to the 5-8 week period of metabolic related mortality and culls at high SD.

To the best of the author's knowledge, only one study has evaluated flock uniformity in relation to SD in turkeys (Beaulac et al., 2019). Flock uniformity was not impacted by SD in the current study, and this is in accordance with the study by Beaulac et al. (2019). This may differ from broiler data. Broilers exhibited poorer uniformity at low SD but higher BW which may indicate that they grew to their genetic potential, whereas broilers at high SD were more uniform due to reduced space (Feddes et al., 2002). This may be because of social feeding behaviour where birds are more likely to eat when others are present at the feeder which will result in more coordinated feeding at high SD (Collins and Sumpter, 2007; Beaulac et al., 2019). As no differences were observed in this study nor the study in toms, this may suggest species differences or too few birds sampled (Beaulac et al., 2019).

In conclusion, high SD negatively impacts some aspects of turkey hen performance to 11 weeks of age. It was hypothesized that high SD will negatively affect BW, feed consumption, feed efficiency, and flock uniformity due to reduced space allowance and environmental stressors. It was also hypothesized that at low SD there will be more aggression. Although feed efficiency and uniformity were unaffected by SD, growth and feed consumption were negatively impacted at high SD and higher aggression related mortality and culls occurred at low SD. It is important to note that although high SD resulted in a greater economic return despite poor performance effects, additional factors such as management costs, equipment damage, labour costs, and potential carcass condemnations were not evaluated and may affect income at high SD. In addition, the birds' health and welfare needs to be evaluated in order to make recommendations.

2.7 Acknowledgements

The author would like to acknowledge the Canadian Poultry Research Council, Agriculture and Agri-Food Canada, Aviagen Turkeys Inc., and Charison's Turkey Hatchery Ltd.

for their financial support and thank the students and staff at the University of Saskatchewan Poultry Centre for their help with these turkey trials.

3.0 Chapter 3: Evaluating the effects of stocking density on the behaviour, health and welfare of turkey hens to 11 weeks of age

Chapter 3 evaluates the impacts of graded levels of SD on the health and welfare of turkey hens to 11 weeks of age. The health and welfare parameters studied in this chapter include footpad lesions, mobility, feather cover and cleanliness, the incidence of aggressive damage, H/L ratios, behaviour, and litter moisture.

3.1 Abstract

Stocking density (SD) is an important factor that impacts the health and welfare of birds by potentially affecting footpad lesions, mobility, feather condition and behaviour. Few studies exist that evaluate turkey health and wellbeing, with no published studies on the effects of SD in turkey hens in the past 22 years. Nicholas Select hens (n=3550 poults in each of 2 blocks) were randomly placed in one of four final estimated SD treatments of 30, 40, 50, or 60 kg/m^2 in two blocks from 0-11 weeks of age. Birds were housed in open rooms (67.5m^2) with four replications per treatment. Feeder and drinker space were equalized on a per bird basis. Ventilation was adjusted to equalize air quality across all treatments based on carbon dioxide (CO_2) and ammonia measures. At week 8 and 11, footpad lesions (scale 0-4), mobility (subjective gait score scale 0-5), feather condition (score 1-4) cleanliness (score 1-4) (30 birds/replicate), and litter moisture (block 1 only) were evaluated. Incidences of aggressive pecking were recorded daily, and birds were treated with a deterrent or culled depending on severity. Heterophil/lymphocyte ratios were evaluated at 3, 5, 8, and 11 weeks of age as a measure of chronic stress (20 birds/replicate). Behaviour was recorded and scan sampled (field of view observations) at 8 and 11 weeks of age. Data were analyzed using regression analysis in SAS 9.4 (Proc Reg and Proc RSReg; SD as independent variable). An analysis of variance was performed for all data (Proc Mixed; SAS 9.4) and Tukey's range test was used to separate means. Differences were considered significant when $P \leq 0.05$. All values reported are in ascending order of SD treatments 30, 40, 50, and 60 kg/m^2. Average footpad scores worsened at week 8 as SD increased (linear; 1.43, 1.55, 1.86, 1.80; $P=0.03$), but no relationship was noted at week 11. Total feather cover scores (sum of five body parts) were poorer in the high SD treatment at week 8 (linear; 17.96, 18.06, 17.30, 16.78; $P<0.01$) and 11 (linear; 17.09, 16.01, 15.68, 14.63; $P<0.01$). Average feather cleanliness scores were poorer in the high SD treatment at week 8 (linear; 1.69, 1.78, 2.64, 3.15; $P<0.01$) and 11 (linear; 1.79, 2.37, 2.80, 3.00; $P<0.01$). Conversely, the total percentage of birds falling into the "incidence of aggressive pecking and culls for aggressive damage" demonstrated a linear increase as SD decreased (7.88, 7.60, 4.05, 2.67 %; $P<0.01$). At 5 weeks of age, H/L ratios linearly increased as SD increased ($P<0.01$) but at 8 weeks, H/L ratios were highest in the 40 kg/m^2 treatment (quadratic; $P=0.03$). At 11 weeks of age, H/L ratios increased with increasing density (quadratic; $P<0.01$). Behaviour was impacted at 8 weeks of age with the percentage of birds at the feeding, resting, and total disturbances linearly increasing as SD increased. The percentage of birds standing, walking, litter pecking (linear), and fighting and aggressive pecking (quadratic) increased with decreasing SD. At 11 weeks of age, the percentage of birds present at the drinker, walking, and environmental pecking increased with decreasing SD while resting, feather pecking, and severe disturbances decreased as SD decreased. Average gait scores tended (*P*=0.06) to be higher in the highest SD and litter moisture tended to increase linearly with increasing SD (*P*=0.08). The results indicate that bird health and welfare were negatively affected by higher SD through worsening footpad lesions, poorer feather cover and cleanliness, increased stress and behavioural changes such as decreased mobility and more disturbances. However, more aggressive behaviour and aggressive damage occurred in the lowest SD.

Keywords: footpad lesions, mobility, feather cover, cleanliness, heterophil/lymphocyte ratio

3.2 Introduction

The health and welfare of poultry raised at high and low stocking densities (SD) has been evaluated by examining footpad lesions, gait scores, feather condition, stress (heterophil/lymphocyte ratio or corticosterone), and behaviour (Martrenchar et al., 1999; Buchwalder and Huber-Eicher, 2004; Gunther and Bessei, 2006; Beaulac and Schwean-Lardner, 2018). The effects of SD on these parameters have been studied more so in broiler chickens compared to turkeys. In the few studies of turkey health and welfare in relation to SD, authors often included only one or two health parameters and more often focused on performance (Denbow et al., 1984; Leighton et al., 1985; Martrenchar et al., 1999; Hafez et al., 2016). However, evaluating bird welfare can be difficult and it is important to establish appropriate measures of welfare. For example, when assessing stress, it is important to choose methods of evaluation that will provide accurate measures for what hypothetical impacts would occur. Examples include measures of acute versus chronic stress and appropriate timing in the diurnal patterns of corticosterone release. In addition, understanding why birds perform specific behaviours in intensive production systems can aid in welfare evaluations and combining physiological parameters with behaviour can help determine if a bird's wellbeing is affected.

Footpad dermatitis (FPD), mobility, and feather condition are important measures when determining turkey wellbeing in relation to SD. Studies have found an increase in the incidence of footpad lesions with increasing density (Martrenchar et al., 1999; Beaulac and Schwean-

Lardner, 2018), and in turn, higher litter moisture was observed at higher densities (Martrenchar et al., 1999). Martrenchar et al., (1999) found turkey hens to have poorer mobility (measured with gait scoring) at higher SD (62.7 kg/m^2) at 12 weeks of age and the same trend was found in toms at 16 weeks of age, which correlated to the presence of FPD at that age (Beaulac and Schwean-Lardner, 2018). Feather cover was found to be poorer in higher densities (48 kg/m^2) in 14-week-old hens (Coleman and Leighton, 1969) and 16-week-old toms housed in higher densities (60 kg/m^2) had poorer feather cover and cleanliness (Beaulac and Schwean-Lardner, 2018).

The ratio of heterophils to lymphocytes (H/L ratio) has previously been used to assess chronic stress. Heterophil/lymphocyte ratios have been studied in relation to SD in turkey toms and no significant effects were observed at 7, 12, 16, or 20 weeks of age (25 to 58 kg/m^2; Hafez et al., 2016). Beaulac and Schwean-Lardner (2018) evaluated H/L ratios at 4, 12, and 16 weeks of age in toms, and H/L ratios were higher with increasing SD at 4 weeks of age. No significant differences were found at 12 or 16 weeks of age. The increased stress response found at 4 weeks of age corresponded with numerical increases in aggressive damage up to 4 weeks of age. Between 4 and 8 weeks of age, a quadratic relationship was observed for aggressive damage with the lowest (30 kg/m^2) and highest density (60 kg/m^2) SD treatments having the highest incidence of aggressive damage. This is an example of the importance of evaluating multiple health and behaviour parameters when assessing the wellbeing of birds.

Behaviour studies in relation to SD have been limited in turkey hens, and few have been conducted with turkey toms. Contradictory behavioural results are common in SD studies due to confounding factors. For example, aggression increased with increasing density in one study (Buchwalder and Huber-Eicher, 2004) and another found that at 16 weeks of age, aggressive pecking linearly increased with increasing SD in toms (Beaulac and Schwean-Lardner, 2018). However, older studies found no increase in aggression with increasing SD levels (Leighton et al., 1985; Martrenchar et al., 1999). Bird activity was reduced in higher SD treatments in some studies (Gunther and Bessei, 2006; Beaulac and Schwean-Lardner, 2018), whereas, others found no effect of SD on walking or resting activity (Martrenchar et al., 1999). Another example of contradicting results for low incidence behaviours such as preening was evident in the study by Gunther and Bessei (2006) where time spent preening was not affected by SD (2.5 to 3.4 birds/m^2; body weight not reported) whereas Beaulac and Schwean-Lardner (2018) observed more toms preening with increasing SD (30 to 60 kg/m^2) at 14 and 16 weeks of age. The above-mentioned studies found no effects of SD on other behaviours evaluated. Thus, with very few studies evaluating the effects of SD on turkey behaviour, stress, and other health parameters, it is imperative that these are studied in turkey hens to determine an appropriate SD that does not negatively affect bird health and welfare.

The objectives of this chapter were to evaluate the effects of SD on turkey hen health and welfare to 11 weeks of age while eliminating confounding factors including air quality (CO_2 and ammonia) and feeder and drinker space. It was hypothesized that increasing SD will negatively impact the health of turkey hens resulting from increased incidence of FPD, poor mobility, and poor feather coverage and cleanliness. This will occur in higher densities as there will be increased litter moisture and less available space for mobility behaviours such as walking to occur. It is also hypothesized that higher H/L ratios will occur at higher densities as a result of chronic stress from increased aggression. These changes will alter bird behaviour at higher SD. It is hypothesized that low SD levels will result in the expression of comfort behaviours as there is more space for this to occur, however, there will be higher aggression seen at low SD levels due to more birds being disturbed by other birds.

3.3 Materials and Methods

The experimental procedures for this experiment were approved by the University of Saskatchewan Animal Care Committee and all birds were cared for as specified in the Guide to the Care and Use of Experimental Animals by the Canadian Council of Animal Care (2009).

3.3.1 Experimental Design

This experiment was comprised of two blocks, starting in January 2019 and November 2019, to allow for increased replication (four room replications per treatment). The study was conducted at the University of Saskatchewan Poultry Centre in a floor facility that includes individual, independently controlled rooms for environmental parameters, allowing for appropriate replication of this study. The four target stocking densities used were 30, 40, 50, and 60 kg/m^2, and parameters were evaluated from placement (day 0) to 11 weeks of age.

3.3.2 Birds and Housing

A total of 3,550 Nicholas Select turkey hens were placed in each block. The birds were obtained from a commercial hatchery, where their beaks and front three toes were treated (infrared trimmed). The birds were randomly selected and placed in one of the four SD treatments of with estimated final densities of 30, 40, 50, or 60 kg/m^2. An additional 5% of birds were placed to account for predicted mortality, allowing for the final target stocking densities to be reached. The number of birds placed in each treatment was calculated according to the final predicted body weight of turkey hens at 11 weeks of age (Aviagen, 2015a; 295, 388, 482, and 571 birds per room for the final predicted SD treatments of 30, 40, 50, and 60 kg/m^2, respectively).

Birds were housed in large open rooms (6.7 x 10.0m = 67.5m^2), with each treatment replicated twice per block, resulting in four replications per treatment. Birds were brooded on wood shavings 7-10 cm thick for the first 10 days, then wheat straw (depth of 10-13 cm) for the rearing period. Brooder rings, 7.0 m in diameter, and heat lamps were used for the first 10 days. Birds were fed *ad libitum* using aluminum tube feeders with a pan diameter of 36 cm for the first 40 d and a pan diameter of 44 cm for the remaining time. Water was provided through Lubing EasyLine™ pendulum turkey nipple drinkers (Lubing, Cleveland, TN). Feeder and drinker space were equalized on a per bird basis for each SD treatment (35 birds/feeder; 30 birds/nipple), thereby eliminating impacts of variable feeder and drinker space. Birds were fed a commercial five-phase diet (Chapter 2; Table 2.1) in specific quantities (kg/bird; see Chapter 2 Table 2.2) and supplemental feeders and drinkers were provided throughout the first ten days. Diet changes were made when the ration was finished, and the total feed amount was adjusted at each diet change to account for mortality.

The standard room temperature curve and lighting program can be seen in Tables 2.3 and 2.4, respectively. LED (light emitting diode) bulbs were used as the light source with daylength starting at 23L:1D (40 lux) and gradually reduced to 18L:6D (5 lux) by day 10. Intact straw bales were provided as environmental enrichment devices (1 bale/90 birds). As straw bales were destroyed, the straw was spread throughout the room and the bale was replaced, thus, no additional litter management was applied.

Air quality was controlled and monitored from day 1 and humidifiers were added during the first 7 days to maintain relative humidity (RH) at approximately 50% (Aviagen, 2015b). Carbon dioxide (CO_2) was measured three times per week using a handheld CO_2 meter (CO240; Extech Instruments; Nashua, NH) and ammonia was monitored once per week until differences were noted, then measured twice per week using ammonia Dräger-Tubes and a handheld pump (Draeger, Inc.; Houston, TX). If CO_2 levels varied by 20% or ammonia differed by 5ppm between rooms, ventilation was adjusted individually in each room in an attempt to match air quality and temperature across all rooms (Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019).

Mortality and morbidity were monitored twice daily (morning and afternoon), and birds were sent for necropsy to an independent pathology lab for identification of cause of illness or death. Birds were culled when necessary due to illness and/or skeletal or growth abnormalities. During the first 3-days, all mortalities and culls were replaced with extra poults in an attempt to maintain the final predicted SD. Birds that were targets of feather pecking and aggression that sustained mild open wounds were treated with pine tar, a deterrent to birds that has anti-microbial properties (Barnes and Greive, 2017), on the affected area. All pine tar treatments were recorded, including the area of the body that was treated. Birds that had more severe wounds were recorded as cull birds and placed in a hospital pen.

In block 1, additional space was blocked off at week 3 to account for high mortality rates during the first 3 weeks of the trial, however, at week 8 space was opened up to maintain the estimated final stocking density for each treatment. In block 2, birds were removed from each treatment at week 9 to account for low mortality rates during block 2, thus the final predicted stocking density could be reached at week 11.

3.3.3 Data Collection

Health Parameters. Heterophil/lymphocyte ratio data was used to measure chronic stress in the birds. Blood was collected from a subsample of 20 birds per replication at 3, 5, 8, and 11 weeks of age from the brachial vein into tubes containing EDTA using vacutainers. Blood smears were prepared on the same day blood was collected. After drying, slides were stored in slide boxes, then stained after each block. Slides were stained with PROTOCOL™ Hema 3™ (Fisher Scientific; Ottawa, Canada) and stored in slide boxes until read. Heterophil/lymphocyte ratios were determined by counting the number of heterophils and lymphocytes within a field of view (Thaxton et al., 2006; Beaulac and Schwean-Lardner, 2018) under 100X oil magnification until a total number of 100 cells were reached (microscope B-290TB; Optika©; Bergamo, Italy).

Bird mobility was tested using a gait scoring technique modified from broilers (Garner et al., 2002) for turkeys by Vermette et al. (2016) (Table 3.1). Thirty birds per replicate were randomly selected and gait scored at 8 and 11 weeks of age. The turkey hens were separated from pen mates and walked down a pathway in the pen in which two individuals scored birds on a six point scale of 0 to 5, where 0 represents no abnormality in gait and 5 represents a complete loss of mobility (Vermette et al., 2016). The two scores were then averaged for each bird. Footpad lesion scores were assessed from the same subsample of 30 birds per replicate at week 8 and week 11. Footpad lesion scoring was conducted by one individual by washing the right footpad with a scrub brush, and then scoring using a method developed by Hocking et al. (2008), as shown in Table 3.2. Using the same subsample of birds, feather cover and cleanliness was assessed. Feather cover was scored on five key areas of the body, including the neck, breast, wings, tail, and back, by one scorer on a scale from 1-4 developed from Davami et al. (1987) and Sarica et al. (2008), as shown in Table 3.3. Total feather cover is expressed as the sum of those five parts (neck, back, wings, tail, and breast) for a maximum score of 20. The feather cleanliness scoring system is shown in Table 3.4.

Litter samples were collected on week 8 and 11 of block 1. Three samples per room were collected by digging a 10x10cm area from the top of the litter to the floor below for each time point. Samples were taken from the front, centre, and back of the room. Samples were not taken from directly below the feeder or drinker lines. Litter was placed into paper bags and weighed before freezing at -18°C until all samples were ready for drying. The week 8 and week 11 samples were placed in an oven (1330GSM Safety Oven; VWR Scientific™; Plainfield, NJ, USA) at 34°C and left for 24 and 36 hours, respectively, after which samples were reweighed to calculate moisture content.

Behaviour Data. Bird activity was recorded using infrared video cameras (Panasonic WV-CF224FX; Panasonic Corporation of North America, Secaucus, NJ) located on the ceiling in each room. Video recordings were taken over a 24-hour period at week 8 and 11 in each room for both blocks. Field of view observations were performed (video playback via Genetec Omnicast

Software, Genetec Inc., Montreal, Canada) using an instantaneous scan sampling technique at 20-minute intervals and the number of birds within the field of view performing each behaviour was recorded (Torrey et al., 2013; Beaulac and Schwean-Lardner, 2018). Behaviours evaluated included those falling into the categories of mobility, comfort and maintenance, exploratory, nutritive, disturbances, and aggression, as defined in the ethogram (Table 3.5).

3.4 Statistical Analyses

The experimental design was a randomized complete block design (RCBD; block as trial) with rooms as the experimental unit. Data from both blocks of this study were analyzed together using regression analysis in SAS (SAS®9.3, Cary, NC, USA) via the Regression Procedure (Proc Reg) and Surface Response Regression Procedure (Proc RSReg) to determine a relationship between stocking density and the health and welfare parameters being evaluated (Beaulac and Schwean-Lardner, 2018). An analysis of variance was performed for all data using the Proc Mixed Procedure (SAS®9.4, Cary, NC, USA) with SD as the fixed factor and block as a random factor. A Tukey's range test was used to separate means. Data were checked for normality (Proc Univariate) and log transformed if needed. If $P \leq 0.05$, differences were considered significant and if $P \leq 0.10$, trends were noted.

Table 3.1. Gait scoring technique modified from broilers (Garner et al., 2002) for turkeys by Vermette et al. (2016)

Score	Degree of impairment		Description
0	None	Original	Smooth, fluid locomotion. The foot is furled while raised.
		Modified	Straight legs.
1	Detectable, but unidentifiable abnormality	Original	The bird is unsteady or wobbles when it walks. However, the problem leg is unclear, or cannot be identified in the first 20s of observation. The bird readily runs from the observer in the pen. The foot may remain flat when raised, but the rest of the stride is fluid and appears unimpaired.
		Modified	Gait appears unstable (shaky or stomping)
2	Identifiable abnormality, that has little impact on overall function	Original	The leg producing the gait defect can be identified within 20s of observation. If a problem leg is identified after 20s of observed locomotor behaviour, then the bird is classed as gait score 1. However, the defect has only a minor impact on biological function. Thus, the bird will run from the observer spontaneously or if touched or nudged with the padded stick. If the bird does not run at full speed, it runs, walks or remains standing for at least 15s after the observer in the pen has ceased to move towards or nudge it. Birds in this, and previous, scores are often observed to scratch their face with their feet-again indicating little impact on function. (The most common abnormality in this score is for the bird to make short, quick, unsteady steps with one leg, where the foot remains flat during the step.)
3	Identifiable abnormality which impairs function	Original	Although the bird will move away from the observer when approached or touched, or nudged, it will not run, and squats within 15s or less of the observer in the pen ceasing to approach or nudge it. If the bird squats after 15s have elapsed, it is classified as gait score 2.
4	Severe impairment of function, but still capable of walking	Original	The bird remains squatting when approached or nudged. This criterion is assessed by approaching the bird, and if it remains squatting, gently nudging or touching the animal for 5s. Animals may appear to rise but still resting upon their hocks. Only rising to stand on both feet within 5s of handling is counted—a bird which takes longer than 5s to rise, or which does not rise at all is scored as 4, while a bird that rises in 5s or less is counted as a 3 (or lower if its gait is good). Nevertheless, the bird can walk when picked up by the observer and placed in a standing position, but squats immediately following one or two steps. (Squatting often involves a characteristic ungainly backwards fall.)
		Modified	Bird requires wings for balance.
5	Complete lameness	Original	The bird cannot walk, and instead may shuffle along on its hocks. It may attempt to stand when approached but is unable to do so, and when placed on feet unable to complete a step with one or both legs.

Table 3.2. Footpad dermatitis (FPD) scoring system (Hocking et al., 2008)

Score	Description of Foot Pad
0	No external signs of FPD. The skin of the footpad feels soft to the touch and no swelling or necrosis is evident.
1	The pad feels harder and denser than a non-affected foot. The central part of the pad is raised, reticulate scales are separated, and small black necrotic areas may be present.
2	Marked swelling of the footpad. Reticulate scales are black, forming scale shaped necrotic areas. The scales around the outside of the black areas may have turned white. The area of necrosis is less than one quarter of the total area of the footpad.
3	Swelling is evident, and the total footpad size is enlarged. Reticulate scales are pronounced, increased in number and separated from each other. The amount of necrosis extends to one half of the footpad.
4	As score 3, but with more than half the footpad covered by necrotic cells.

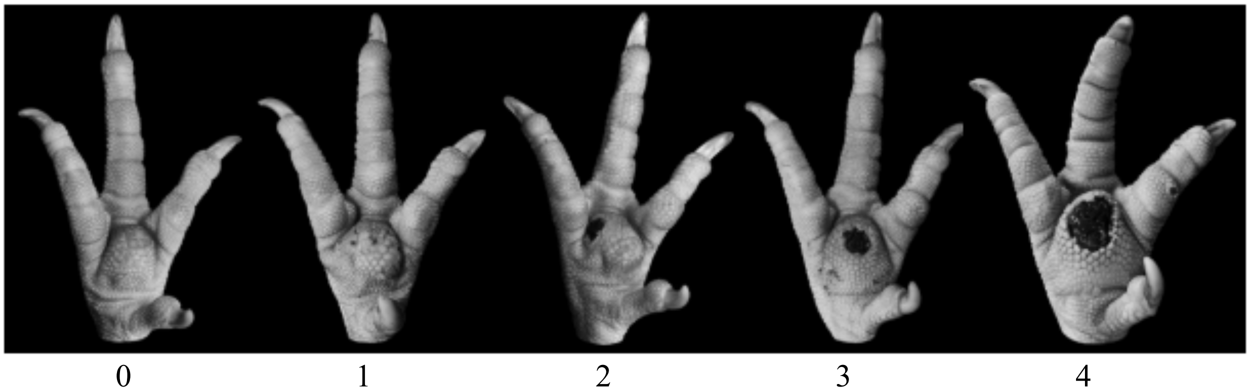

Figure 3.1. Photographs of footpad lesions in turkeys. The lesions for scores 2, 3 and 4 are the minimum size required for each score based on the proportion of the footpad area that is covered by necrotic cells indicated by a dark scab. (Hocking et al., 2008)

Table 3.3. Feather scoring technique developed from Davami et al. (1987) and Sarica et al. (2008)

Score	Description[1]
1	No feather cover
2	More than half of the plumage is missing.
3	Few or less than half of the plumage is missing.
4	Full, intact plumage.

[1]Each area was scored by two individuals and then averaged. The areas of the body scored include the neck, breast, wings, tail, and back. A total of these scores from each area of the body is the total feather condition score.

Table 3.4. Feather cleanliness scoring developed from Forkman and Keeling (2009) as adapted from Wilkins et al. (2003)

Score	Description
1	Very clean – more than 75% of the body feathers free from soiling
2	Moderately clean – 50-75% of the body feather are free from soiling
3	Moderately dirty – 25-50% of the body feathers are free from soiling
4	Very dirty – less than 25% of the body feathers are free from soiling

Table 3.5. Behavioural ethogram for turkey toms, as modified from Martrenchar et al. (1999) and Vermette et al. (2016)

Behaviour	Description of Behaviour
Feeding	Standing or sitting with head in the feeder.
Drinking	Standing or sitting with head in the drinker.
Resting	Lying down, not performing any other behaviour. May or may not be sleeping.
Standing	Standing, not performing any other behaviour.
Walking	Bird walking or running. Must take 2 or more consecutive steps.
Fighting	Two or more individuals, where at least one bird is posturing with head back and breast thrust forward. May or may not include one individual running or jumping at the other.
Preening	Manipulating own feathers with the beak while standing or resting.
Stretching	Extension of the wings and/or legs.
Wing Flapping	Flapping both wings.
Dust Bathing	Fluttering movement of the bird in a lying position on the litter while pulling the loose substrate close to the body and into the feathers.
Feather Ruffle	Full body shake while standing or resting.
Environmental Pecking	Pecking at walls, feeder tubes (not feed pan), drinker lines (away from the drinker cups), or litter while standing or resting.
Feather Pecking	Pecking at a pen mate's feathers while standing or resting. The pen mate typically does not move away.
Aggressive Pecking	Forceful pecking at a pen mate's head, body, or snood while standing or resting. The pen mate typically moves away.
Moderate Disturbance	A bird in a laying posture opens its eyes, lifted its head or moved its body as a result of another bird walking in front of it, on top of it, touching it, or flapping near it.
Severe Disturbance	A bird in a lying posture stands up as a result of another bird walking in front of it, on top of it, or flapping near it.

3.5. Results

3.5.1 Mobility and footpad scores

At 8 weeks of age, SD treatments did not impact turkey hen mobility, as measured by gait scoring (Table 3.6). At 11 weeks of age, no relationship was observed between SD and average gait scores, however, a larger percentage of tested birds scored in the gait score category 2 (identifiable abnormality, that has little impact on overall function) in the 60 kg/m^2 treatment compared to the 30 kg/m^2 treatment (23.75% vs 12.50%, respectively; ANOVA; P=0.03).

Table 3.6. Effect of estimated final stocking density on turkey hen gait scores expressed as the percentage of sampled birds in each scoring category (scale 0-5[1]) and average gait scores at 8 and 11 weeks of age

% In category	n	Stocking density (kg/m^2)				SEM[2]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[3]
		30	40	50	60					
Week 8 gait scores										
0	4	72.92	77.92	74.17	61.25	3.659	0.44	0.25	0.23	-
1	4	18.75	18.75	22.50	29.58	2.573	0.41	0.12	0.50	-
2	4	7.50	3.33	3.33	8.33	1.299	0.40	0.84	0.09	-
3	4	0	0	0	0.83	0.208	0.42	0.19	0.32	-
4	4	0.83	0	0	0	0.208	0.43	0.19	0.32	-
5	4	0	0	0	0	-	-	-	-	-
Average Score	4	0.37	0.25	0.29	0.49	0.051	0.42	0.41	0.13	-
Week 11 gait scores										
0	4	59.17	47.50	43.33	33.75	7.048	0.10	0.21	0.94	-
1	4	28.33	35.42	42.92	41.67	4.606	0.43	0.26	0.66	-
2	4	12.50[b]	16.25[ab]	13.75[ab]	23.75[a]	3.265	0.03	0.30	0.65	-
3	4	0	0.83	0	0	0.208	0.43	0.67	0.35	-
4	4	0	0	0	0	-	-	-	-	-
5	4	0	0	0	0.83	0.208	0.43	0.19	0.32	-
Average Score	4	0.53	0.70	0.70	0.93	0.098	0.60	0.18	0.89	-

[1]Score of 0= no impairment and 5=complete lameness (adapted from Garner et al., 2002 by Vermette et al., 2016).
[2]Standard error of the mean; [3]Regression considered significant if P≤0.05.
[a,b] Means with common letter within a row do not differ significantly (P≤0.05).

Average footpad scores and the percentage of birds that scored within each category (scale 0-4) are shown in Table 3.7. At 8 weeks of age, average footpad scores increased linearly with increasing density (1.43, 1.55, 1.86, 1.80 for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; *P*=0.03), indicating more severe lesions at high SD. The percentage of birds in the score of 0 category increased with decreasing density (linear; *P*=0.02), thus indicating more birds in the 30 kg/m^2 showed no external signs of a lesion compared to the highest SD. The percentage of birds in the score of 2 category (lesion is less than one quarter of the total area of the footpad) increased linearly as SD increased (*P*=0.03). At 11 weeks of age, no significant regression relationships between footpad score categories or average footpad scores and SD were observed. However, average footpad scores were higher in the 60 kg/m^2 treatment (2.83) compared to the 30 kg/m^2 (2.02) and 40 kg/m^2 treatments (2.18) (ANOVA; *P*=0.01).

Table 3.7. Effect of estimated final stocking density on turkey hen footpad scores expressed as the percentage in each scoring category (scale 0-4[1]) and average footpad score at 8 and 11 weeks of age

% In category	n	Stocking density (kg/m^2)				SEM[2]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[3]
		30	40	50	60					
Week 8 footpad scores										
0	4	15.83	10.00	2.50	2.50	2.332	0.11	0.02	0.48	Y = -0.48x+29.08
1	4	34.17	39.17	23.33	30.00	3.206	0.36	0.34	0.90	-
2	4	40.83^{b}	37.50^{b}	61.67^{a}	53.33^{ab}	3.319	<0.01	0.03	0.68	Y = 0.62x+20.58
3	4	9.17	12.50	10.83	13.33	2.018	0.91	0.56	0.92	-
4	4	0	0.83	1.67	0.83	0.373	0.52	0.33	0.28	-
Average Score	4	1.43	1.55	1.86	1.80	0.075	0.13	0.03	0.52	Y = 0.014x+1.03
Week 11 footpad scores										
0	4	10.83	8.33	0.83	3.33	2.184	0.10	0.13	0.57	-
1	4	29.17^{a}	22.50^{ab}	18.33^{ab}	5.00^{b}	5.543	0.05	0.13	0.76	-
2	4	22.50	30.00	23.33	26.67	2.866	0.70	0.83	0.74	-
3	4	22.50	21.67	35.00	35.00	4.503	0.18	0.22	0.96	-
4	4	15.00	17.50	22.50	30.00	5.045	0.22	0.28	0.81	-
Average Score	4	2.02^{b}	2.18^{b}	2.60^{ab}	2.83^{a}	0.223	0.01	0.16	0.93	-

[1]Score 0 = no external signs of a lesion, score 4 = greater than 50% of the footpad covered with necrotic cells (Hocking et al., 2008).
[2]Standard error of the mean.
[3]Regression considered significant if P≤0.05.
[a,b] Means with common letter within a row do not differ significantly (P≤0.05).

3.5.2 Feather cover and cleanliness

Table 3.8 shows the percentage of birds that fell within each feather cover score category (scale of 1-4) for the neck, breast, wing, tail, and back at 8 weeks of age. No differences were observed for neck feather scores. Breast feather cover was significantly reduced in the 60 kg/m^2 treatment as more birds fell in the score of 1 (no feather cover) or 2 (more than 50% of plumage is missing) (linear; P=0.03 and P=0.02, respectively). More birds in the lowest density treatment scored a 4 (full intact plumage) for breast feather cover (linear; P=0.02). No differences were observed for wing feather scores with increasing density. Tail feather cover followed a similar pattern to breast feather cover as tail cover was poorer in the birds of the highest density treatment as more birds scored a 1 or 2 (linear; P=0.01 and P<0.01, respectively) and the percentage of birds that scored a 4 was highest in the lowest SD (linear; P<0.01). Back feather cover scores were indicative of good feather condition. Only 3.33% of birds in the highest SD scored a 3 (less than 50% of the plumage is missing; linear; P=0.05), and 100% of birds in the 30, 40, and 50 kg/m^2 treatments and 96.67% of birds in the 60 kg/m^2 treatment scored a 4 (full intact plumage; linear; P=0.05).

Table 3.8. Effect of estimated final stocking density turkey hen feather cover score (% of birds that scored between 1-4[1]) and feather cleanliness score (% of birds that scored between 1-4[2]) at 8 weeks of age

% In category	n	Stocking density (kg/m^2) 30	40	50	60	SEM[3]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[4]
Neck feather score										
1	4	0	0	0	0	0	-	-	-	-
2	4	0	0	0	0	0	-	-	-	-
3	4	3.33	3.33	2.50	1.67	0.872	0.87	0.47	0.82	-
4	4	96.67	96.67	97.50	98.33	0.872	0.87	0.47	0.82	-
Breast feather score										
1	4	0.83	0.83	5.00	8.33	1.390	0.16	0.03	0.51	Y = 0.27x-8.25
2	4	25.00^{ab}	13.33^{b}	35.00^{ab}	44.17^{b}	4.085	0.03	0.02	0.14	Y = 0.79x-6.25
3	4	53.33	62.50	42.50	42.50	3.502	0.11	0.09	0.50	-
4	4	20.83	23.33	17.50	5.00	2.739	0.06	0.02	0.11	Y = -0.53x+40.67
Wing feather score										
1	4	0	0	0	0	0	-	-	-	-
2	4	0	0	0	0	0	-	-	-	-
3	4	0.83	1.67	8.33	7.50	1.969	0.25	0.13	0.83	-
4	4	99.17	98.33	91.67	92.50	1.969	0.44	0.13	0.83	-
Tail feather score										
1	4	0	0	1.67	6.67	1.049	0.06	0.01	0.16	Y = 0.22x-7.67
2	4	14.17^{b}	8.33^{b}	31.67^{ab}	42.50^{b}	4.425	<0.01	<0.01	0.21	Y = 1.08x-24.58
3	4	65.83^{ab}	80.83^{a}	63.33^{ab}	49.17^{b}	4.270	0.05	0.08	0.06	-
4	4	20.00^{a}	10.83^{ab}	3.33^{b}	1.67^{b}	2.613	0.02	<0.01	0.36	Y = -0.63x+37.08
Back feather score										
1	4	0	0	0	0	0	-	-	-	
2	4	0	0	0	0	0	-	-	-	
3	4	0	0	0	3.33	0.569	0.06	0.05	0.10	Y = 0.10x-3.67
4	4	100.0^{a}	100.0^{a}	100.0^{a}	96.67^{b}	0.569	0.05	0.05	0.10	Y = -0.10x+103.67
Cleanliness score										
1	4	42.50^{a}	38.33^{a}	5.83^{b}	1.67^{b}	5.868	<0.01	<0.01	1.00	Y = -1.55x+91.83
2	4	45.83	45.83	35.00	21.67	3.859	0.06	0.01	0.30	Y = -0.83x+74.58
3	4	11.67^{b}	15.00^{b}	48.33^{a}	36.67^{ab}	5.109	<0.01	0.01	0.40	Y = 1.08x-20.83

4	4	0[c]	0.83[c]	10.83[b]	40.00[a]	5.977	0.04	0.01	0.15	Y = 1.30x-45.58

[1]Score of 1=no feather cover, 2= greater than 50% of the plumage is missing, 3= less than 50% of the plumage is missing, and 4=full intact plumage (Davami et al., 1987 and Sarica et al., 2008); [2]Score of 1= very clean, 2= moderately clean, 3=moderately dirty, and 4= very dirty (Forkman and Keeling (2009) as modified from Wilkins et al., 2003).

[3]Standard error of the mean;

[4]Regression considered significant if $P \leq 0.05$

[a,b] Means with common letter within a row do not differ significantly ($P \leq 0.05$).

Table 3.9 shows the percentage of birds that fell within each feather cover score category (scale of 1-4) for the neck, breast, wing, tail, and back at 11 weeks of age. Birds' neck feather cover scores were unaffected by SD. Breast feather cover was poorer in the highest density as more birds scored a 1 (no feather cover) (linear; *P*=0.02). For wing feather cover, a linear effect was noted with the highest percentage of birds scoring 2 (more than 50% of plumage missing) and 3 (less than 50% of plumage missing) in the 60 kg/m^2 treatment (*P*=0.05 and *P*<0.01, respectively). Wing feather cover was better as density decreased with more birds scoring a 4 (full intact plumage; linear; *P*<0.01). Tail feather cover was poorer in the 60 kg/m^2 treatment as more birds scored a 1 and 2 (linear, *P*<0.01 and quadratic *P*<0.01, respectively). Thus, tail feather cover was significantly better as SD decreased, with more birds scoring a 3 and 4 (linear; *P*<0.01 and *P*=0.01, respectively). For back feather scores, a linear effect was observed with the highest percentage of birds scoring a 1 and 3 in the 60 kg/m^2 treatment (linear; *P*=0.05 and *P*=0.04, respectively). There was also a linear tendency for more birds to score a 2 in the 60 kg/m^2 treatment (*P*=0.06) for back feather scores and therefore, as density decreased, back feather cover improved as a higher percentage of birds scored a 4 (linear; *P*=0.03).

Feather cleanliness scores (scale of 1-4, where 1 is very clean and 4 is very dirty) were recorded for the whole body of the bird. The percentage of birds that scored within each score category can be seen in Tables 3.8 and 3.9 for week 8 and 11, respectively. At week 8, more birds were very clean (score 1) and moderately clean (score 2) as SD decreased (linear; *P*<0.01 and *P*=0.01, respectively). Therefore, more birds were moderately dirty (score 3) and very dirty (score 4) as SD increased (linear; *P*=0.01 for both), indicating that birds were dirtier in the higher density treatments at week 8. The same pattern was observed for feather cleanliness at 11 weeks of age (Table 3.9). More birds were very clean (score 1) and moderately clean (score 2) as SD decreased (quadratic, *P*=0.01 and linear, P<0.01, respectively) and more birds were moderately dirty (score 3) and very dirty (score 4) as SD increased (quadratic; *P*<0.01 and *P*=0.02, respectively).

Table 3.9. Effect of estimated final stocking density turkey hen feather cover score (% of birds that scored between 1-4[1]) and feather cleanliness score (% of birds that scored between 1-4[2]) at 11 weeks of age

% In category	n	Stocking density (kg/m^2) 30	40	50	60	SEM[3]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[4]
Neck feather score										
1	4	0	0	0	0	0	-	-	-	-
2	4	4.17	12.50	11.67	8.33	1.936	0.42	0.52	0.15	-
3	4	20.00	26.67	25.83	27.50	3.088	0.83	0.45	0.71	-
4	4	75.83	60.83	62.50	64.17	4.156	0.62	0.39	0.34	-
Breast feather score										
1	4	0.83^{b}	14.17^{ab}	18.33^{ab}	33.33^{a}	4.944	0.05	0.02	0.92	$Y = 1.02x-29.08$
2	4	60.83	58.33	53.33	50.00	3.635	0.73	0.26	0.96	-
3	4	33.33^{a}	25.83^{ab}	26.67^{ab}	15.00^{b}	4.082	0.03	0.14	0.80	-
4	4	5.00	1.67	1.67	1.67	0.938	0.38	0.25	0.39	-
Wing feather score										
1	4	0	0	0	0	0	-	-	-	-
2	4	0	0	0	1.67	3.635	0.07	0.05	0.10	$Y = 0.05x-1.83$
3	4	2.50^{b}	2.50^{b}	7.50^{ab}	16.67^{a}	4.082	0.01	<0.01	0.16	$Y = 0.48x-14.08$
4	4	97.50^{a}	97.50^{a}	92.50^{ab}	81.67^{b}	0.938	0.01	<0.01	0.10	$Y = -0.53x+115.92$
Tail feather score										
1	4	0.83^{c}	9.17^{bc}	15.00^{b}	35.83^{a}	3.613	<0.001	<0.01	0.07	$Y = 1.11x-34.67$
2	4	11.67^{b}	43.33^{a}	56.67^{a}	58.33^{a}	5.327	<0.001	<0.01	<0.01	$Y = -0.075x^2+8.28x-169.00$
3	4	76.67^{a}	45.83^{b}	28.33$_{bc}$	5.83^{c}	7.217	<0.001	<0.01	0.50	$Y = -2.30x+142.67$
4	4	10.83^{a}	1.67^{ab}	0^{b}	0^{b}	1.652	0.02	0.01	0.10	$Y = -0.34x+18.50$
Back feather score										
1	4	0	0	0	1.67	0.285	0.07	0.05	0.10	$Y = 0.050x-1.83$
2	4	0	0	0	5.00	0.907	0.09	0.06	0.13	-
3	4	0^{b}	0^{b}	0.83^{ab}	13.33^{a}	2.287	0.05	0.04	0.13	$Y = 0.41x-14.83$
4	4	100.00^{a}	100.00^{a}	99.17^{ab}	80.00^{b}	3.248	0.03	0.03	0.09	$Y = -0.61+122.17$
Cleanliness score										
1	4	26.67^{a}	3.33^{b}	0^{b}	0.83^{b}	3.425	<0.01	<0.01	0.01	$Y = 0.060x^2-6.25x+158.88$
2	4	67.50^{a}	57.50^{ab}	29.17^{bc}	25.83^{c}	5.553	<0.01	<0.01	0.65	$Y = -1.53x+114.00$
3	4	5.83^{c}	38.33^{b}	61.67^{a}	45.83^{ab}	5.651	<0.001	<0.01	<0.01	$Y = -0.12x^2+12.31x-256.17$

4	4	0[b]	0.83[b]	9.17[b]	27.50[a]	3.223	<0.01	<0.01	0.02	$Y = 0.044x^2-3.03x+51.63$

[1]Score of 1=no feather cover, 2= greater than 50% of the plumage is missing, 3= less than 50% of the plumage is missing, and 4=full intact plumage (Davami et al., 1987 and Sarica et al., 2008); [2]Score of 1= very clean, 2= moderately clean, 3=moderately dirty, and 4= very dirty (Forkman and Keeling (2009) as modified from Wilkins et al., 2003).

[3]Standard error of the mean;

[4]Regression considered significant if $P \leq 0.05$

[a,b] Means with common letter within a row do not differ significantly ($P \leq 0.05$).

At 8 weeks of age, total feather cover (sum of five parts; max score of 20) linearly decreased as SD increased (17.96, 18.06, 17.30, 16.78 for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; $P<0.01$; Table 3.10). Overall, at 11 weeks of age, total feather cover linearly decreased as SD increased (17.09, 16.01, 15.68, 14.63 for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; $P<0.01$). The average feather cleanliness scores worsened in a linear manner as SD increased at 8 weeks of age (1.69, 1.78, 2.64, 3.15 for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; $P<0.01$). At 11 weeks of age, the average feather cleanliness scores were 1.79, 2.37, 2.80, and 3.00 for SD treatments 30, 40, 50, and 60 kg/m^2, respectively, which also shows that cleanliness worsened in a linear manner as SD increased at 11 weeks of age ($P<0.01$).

Table 3.10. Effect of estimated final stocking density on turkey hen overall feather cover (scale 1-4[1]) and cleanliness scores (scale 1-4[2]) at 8 and 11 weeks of age

Week	n	Stocking density (kg/m^2)				SEM[3]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[4]
		30	40	50	60					
Total feather cover score (sum of five parts)[5]										
8	4	17.96^{a}	18.06^{a}	17.30^{ab}	16.78^{b}	0.159	<0.01	<0.01	0.13	Y = -0.043x+19.46
11	4	17.09^{a}	16.01^{ab}	15.68^{bc}	14.63^{c}	0.263	<0.01	<0.01	0.96	Y = -0.077x+19.33
Average feather cleanliness score										
8	4	1.69^{b}	1.78^{b}	2.64^{a}	3.15^{a}	0.181	<0.01	<0.01	0.33	Y = 0.052x-0.038
11	4	1.79^{c}	2.37^{b}	2.80^{a}	3.00^{a}	0.129	<0.001	<0.01	0.09	Y = 0.041x+0.66

[1]Score of 1=no feather cover, 2= greater than 50% of the plumage is missing, 3= less than 50% of the plumage is missing, and 4=full intact plumage (Davami et al., 1987 and Sarica et al., 2008).
[2] Score of 1= very clean, 2= moderately clean, 3=moderately dirty, and 4= very dirty (Forkman and Keeling (2009) as modified from Wilkins et al., 2003).
[3]Standard error of the mean.
[4] Regression considered significant if $P \leq 0.05$.
[5]Sum of five parts for a max of 20: neck, back, wings, tail, breast, scored on a scale of 1-4.

2.5.3 Incidence of aggressive damage

The overall percentage of birds treated by location, time period, and the percentage of birds treated by time period that included culls for aggressive damage can be seen in Table 3.11. The percentage of birds treated for aggressive damage on the tail (P<0.01) and the head (P=0.05) linearly increased with decreasing density. There was a tendency for aggressive damage to the neck to be highest in the 40 kg/m^2 treatment and lowest in the 60 kg/m^2 treatment (quadratic; P=0.07). A quadratic tendency was noted for skin tears to be highest in the middle SD (40 and 50 kg/m^2; P=0.07). When evaluating by time period, during week 5-8 and 8-11, incidence of aggressive damage was highest in the lowest SD of 30 kg/m^2 (linear; P<0.01 for both). The total percentage of birds treated for aggressive damage from 0-11 weeks of age was 7.20, 7.02, 3.53, and 2.58% for SD treatments 30, 40, 50, and 60 kg/m^2, respectively (linear; P<0.01). The total percentage of birds treated for aggressive damage plus culls related to aggressive damage from 0-11 weeks of age linearly increased as SD decreased (7.88, 7.60, 4.05, and 2.67% for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; P<0.01).

Table 3.11. Effect of estimated final stocking density on the incidence and location of aggressive damage and skin tears (% of birds placed) treated with a deterrent up to 11 weeks of age

Location	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
% of birds treated with a deterrent by location										
Tail	4	2.96	2.13	0.73	0.66	0.381	0.08	<0.01	0.51	Y = -0.083x+5.37
Wing	4	0.76	0.90	0.41	0.74	0.158	0.75	0.52	0.86	
Back	4	0.08	0.06	0	0.04	0.027	0.77	0.48	0.58	
Neck	4	0.42	0.58	0.47	0.13	0.096	0.12	0.12	0.07	
Head	4	2.88	2.84	1.50	0.74	0.455	0.20	0.05	0.49	Y = -0.077x+5.48
Snood	4	0.08	0.26	0.26	0.26	0.067	0.78	0.52	0.32	
Skin tear	4	0	0.26	0.16	0	0.056	0.29	0.84	0.07	
Total	4	7.20[a]	7.02[a]	3.53[ab]	2.58[b]	0.829	0.01	<0.01	0.45	Y = -0.17x+12.89
% of birds treated with a deterrent by time period										
Week 0-3	4	0	0	0	0.04	0.011	0.43	0.19	0.32	
Week 3-5	4	1.95	2.13	1.19	1.05	0.243	0.29	0.09	0.81	
Week 5-8	4	2.88[a]	2.64[ab]	1.14[ab]	1.01[b]	0.327	0.03	<0.01	0.73	Y = -0.071x+5.12
Week 8-11	4	2.88[a]	2.26[ab]	1.20[ab]	0.48[b]	0.463	0.03	<0.01	0.54	Y = -0.08x+5.42
Week 0-11	4	7.20[a]	7.02[a]	3.53[ab]	2.58[b]	0.829	0.01	<0.01	0.45	Y = -0.17x+12.89
% of birds treated with a deterrent plus all culls related to aggressive damage by time period										
Week 0-3	4	0	0	0	0.04	0.011	0.43	0.19	0.32	
Week 3-5	4	1.95	2.19	1.19	1.09	0.240	0.26	0.09	0.82	
Week 5-8	4	2.97[a]	2.71[a]	1.14[ab]	1.05[b]	0.343	0.03	<0.01	0.77	Y = -0.073x+5.26
Week 8-11	4	3.47[a]	2.71[a]	1.71[ab]	0.48[b]	0.506	<0.01	<0.01	0.22	Y = -0.10x+6.58
Week 0-11	4	7.88[a]	7.60[a]	4.05[ab]	2.67[b]	0.883	<0.01	<0.01	0.34	Y = -0.19x+14.18

[1]Standard error of the mean.

[2]Regression considered significant if P≤0.05.

[a,b] Means with common letter within a row do not differ significantly (P≤0.05).

The incidence of aggressive damage and skin tears (% of birds placed) by location of damage from 0-3, 3-5, 5-8, and 8-11 weeks of age can be seen in Table 3.12. During week 0-3 and 3-5, SD did not impact incidence of aggressive damage. During week 5-8, incidence of aggressive damage to the tail was highest in the 30 kg/m^2 treatment (linear; $P<0.01$). A quadratic relationship resulted for aggressive damage to the neck, with the middle densities (40 and 50 kg/m^2) having the highest incidence ($P=0.04$). During week 8-11, a quadratic relationship is observed for incidence of aggressive damage to the tail, with the 30 and 60 kg/m^2 treatments having the highest incidence ($P=0.03$). Incidence of aggressive damage to the snood linearly increased with decreasing SD ($P=0.03$) during week 8-11.

Table 3.12. Effect of estimated final stocking density on the incidence of aggressive damage and skin tears (% of birds placed) by location of damage from 0-3, 3-5, 5-8, and 8-11 weeks of age

% tarred in each location	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
Week 0-3										
Tail	4	0	0	0	0.04	0.011	0.43	0.19	0.32	-
Wing	4	0	0	0	0	0	-	-	-	-
Back	4	0	0	0	0	0	-	-	-	-
Neck	4	0	0	0	0	0	-	-	-	-
Head	4	0	0	0	0	0	-	-	-	-
Snood	4	0	0	0	0	0	-	-	-	-
Skin tear	4	0	0	0	0	0	-	-	-	-
Week 3-5										
Tail	4	1.10	1.48	0.47	0.35	0.221	0.35	0.12	0.64	-
Wing	4	0.17	0.13	0	0.22	0.042	0.31	0.99	0.15	-
Back	4	0	0	0	0	0	-	-	-	-
Neck	4	0.25	0.06	0.26	0.13	0.060	0.67	0.80	0.80	-
Head	4	0	0.13	0.05	0.22	0.047	0.41	0.26	0.77	-
Snood	4	0.42	0.32	0.41	0.13	0.109	0.83	0.45	0.69	-
Skin tear	4	0	0	0	0	0	-	-	-	-
Week 5-8										
Tail	4	1.36[a]	0.58[ab]	0.16[b]	0.13[b]	0.175	<0.01	<0.01	0.10	$Y = -0.04x+2.40$
Wing	4	0.42	0.71	0.36	0.44	0.113	0.72	0.64	0.65	-
Back	4	0	0.06	0	0.04	0.019	0.58	0.69	0.82	-
Neck	4	0[b]	0.32[a]	0.05[ab]	0[b]	0.046	0.01	0.52	0.04	$Y=-0.9e^{-4}x^2+0.08x-1.56$
Head	4	0.08	0.06	0.05	0.04	0.029	0.98	0.65	0.94	-
Snood	4	1.02	0.64	0.36	0.35	0.173	0.57	0.17	0.76	-
Skin tear	4	0	0.26	0.16	0	0.056	0.29	0.84	0.07	-
Week 8-11										

Tail	4	0.68[a]	0.06[b]	0.10[ab]	0.13[ab]	0.088	0.04	0.04	0.03	$Y = 0.16e^{-2}x^2-0.16x+4.01$
Wing	4	0.25	0.06	0.05	0.09	0.039	0.25	0.10	0.32	-
Back	4	0.08	0	0	0	0.021	0.43	0.19	0.32	-
Neck	4	0.08	0.19	0.16	0	0.033	0.15	0.22	0.22	-
Head	4	0	0.06	0.16	0	0.031	0.25	0.76	0.08	-
Snood	4	1.78	1.87	0.73	0.26	0.300	0.11	0.03	0.48	$Y = -0.057x+3.72$
Skin tear	4	0	0	0	0	0	-	-	-	-

[1]Standard error of the mean.

[2]Regression considered significant if $P \leq 0.05$.

[a, b] Means with common letter within a row do not differ significantly ($P \leq 0.05$).

3.5.4 Heterophil/Lymphocyte Ratio

Turkey hen H/L ratio was not affected by SD at 3 weeks of age (Table 3.13). At 5 weeks of age, H/L ratio linearly increased as SD increased (0.76, 0.85, 0.88, 0.89 for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; P<0.01). At 8 weeks of age, H/L ratio in hens demonstrated a quadratic relationship with the 40 and 30 kg/m^2 treatments having the highest ratios (1.09, 1.4, 0.83, 0.89 for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; P=0.03). At 11 weeks of age, H/L ratios were 0.87, 0.96, 1.13, 1.06 for SD treatments 30, 40, 50, and 60 kg/m^2 respectively, (P<0.01) demonstrating a quadratic relationship.

Table 3.13. Effect of estimated final stocking density on turkey hen heterophil/lymphocyte ratio at 3, 5, 8, and 11 weeks of age

Age (weeks)	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
3	4	0.96	1.06	0.99	1.05	0.033	0.63	0.51	0.73	-
5	4	0.76[b]	0.85[ab]	0.88[a]	0.89[a]	0.017	0.02	<0.01	0.18	$Y = 0.42e^{-2}x+0.65$
8	4	1.09[b]	1.40[a]	0.83[c]	0.89[bc]	0.032	<0.01	<0.01	0.03	$Y = -0.67e^{-4}x^{2}+0.05x+0.31$
11	4	0.87[c]	0.96[bc]	1.13[a]	1.06[ab]	0.021	<0.01	<0.01	0.05	$Y = -0.41e^{-3}x^{2}+0.04-0.09$

[1]Standard error of the mean.

[2]Regression considered significant if $P \leq 0.05$.

[a,b] Means with common letter within a row do not differ significantly ($P \leq 0.05$).

3.5.5 Behaviour

The behaviour of turkey hens (percentage of birds performing various behaviours based on the percentage of birds within a field of view) 8 weeks of age is shown in Table 3.14. The percentage of birds present at the feeder linearly increased as SD increased (4.63, 3.81, 8.61, and 6.25%, for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; P=0.04). The percentage of birds resting linearly increased as SD increased, with birds in the 60 kg/m^2 treatment resting the most (P<0.01). Therefore, the opposite effects were seen for standing and walking behaviour, where the percentage of birds standing (P=0.02), and walking (P<0.01) linearly decreased with increasing density. The percentage of birds litter pecking linearly increased with decreasing SD (3.20, 1.21, 2.29, 0.97% for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; P=0.03). The percentage of birds dustbathing was highest in the 50 kg/m^2 treatment (quadratic; P<0.01) and the percentage birds hens head scratching also demonstrated a quadratic relationship with the 40 and 50 kg/m^2 treatments having the most birds performing the behaviour (P=0.01), however, these are low incidence behaviours. The percentage of birds that experienced severe disturbances linearly increased as SD increased (P=0.01). The total incidence of disturbances, which included moderate and severe disturbances, linearly increased with increasing density (P=0.05). The percentage of hens fighting linearly increased with decreasing density (P=0.01) and aggressive pecking behaviour demonstrated a quadratic relationship with SD, with birds in the 30 kg/m^2 treatment performing that behaviour more (P=0.02). The percentage of birds performing aggressive behaviours, which includes fighting and aggressive pecking, also demonstrated a quadratic relationship with SD, with the highest aggression seen in the lowest SD treatment (0.60, 0.34, 0.11, 0.19 % for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; P=0.01).

Table 3.14. Effect of estimated final stocking density on percentage of turkey hens performing various behaviours (% of birds within the field of view) at 8 weeks of age

Behaviour	n	Stocking density (kg/m^2)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
Nutritive behaviours										
Feeding	4	4.63^{ab}	3.81^{b}	8.61^{a}	6.25^{ab}	0.652	0.03	0.04	0.60	$Y = 0.10x+1.48$
Drinking	4	2.85	2.69	2.43	1.69	0.259	0.36	0.18	0.80	-
Mobility behaviours										
Standing	4	15.01	14.48	13.71	12.30	0.451	0.10	0.02	0.90	$Y = -0.09x+17.88$
Walking	4	10.06^{a}	6.30^{bc}	8.25^{ab}	5.11^{c}	0.504	<0.01	<0.01	0.96	$Y = -0.13x+13.24$
Resting	4	57.89^{b}	64.52^{a}	59.74^{ab}	67.05^{a}	1.169	<0.01	<0.01	0.54	$Y = 0.23x+52.08$
Exploratory behaviours										
Litter pecking	4	3.20^{a}	1.21^{b}	2.29^{ab}	0.97^{b}	0.302	0.02	0.03	0.48	$Y = -0.06x+4.44$
Environmental pecking	4	0.82	1.25	0.57	0.72	0.108	0.48	0.88	0.85	-
Feather pecking	4	0.51	0.51	0.43	0.49	0.031	0.98	0.91	0.98	-
Comfort and maintenance behaviours										
Preening	4	2.82	3.37	2.47	3.21	0.167	0.06	0.32	0.53	-
Stretching	4	0.25	0.12	0.19	0.17	0.021	0.15	0.95	0.25	-
Wing flapping	4	0.29	0.23	0.29	0.17	0.031	0.78	0.51	0.67	-
Dustbathing	4	0.00^{b}	0.02^{ab}	0.05^{a}	0.00^{b}	0.007	0.01	0.63	<0.01	$Y = -0.16e^{-3}x^2+0.02x-0.30$
Perching	4	0.06	0.00	0.00	0.12	0.027	0.34	0.37	0.11	-
Head scratching	4	0.01^{b}	0.07	0.06	0.03	0.009	0.05	0.49	0.01	$Y = -0.21e^{-3}x^2+0.02x-0.37$
Feather ruffle	4	0.18	0.11	0.28	0.08	0.032	0.09	0.99	0.22	-
Total comfort & maintenace[3]	4	0.73	0.55	0.87	0.45	0.060	0.16	0.81	0.25	-
Aggressive behaviours										
Fighting	4	0.30^{a}	0.16^{ab}	0.00^{b}	0.07^{ab}	0.038	0.01	0.01	0.12	$Y = -0.86e^{-2}x+0.52$
Aggressive pecking	4	0.30^{a}	0.18^{ab}	0.11^{b}	0.13^{ab}	0.028	0.02	0.05	0.02	$Y = 0.34e^{-3}x^2-0.04x+1.09$
Total aggression[4]	4	0.60^{a}	0.34^{ab}	0.11^{b}	0.19^{b}	0.055	<0.01	<0.01	0.01	$Y = 0.86e^{-3}x^2-0.09x+2.59$
Disturbances										

Moderate disturbances	4	0.39	0.26	0.27	0.27	0.033	0.94	0.81	0.84	-
Severe disturbances	4	0.50^{b}	0.63^{b}	0.47^{b}	1.17^{a}	0.101	0.01	0.01	0.06	Y = 0.02x-0.14
Total disturbance[5]	4	0.89	0.88	0.74	1.43	0.114	0.08	0.05	0.07	Y = 0.01x+0.32

[1]Standard error of the mean; [2]Regression considered significant if P≤0.05

[3]**Total comfort and maintenance**: stretching, wing flapping, dustbathing, head scratching, and feather ruffling; [4]**Total aggression**: fighting and aggressive pecking; [5]**Total disturbance**: moderate disturbances and severe disturbances.

[a,b] Means with common letter within a row do not differ significantly (P≤0.05).

The behaviour of turkey hens (percentage of birds performing various behaviours based on the percentage of birds within a field of view) at 11 weeks of age is seen in Table 3.15. The percentage of birds present at the feeder at 11 weeks was not impacted by SD, however, the percentage of birds at the drinker demonstrated a quadratic relationship with SD (2.97, 3.98, 2.79, 2.34 % for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; P=0.02). The percentage of birds resting followed the same trend as week 8 where more birds were observed resting in the 60 kg/m^2 treatment (linear; P<0.01) and the percentage of birds walking was highest in the lowest SD treatment of 30 kg/m^2 (linear; P=0.03). For exploratory behaviours, there was a tendency for environmental pecking to demonstrate a quadratic relationship with SD with the most birds performing environmental pecking behaviour in the 30 kg/m^2 treatment (P=0.07). The percentage of hens feather pecking was 0.56, 1.22, 0.90, 1.04 % for SD treatments 30, 40, 50, and 60 kg/m^2, respectively (linear; P=0.01). The percentage birds head scratching was highest in the 30 kg/m^2 (linear; P=0.02). The percentage of birds that experienced severe disturbances followed a similar trend to week 8 in which severe disturbances linearly increased with increasing density (P=0.02). At 11 weeks of age, the percentage of birds aggressive pecking and total aggression, was highest in the 40 kg/m^2 treatment compared to the 30 and 50 kg/m^2 treatments (ANOVA; P=0.02 and P=0.01, respectively).

3.5.6 Litter moisture

At both 8 and 11 weeks of age, litter moisture was not significantly affected by SD, however, there was a linear tendency for litter moisture to increase as SD increased (29.0, 33.55, 35.84, 34.97% for SD treatments 30, 40, 50, and 60 kg/m^2, respectively; P=0.08) at 11 weeks of age (Table 3.16).

Table 3.15. Effect of estimated final stocking density on percentage of turkey hens performing various behaviours (% of birds within the field of view) at 11 weeks of age

Behaviour	n	Stocking density (kg/m²)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
Nutritive behaviours										
Feeding	4	5.37	7.70	5.30	4.89	0.637	0.51	0.64	0.34	-
Drinking	4	2.97[b]	3.98[a]	2.79[b]	2.34[b]	0.196	<0.01	0.15	0.02	$Y = -0.37e^{-2}x^2+0.30x-2.54$
Mobility behaviours										
Standing	4	13.05	11.16	9.56	9.45	0.653	0.51	0.24	0.62	-
Walking	4	8.86[a]	5.75[ab]	5.08[b]	4.51[b]	0.609	0.02	0.03	0.19	$Y = -0.14x+12.22$
Resting	4	58.90	60.57	69.12	70.28	1.985	0.07	<0.01	0.67	$Y = 0.43x+45.50$
Exploratory behaviours										
Litter pecking	4	1.87[a]	1.44[ab]	0.77[b]	1.14[ab]	0.150	0.04	0.10	0.13	-
Environmental pecking	4	1.94[ab]	1.42[ab]	1.79[a]	0.55[b]	0.222	0.04	0.12	0.07	-
Feather pecking	4	0.56[b]	1.22[a]	0.90[ab]	1.04[a]	0.087	<0.01	0.04	0.09	$Y=0.01x+0.42$
Comfort and maintenance behaviours										
Preening	4	4.75	4.28	3.26	3.69	0.209	0.18	0.17	0.23	-
Stretching	4	0.19	0.16	0.10	0.09	0.022	0.70	0.23	0.78	-
Wing flapping	4	0.21	0.22	0.34	0.15	0.034	0.17	0.73	0.10	-
Dustbathing	4	0.03	0.04	0.01	0.02	0.006	0.34	0.25	0.87	-
Perching	4	0.01	0.00	0.00	0.07	0.013	0.16	0.12	0.12	-
Head scratching	4	0.12	0.06	0.04	0.01	0.016	0.17	0.02	0.77	$Y = -0.34e^{-2}x+0.21$
Feather ruffle	4	0.12	0.15	0.08	0.17	0.017	0.12	0.17	0.28	-
Total comfort & maintenance[3]	4	0.66	0.62	0.58	0.43	0.049	0.78	0.51	0.41	-
Aggressive behaviours										
Fighting	4	0.10	0.13	0.04	0.10	0.018	0.31	0.89	0.85	-
Aggressive pecking	4	0.32[b]	1.12[a]	0.23[b]	0.68[ab]	0.118	0.02	0.48	0.74	-
Total aggression[4]	4	0.42[b]	1.25[a]	0.27[b]	0.77[ab]	0.128	0.01	0.55	0.80	-
Disturbances										

Moderate disturbances	4	0.25	0.19	0.22	0.27	0.030	0.79	0.51	0.47	-
Severe disturbances	4	0.38[b]	0.43[ab]	0.35[b]	0.76[a]	0.063	<0.01	0.02	0.18	$Y = 0.01x+0.61e^{-2}$
Total disturbance[5]	4	0.63	0.63	0.57	1.03	0.079	0.08	0.06	0.21	-

[1]Standard error of the mean; [2]Regression considered significant if P≤0.05

[3]**Total comfort and maintenance**: stretching, wing flapping, dustbathing, head scratching, and feather ruffling; [4]**Total aggression**: fighting and aggressive pecking; [5]**Total disturbance**: moderate disturbances and severe disturbances.

[a,b] Means with common letter within a row do not differ significantly (P≤0.05).

Table 3.16. Effect of estimated final stocking density on average[3] litter moisture percentage at 8 and 11 weeks

Age (weeks)	n	Stocking density (kg/m²)				SEM[1]	*P*-value (ANOVA)	*P*-value (Linear)	*P*-value (Quadratic)	Regression Equation[2]
		30	40	50	60					
8	2	28.31	32.60	35.12	31.22	1.52	0.55	0.45	0.21	-
11	2	29.90	33.55	35.84	34.97	1.15	0.31	0.08	0.27	-

[1]Standard error of the mean.
[2]Regression considered significant if P≤0.05.
[3]Average moisture of samples taken from front, middle, and back of room

3.6 Discussion and Conclusions

Stocking density can impact the productivity and welfare of birds. Providing evidence that clearly defines what these impacts are is important for the development of SD guidelines. As discussed in Chapter 2, higher SD can result in improved economic return despite negative performance impacts. However, determining the relationship between SD and bird wellbeing is complicated but necessary to make recommendations. By encompassing a multidisciplinary approach to studying the effects of SD in turkey hens, it can better aid in developing SD guidelines that accounts for both performance and welfare effects. Therefore, this chapter evaluates the health and welfare effects in relation to SD for turkey hens by evaluating footpad lesions, mobility, litter moisture, feather condition, incidence of aggression, chronic stress, and behaviour.

Bird mobility, evaluated by subjective gait scoring, is a measure of bird welfare and has been previously evaluated in relation to SD. When a bird's mobility is reduced, it can impact growth from limiting access to feed and water and more severe issues may potentially cause pain from skeletal abnormalities (Jong et al., 2012; Jankowski et al., 2015). Previous studies have found gait scores to be poorer in 12-week old hens and toms and 16-week toms reared at higher densities (60 kg/m^2 and above) (Martrenchar et al., 1999; Beaulac and Schwean-Lardner, 2018). At week 8 of this study, mobility was not affected by increasing SD. However, at 11 weeks of age, the percentage of birds with an identifiable abnormality that does not impact function (score 2) was higher in the 60 kg/m^2 treatment compared to the 30 kg/m^2 at 11 weeks of age. This may relate to the previous studies listed where gait differences were only noted from 12 weeks of age in both hens and toms, thus suggesting that hens raised to 11 weeks of age at various densities may not experience severe gait abnormalities up until that age. In broilers, high SD has been found to reduce mobility when bird movement was restricted (Sørensen et al., 2000). At both 8 and 11 weeks of age, bird behaviour showed that they were less active at high SD and rested more. Therefore, at higher densities, there is reduced floor space which hinders the bird's ability to be active and this can lead to poor mobility and reduced growth (Chapter 2), especially at older ages.

The presence of FPD in turkeys can be influenced by many factors including litter substrate quality, breed, diet, and management practices (Mayne, 2005). Footpad lesions have been linked to poor gait and could indicate discomfort and pain (Martland, 1984; Weber Wyneken et al., 2015). Martrenchar et al., (1999) and Beaulac and Schwean-Lardner (2018) found footpad lesions to worsen with increasing density from 38.8 to 62.7 kg/m^2 at 12 (hens) and 16 (toms) weeks of age and from 30 to 60 kg/m^2 at 10 and 16 weeks of age (toms), respectively. In this study, average footpad scores worsened with increasing density at 8 and 11 weeks of age. The effect of SD on footpad lesions at week 8 and the numerical differences observed at week 11 are in accordance with the studies listed above. Another significant contributing factor to the presence of FPD is litter moisture. It is documented that litter moisture is directly affected by ventilation and the ability for the air to dry the litter as well as high SD when more birds produce more fecal matter (Proudfoot et al., 1979b; Noll et al., 1991; Zuidhof et al., 1993, 1995; Martrenchar et al., 1997, 1999; Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019). In this study, litter moisture did not differ between SD treatments at week 8 but a trend was observed at week 11 for increasing litter moisture with increasing SD. Beaulac et al. (2019) suggested that despite adequate ventilation for air quality, at high densities when more birds are resting, the restricted ability for the air to reach the litter and dry it can contribute to higher litter moisture. In accordance with this, a larger percentage of hens were observed resting at high SD which could have contributed to the higher presence of FPD observed. At high SD, birds exhibited less active behaviours and rested more on wet litter, thus contributing to the presence of footpad lesions.

Evaluating feather cover and cleanliness can be a measure of bird health and wellbeing. Feather cover is important for thermoregulation and protection from scratches. Poor feather cover can also affect feed efficiency because more energy is required to thermoregulate after heat loss, therefore, the birds consume more feed to compensate for energy losses (Leeson and Morrison, 1978; Sarica et al., 2008). Furthermore, reduced feather cover caused by feather pecking results in a negative impact on bird wellbeing. Feather pecking affects birds as it can lead to cannibalism if the skin tears and the removal of feathers can be painful (Gentle and Hunter, 1991). Thus, feather cover is an important welfare measure. Feather cover worsened with increasing SD at both 8 and 11 weeks. This same effect was observed in two previous studies, where feather cover was poorer at high SD (60 kg/m^2) at 10, 12, and 16 weeks in toms (Beaulac and Schwean-Lardner, 2018) and at 14 weeks in hens and toms at 48 kg/m^2 (Coleman and Leighton, 1969) suggesting that feather cover can be affected at younger ages, even before maximum SD is reached. When evaluating feather cover, it is important to evaluate the different areas of the body that can be affected as a result of space allowance, behaviour, and management practices; these include the neck, breast, wings, tail and back. Neck feather scores were unaffected by SD and though only a hypothesis, this could suggest that providing feeders on a per bird basis can reduce the impact of feeder space competition at higher densities when friction from the feeders against the neck can cause feather loss. Laying hens had significant neck feather loss at reduced space allowance where more competition for feeder space was observed (Sarica et al., 2008). Breast feather scores worsened with increasing SD at 8 and 11 weeks and this was also observed in the study by Beaulac and Schwean-Lardner (2018). The authors suggested that although turkeys do not have many feathers on the breast, these results may be caused by increased contact with wet litter (Thomas et al., 2004) and birds in this study exhibited more resting behaviour at higher SD. The wings had poorer feather cover at higher densities at 11 weeks of age. This may be a result of birds brushing against pen mates when maneuvering at higher SD which may cause damage to the wing feathers. Tail feather cover was poorer with increasing density, however, the percentage of birds treated for aggressive damage to the tail was lowest at high SD. This suggests that reduced space can cause damage to tail feathers. Beaulac and Schwean-Lardner (2018) did not observe effects of SD on tom wing feather cover but did see similar tail feather cover results. The authors suggested that reduced space as the birds develop and grow may cause the tail feathers to be affected when birds are moving past each other at higher SD, causing more friction to the tail, or by birds stepping on other bird's tails when resting. This may explain the wing and back feather cover in which they worsened with increasing density at 8 weeks and more severely at 11 weeks when space was limited in this study. This was also observed in toms at 12 and 16 weeks of age (Beaulac and Schwean-Lardner, 2018).

Feather cleanliness has only been studied in relation to SD in one previous study (Beaulac and Schwean-Lardner, 2018). Feather cleanliness worsened with increasing SD over the course of the 16 week trial with toms (Beaulac and Schwean-Lardner, 2018) and this same effect was seen in this study, as birds were dirtier at high SD at both 8 and 11 weeks of age. When feathers come into contact with fecal matter and wet litter, the feathers become wet and dirty which may cause the birds to become colder and require more energy to maintain body temperature (Hunter et al., 2010; Beaulac and Schwean-Lardner, 2018), thus, feather cleanliness is important for thermoregulation and comfort. There is also a human health risk associated with feather cleanliness as dirty and fecal contaminated feathers at slaughter pose a meat quality and safety issue (Wilkins et al., 2003; Beaulac and Schwean-Lardner, 2018).

Aggressive pecking causing damage to the skin, often referred to as injurious pecking, is a major welfare concern and is common in commercial strains of turkeys (Martrenchar et al., 2001). Aggressive damage can lead to serious injuries or culling (Buchwalder and Huber-Eicher, 2004). In this study, aggressive damage was recorded when a bird was treated with a deterrent for an open lesion as a result of pecking or as a result of a skin tear that can attract other birds to start pecking at the open wound. Aggressive damage was highest at low SD for week 5-8, 8-11, and for the duration of the study from 0-11 weeks. This is the opposite effect observed in toms at 13 weeks of age in which aggressive damage was highest at high SD (Buchwalder and Huber-Eicher, 2004). Though not statistically significant, Beaulac and Schwean-Lardner (2018) observed a high overall incidence of aggressive damage in toms in the highest and lowest SD as well as a quadratic relationship with SD at 4-8 weeks of age where aggressive damage was highest at low and high SD. Therefore, sex differences may explain why hens did not exhibit aggressive damage at higher SD and only at lower SD. Incidence of aggressive pecking and damage have been found to be lower in hens compared to toms, who reach sexual maturity earlier (Denbow et al., 1984; Leighton et al., 1985; Buchwalder and Huber-Eicher, 2003; Dalton et al., 2013). Turkey hens can reach sexual maturity at approximately 21 weeks of age (Siopes, 2010) yet turkey hens are often raised to 12 weeks (with some reared for longer) in Canada which is well before hens may show aggression or dominance behaviour. This may be why the studies listed above observed less aggressive damage in hens. At lower SD in the current study, birds were more active and this may have resulted in more aggressive behaviours to be exhibited in the turkey hens (Beaulac and Schwean-Lardner, 2018).

Incidence of aggressive damage by location on the body shows that the tail and head were targeted more by birds in the lowest SD. Aggression is often targeted at the head in turkeys (Moinard et al., 2001) and this leads to mortality and culls resulting from aggressive damage. Head pecking is a learned behaviour by poults as a fighting method used by older birds to establish a dominance hierarchy (Buchholz, 1997; Dalton et al., 2013). It is also thought that head pecking may be influenced by frequent environmental disturbances at high SD and is performed by hens and toms after a disturbance to re-establish the dominance hierarchy and settle the flock (Gill and Leighton, 1984; Buchwalder and Huber-Eicher, 2003). It has been suggested that birds at high SD need to continuously re-establish a dominance hierarchy when encountering unfamiliar birds in the pen, as a low level of familiarity between birds has been found to result in a higher level of aggressive head pecking (Buchwalder and Huber-Eicher, 2003, 2004). However, the studies listed had total group sizes of 5 (Buchwalder and Huber-Eicher, 2004) or 8 toms (Buchwalder and Huber-Eicher, 2003) housed in small pens (2x3m and 3x6m, or 3.1x3.7m, respectively) where one unfamiliar conspecific was introduced to either 4 or 7 toms. At low SD in this study, the group size (295 hens at placement) may have been too large for the birds to establish a dominance hierarchy. It has also been suggested that in large group sizes (>100 birds) social hierarchy could be too difficult to establish so aggressive behaviours occur independently of group size (Denbow et al., 1984; Hughes et al., 1997; Dalton et al., 2013).

Stress may also have a direct effect on bird wellbeing. There are few studies that have evaluated chronic stress (in this study measured via H/L ratio) in response to various SD levels in turkeys. One study found SD (high SD treatment of 58 kg/m^2) to have no effect on stress (H/L ratio) in toms at 7, 12, 16, or 20 weeks of age (Hafez et al., 2016). In comparison, Beaulac and Schwean-Lardner (2018) observed a linear increase of H/L ratio in toms housed at increasing density at 4 weeks of age and a similar trend was observed at 12 weeks of age, suggesting increased stress with increased SD. The authors suggested that SD or group size may impact turkeys' stress response earlier in life compared to older ages as the higher H/L ratios observed at 4 weeks were correlated to increases in aggressive damage and aggression related mortality and culls at that age (Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019). Similarly, the results of this study show that stress increased linearly with increasing SD at 5 weeks of age. This may be related to increased SD or increased group size causing more stress (Buchwalder and Huber-Eicher, 2004, 2005; Beaulac and Schwean-Lardner, 2018). At 8 weeks of age, birds were more stressed in the 40 and 30 kg/m^2 treatments which may be related to the higher incidence of aggression related mortality and culls (Chapter 2) and aggressive damage observed from 5-8 and 8-11 weeks of age at lower SD. Birds in the lower SD treatments were also more active (increased walking, standing and litter pecking and decreased resting). The increase in stress observed at week 8 at lower SD suggests the birds may have had a stress response to the increased activity and aggression observed, however, by week 11, higher H/L ratios were observed in the 50 and 60 kg/m^2 treatments. In the higher SD treatments at 11 weeks of age, there was reduced floor space, more birds being disturbed by other birds, increased incidence of FPD, and they were less active which may have been stressful. It has been documented that the H/L response to mild and moderate stressors results in increased heterophils, whereas severe stressors result in basophilia (increased number of basophils) and heteropenia (lower than normal levels), thus, the H/L ratio becomes unreliable depending on the degree of the stressor (Maxwell, 1993; Maxwell and Robertson, 1998). Therefore, evaluating the additional health and welfare parameters may more accurately help identify the effects of SD on the wellbeing of the birds.

Evaluating turkey behaviour aids in understanding the birds affective state as a measure of welfare. It is thought that expressing certain behaviours can also aid in improving the health and physical condition of birds which will improve welfare (Duncan, 1998). Positive affective states can occur when an environment or other factors allow the expression of behaviours that are driven by motivation (Fraser, 2008). Therefore, by providing an environment that allows birds to perform behaviours such as comfort and maintenance, exploratory, and social behaviour can improve welfare by promoting a positive affective state (Fraser, 2008). There have not been many studies that have evaluated the effects of SD on turkey behaviour, and more have focused on toms not hens. However, variations in group size, pen size, and SD levels can be confounding factors in previous studies and this has caused conflicting results between studies and some behaviours.

Feeding and drinking behaviours – classed as **nutritive behaviours** in this study– are important for turkey production as they directly impact performance. Martrenchar et al. (1999) found no differences in the percentage of time hens or toms spent feeding or drinking in relation to SD. In the current study, a larger percentage of hens were present at the feeder with increasing SD at 8 weeks of age, but drinking was not affected by SD. This has not been observed in previous studies. At 8 weeks of age, mobility was not affected by SD, meaning birds were able to reach the feeder without gait affecting them, ruling out the potential of mobility resulting in these differences. As discussed in Chapter 2, broilers were found to approach a feeder with birds already present and would stay at that feeder for longer when the other birds were present (Collins and Sumpter, 2007). This may explain that birds housed at higher densities were motivated by social feeding behaviour and were able to move to feeders at 8 weeks of age despite presence of FPD. At 11 weeks of age, the percentage of birds at the feeder was unaffected by SD and drinking demonstrated a quadratic relationship with the largest percentage of birds present at the drinker observed in the 40 kg/m^2 treatment. In the study by Beaulac and Schwean-Lardner (2018), toms feeding behaviour was unaffected at 12 weeks of age which is similar to this study, however, in the older toms (14 and 16 weeks of age) feeding reduced with increasing SD.

Standing, walking, and resting – classed as **mobility behaviours** in this study– are important indicators of bird activity housed at various SD. Martrenchar et al. (1999) did not observe any differences in activity of toms and hens relative to SD (38.8 to 62.7 kg/m^2). However, Beaulac and Schwean-Lardner (2018) found a decrease in the percentage of toms walking and an increase in the percentage of toms resting at high SD (60 kg/m^2) at week 12, 14, and 16. In the current study, birds were more mobile at 8 weeks of age in the low SD treatment as the amount of birds walking and standing linearly decreased with increasing density. In addition, a greater percentage of birds were resting with increasing SD. This suggests that with increased floor space and smaller group size, birds will be more active.

At 11 weeks of age, the percentage of birds walking was higher at low SD and they rested more at high SD. However, despite more birds resting at high SD, no differences were observed for the percentage of birds standing in the high SD treatment compared to the lower densities. This may indicate that the birds observed standing in the higher SD treatments did not have enough space to rest comfortably (Beaulac and Schwean-Lardner, 2018). Beaulac and Schwean-Lardner (2018) observed more birds standing in the 30 and 60 kg/m^2 treatments and more birds resting in the 50 kg/m^2 which supported the idea that above 50 kg/m^2, birds could not rest comfortably and had to stand as a result. There may also give more opportunity for the few birds not resting at high SD to disturb those that are.

With increasing SD, there was a greater percentage of hens disturbing other birds. Similar results were observed for disturbances in turkey hens as early as 6 weeks of age up to 12 weeks of age at high SD (62.7 kg/m^2; Martrenchar et al., 1999). Conversely, toms experienced more disturbances at low density at 12 weeks which may have been a result of better mobility observed in toms in the 30 kg/m^2 treatment (Beaulac and Schwean-Lardner, 2018). Broilers housed at high SD have been observed to be less active and experience more disturbances when birds move to the feeders or drinkers and they bump into or walk over one another (Martrenchar et al., 1997; Simitzis et al., 2012). Thus, is it evident that the wellbeing of birds housed at high SD may be affected as they were much less active and experienced more disturbances when resting in comparison to those housed at low SD.

Exploratory behaviours – including litter pecking, environmental pecking, feather pecking – are performed to explore stimuli in the environment and if these behaviours cannot be performed it may lead to boredom (Newberry, 1999). To the best of the author's knowledge, environmental pecking and feather pecking behaviours in relation to SD in turkeys have only been studied once, but no effects were found with increasing SD (Beaulac and Schwean-Lardner, 2018). In the current study, a greater percentage of hens were litter pecking with decreasing SD which further supports that birds are more active at low SD. Litter pecking is related to foraging, which is an innate behaviour that birds perform as a feeding and exploratory behaviour (Duncan, 1998; Miller and Mench, 2005). Environmental pecking was higher at low SD, similar to the results observed by Beaulac and Schwean-Lardner (2018), and the authors suggested that the birds have more space to perform this behaviour at low SD, however, it is considered a low incidence behaviour and may not show significant results. The percentage of birds feather pecking (gentle pecks at a pen mates feathers that does not cause damage) increased with increasing density. Feather pecking is thought to be redirected foraging behaviour (Dalton et al., 2013) and not performed as an aggressive act. This suggests that at high SD there is reduced floor space and more birds resting in close proximity resulting in more feather pecking and fewer birds engaged in exploratory behaviours like litter pecking. In addition, poor feather cleanliness observed at high SD may have contributed to more birds feather pecking which can be performed as a social preening behaviour (Savory, 1995)

Preening, stretching, wing flapping, dust bathing, perching, head scratching, and feather ruffling have been classed together as **comfort and maintenance behaviours** in this study. Preening (Duncan and Wood-Gush, 1972), dustbathing (Lindberg and Nicol, 1997), and head scratching (Nicol, 1989; Duncan, 1998) have been categorised as maintenance behaviours, however, high incidence of these behaviours in some cases can be signals of poor health or welfare. Stretching, feather ruffling, and wing flapping are considered comfort movements and feather maintenance behaviours (Appleby et al., 2004). Increased stress and social pressure at high and low SD could cause preening to be performed as a displacement behaviour (Duncan, 1998). Dustbathing is thought to be triggered by internal (Vestergaard, 1982) and external factors

(Petherick et al., 1995) such as the need for feather maintenance and stimulation from the presence of an adequate litter substrate. Head scratching is performed to maintain plumage and skin condition (Nicol, 1989; Duncan, 1998) but a high incidence of this can occur in other situations, such as if birds are infected with ectoparasites (Kilpinen et al., 2005; Temple et al., 2020). At 8 weeks of age, the percentage of birds dust bathing and head scratching demonstrated a quadratic relationship with SD where more birds performed those behaviours at moderate density (40 and 50 kg/m^2). These results may suggest that birds at moderate SD had adequate floor space and were experiencing a positive affective state compared those at low and high SD. In this study, the percentage of birds preening was numerically higher for birds in the 40 and 60 kg/m^2 treatment at week 8. In comparison, Beaulac and Schwean-Lardner (2018) observed more toms preening at high SD at week 14 and 16. This coincided with poor feather cleanliness at high SD resulting in an increased need for feather maintenance because of dirty feathers. At week 8 in the current study, it is likely that this same occurrence at high SD was due to poor feather cleanliness and as a displacement behaviour to some extent. In addition, aggressive behaviour at week 8, including the percentage of birds fighting and aggressive pecking, increased with decreasing SD and this may have added to social stress. This relates to the higher incidence of aggressive damage and more aggression related mortality and culls (Chapter 2) observed at low SD.

It is evident that high SD can negatively impact turkey hen health and welfare raised to 11 weeks of age. It was hypothesized that high SD would negatively impact the health of the turkey hens resulting from increased incidence of FPD, poorer mobility, and poor feather coverage and cleanliness. Birds housed at high SD (60 kg/m^2) had an increased incidence of FPD, poorer mobility at 11 weeks, and poor feather cover and cleanliness which supports this hypothesis. It was also hypothesized that high SD would result in decreased activity and increased aggression, leading to an increase in aggression related mortality and, higher stress levels (H/L ratios). The decreased activity and higher H/L ratios observed at high SD support this hypothesis; however, aggression related mortality was lowest at high SD and highest in the low SD treatment (30 kg/m^2). At low SD, it was hypothesized that there would be an increased expression of comfort behaviours as there is more space for this to occur, however, higher aggression would be seen at low SD levels due to more birds being disturbed by other birds. This was partially supported as there was a numerically higher expression of comfort behaviours at low SD and more aggressive behaviour and incidence of aggressive damage, but this was caused from increased activity and not caused from increased disturbances. In conclusion, high and low SD (30 and 60 kg/m^2) had negative impacts on the health and wellbeing of turkey hens. Thus, moderate densities may be more ideal to achieve optimal bird health and welfare.

3.7 Acknowledgements

The author would like to acknowledge the Canadian Poultry Research Council, Agriculture and Agri-Food Canada, Aviagen Turkeys Inc., and Charison's Turkey Hatchery Ltd. for their financial support and thank the students and staff at the University of Saskatchewan Poultry Centre for their help with these turkey trials.

4.0 Chapter 4: Overall Discussion

4.1 Introduction

Stocking density (SD) can have significant impacts on the welfare and productivity of turkeys. With a gap in the literature on the effects of SD in turkey hens, it is important to establish guidelines that allow for efficient production while balancing bird wellbeing. Currently, there are many variations in the SD guidelines for turkeys in North America. For example, the National Turkey Federation (2012) suggests a maximum SD of up 73.2 kg/m^2, whereas the Certified Humane program suggests a maximum standard of 36.6 kg/m^2 (Human Farm Animal Care, 2014). The Canadian Codes of Practice recommend a maximum SD range from 40-65 kg/m^2 depending on the final predicted body weight of the turkeys raised (National Farm Animal Care Council, 2016). For turkeys raised between 6.2-10.8 kg, such as the hens in this study, the recommended SD is 45 kg/m^2 up to 50 kg/m^2 if specific environmental and management requirements are met (National Farm Animal Care Council, 2016). The differences found in SD recommendations may be a result of varying effects of SD on turkeys observed in previous literature (Chapter 1). Therefore, it is important to develop guidelines based on current research, especially for turkey hens, as more studies focus on toms.

The purpose of this research was to assess the impacts of graded levels of SD on the performance, health, and welfare of turkey hens raised to 11 weeks of age. The four SD treatments evaluated in this study were estimated final densities of 30, 40, 50, and 60 kg/m^2. The impacts on the performance of turkey hens were evaluated by measuring body weight (BW), feed consumption, feed efficiency (F:G), flock uniformity, and mortality. An economic analysis was performed to determine economic return for each SD treatment. Mobility (subjective gait scoring), footpad scoring, feather cover and cleanliness scoring, incidence of aggressive damage, heterophil/lymphocyte (H/L) ratios, behaviour, and litter moisture were evaluated to determine the effects of SD on the health and welfare of turkey hens. By encompassing all the parameters mentioned above, a multidisciplinary approach was used to determine SD guidelines for turkey hens raised to 11 weeks of age.

4.2 Discussion

The increasing levels of SD (30, 40, 50, and 60 kg/m^2) evaluated in this study have demonstrated significant effects on the production and welfare of turkey hens raised to 11 weeks of age. Although increasing density resulted in increased income, factors such as increased management costs, equipment damage, and labour costs that could occur when housing turkeys at high SD were not calculated. However, low SD does not necessarily mean better welfare in all measured components despite improved production, as alterations in behaviour and incidence of aggression were noted.

With increasing SD, the performance parameters affected include BW, BW gain, and feed consumption. Body weight was reduced at 11 weeks of age and overall BW gain (from 0-11 weeks) tended to decrease with increasing SD. Feed consumption reduced with increasing SD from 8-11 weeks and overall (0-11 weeks). At 3-5 weeks of age, feed efficiency was poorer in the 30 kg/m^2 treatment compared to the 50 kg/m^2 treatment. However, this was the only age with significant effects of SD on feed efficiency. Poor performance toward the end of the rearing cycle is important to understand as that is when target market BW is reached. These performance effects may be explained by the health, welfare, and behaviour data observed in this study.

The reduction in BW and BW gain observed with increasing SD may be related to poor mobility and reduced floor space. Gait scores indicated that more birds in the 60 kg/m^2 treatment had an identifiable abnormality that did not affect overall function (11 weeks) and a higher incidence of footpad dermatitis (FPD; 8 and 11 weeks). The poorer mobility, presence of FPD, and a trend for increased litter moisture observed at 11 weeks of age could make reaching the feeder more difficult, especially because floor space becomes limited as the final estimated SD is more closely achieved. The presence of FPD in poultry has been linked to poor gait and could indicate discomfort and pain (Martland, 1984; Weber Wyneken et al., 2015).

The difficulty of reaching the feeder is further supported by changes in behaviour. Initially, at 8 weeks of age, a larger percentage of birds in the 60 kg/m^2 treatment were at the feeder despite the presence of FPD. This suggests that birds in higher densities were motivated by social feeding behaviour (Collins and Sumpter, 2007; Beaulac et al., 2019) and were still able to move to feeders when floor space was not as limited at that age. By 11 weeks of age, when the final estimated SD was reached and floor space was limited, the percentage of birds present at the feeder did not differ between treatments, however, feed consumption decreased with increasing SD. Birds housed at high SD also rested more and fewer birds were observed walking suggesting that they may have been less motivated to access feeders as they would need to exert more energy to move between more resting pen mates (Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019).

Reduced growth may be impacted by numerous factors, including stress or an immune response. When birds are exposed to stressors and the hypothalamic-pituitary-adrenal (HPA) axis is activated, adrenocorticotropin hormone (ACTH) stimulates the production and release of corticosterone and plasma corticosterone concentrations increase (He et al., 2018). In a study where a model was created to study stress in broiler chickens by giving ACTH to the birds and monitoring the physiological responses, it was found that continuous delivery of ACTH caused an increase in corticosterone, H/L ratios, and other physiological parameters, resulting in a significant decrease in BW (Puvadolpirod and Thaxton, 2000). In the current study, turkey hen H/L ratios linearly increased with increasing SD at 5 weeks and were highest in the 50 and 60 kg/m^2 treatments at 11 weeks of age which may suggest that stress contributed to the reduced growth performance. Similar results were found in another study of broilers' response to multiple stressors. Broiler BW decreased and H/L ratios increased, thus, it was suggested that the lower BW noted may be caused by the reallocation of resources from the body's growth and more towards the increased immune response to stress (McFarlane and Curtis, 1989; McFarlane et al., 1989).

In addition to the effects of stress on growth, increased stress may have had other effects on the health of the birds housed at high SD. Infectious related mortality and culls were highest in the birds at high SD. This may suggest that birds were experiencing stress (increased H/L ratios observed) as a result of high SD, which impacts the immune function causing more infectious related mortality. At high SD, the close proximity of birds may increase the risk of spreading infections. In addition, footpad lesions have been related to increases in secondary infections (Martrenchar et al., 2002) and it was hypothesized that feather cleanliness may be a factor in infectious disease prevalence (Beaulac et al., 2019). At both 8 and 11 weeks, feather cleanliness and footpad lesions were poorer as SD increased and this could have also contributed to the higher infectious related mortality.

When feathers become wet and dirty from increased contact with fecal matter and wet litter, it may cause the birds to lose their thermoregulatory ability and require more energy to maintain body heat (Hunter et al., 2010; Beaulac and Schwean-Lardner, 2018). Feather cover and cleanliness was poorer at high SD which may have resulted in more heat loss. This can affect feed efficiency, as the birds consume more feed to compensate for energy losses from thermoregulation (Leeson and Morrison, 1978; Sarica et al., 2008). Behavioural observations support this as a larger percentage of birds were present at the feeder at 8 weeks of age in the 60 kg/m^2 treatment, but BW was reduced (8-11 weeks). In addition, more birds were resting on wet litter which may have contributed to dirtier feathers, and this may have resulted in a tendency for more birds to preen. Preening can occur as a displacement behaviour caused by increased stress and social pressure at high SD (Duncan, 1998) as well as a maintenance behaviour for cleaning feathers (Duncan and Wood-Gush, 1972). With both increased stress and poor feather cover and cleanliness, it could be suggested that these factors influenced the tendency for increased preening behaviour at high SD. At high SD, a larger percentage of birds exhibited feather pecking behaviour which may have occurred as a form of social preening (Savory, 1995) when they were in close proximity when resting. This form of gentle feather pecking can be exploratory and directed at food particles or debris on a pen mates feathers (Savory, 1995; Hughes and Grigor, 1996; Dalton et al., 2013). Dirtier feathers observed at high SD may have contributed to this.

One of the objectives of this study was to minimize the impact of confounding factors such as air quality; thus, ventilation was adjusted to match air quality in all treatments which included maintaining carbon dioxide (CO_2) and ammonia within acceptable levels. Litter moisture can be significantly affected by ventilation rate and increased SD which causes increases in CO_2 and ammonia (Zuidhof et al., 1993; Mayne, 2005). Litter moisture at week 8 was unaffected by SD which can be directly related to reducing the impact of air quality in this study. Average CO_2 and ammonia levels in both blocks were consistent between treatments. However, a linear trend for increased litter moisture was noted at 11 weeks at high SD which may have been due to maximum allowable levels of ammonia being reached when external ambient temperatures were extremely low, and ventilation had to be reduced. It is well documented that high SD and poor ventilation causes higher litter moisture due to increased fecal matter (Proudfoot et al., 1979a; Noll et al., 1991; Zuidhof et al., 1993, 1995; Martrenchar et al., 1997, 1999). However, Beaulac et al., (2019) found litter moisture to be highest in the middle SD treatments of 40 and 50 kg/m^2. The authors suggest that in addition to ventilation rate and excreta output, the ability for air to circulate and reach the litter to dry it can be a contributing factor to increased litter moisture as birds in those treatments rested more, therefore, there was not enough open space to dry the litter (Beaulac and Schwean-Lardner, 2018; Beaulac et al., 2019). This is supported in the current study as the larger percentage of birds were resting on wet litter at high SD may have prevented the litter from drying due to restricted air flow. Thus, increased litter moisture at high SD may have contributed to poorer feather cover and cleanliness, increased incidence of FPD, and poorer mobility at 11 weeks.

Birds housed at high SD were further impacted by reduced floor space, resulting in more birds being disturbed by others. Because more birds were resting, when pen mates had to manoeuvre between each other to reach resources, it resulted in more disturbances. As a larger percentage of birds were observed resting on wet litter, this may have contributed to poor breast feather cover observed at high SD (Thomas et al., 2004; Beaulac and Schwean-Lardner, 2018). The increase in disturbances could have resulted in the poorer tail, wing and back feather cover observed because of the limited floor space when birds were brushing past or walking on top of each other resulting in friction and the removal of a pen mates feathers (Beaulac and Schwean-Lardner, 2018).

The close proximity of birds at high SD suggests the birds had less space for activity and may have experienced frustration as seen through a numerically higher percentage of birds preening which may have been performed as a displacement behaviour (Duncan and Wood-Gush, 1972). In comparison, a larger percentage of birds in the middle densities (40 and 50 kg/m^2) were observed dustbathing compared to the 30 and 60 kg/m^2 treatments which may suggest that they had more space and were experiencing a positive affective state to perform this behaviour.

The effects of low SD on turkey hen welfare can be negative despite better performance. Incidence of aggressive pecking and damage is generally lower in hens compared to toms (Denbow et al., 1984; Leighton et al., 1985; Buchwalder and Huber-Eicher, 2003; Dalton et al., 2013) as hens often don't reach sexual maturity during the typical rearing cycle. Thus, sex differences in aggressive behaviour are evident. However, toms are more aggressive when housed at high SD (Buchwalder and Huber-Eicher, 2004; Beaulac and Schwean-Lardner, 2018). However, low densities may also be problematic. One study found more aggression in toms at low SD between 4 and 8 weeks of age (Beaulac and Schwean-Lardner, 2018). To the best of the author's knowledge, the current study is the first to show an increased incidence of aggression and aggression related mortality and culls at low SD in hens. However, this provides important information regarding the wellbeing of turkey hens housed at low SD and the social stressors affecting them. At low SD, there was an increased incidence of aggression related mortality and culls and aggressive damage observed from 5-8 and 8-11 weeks of age. A larger percentage of birds in the 30 kg/m^2 treatment were observed standing, walking, litter pecking and environmental pecking and it is thought that the more active birds are at low SD, the more aggressive encounters they participate in (Beaulac and Schwean-Lardner, 2018).

4.3 Conclusions

High SD (60 kg/m^2) negatively affected the performance of turkey hens as BW and feed consumption were reduced, and trends were noted for BW gain. Birds in the low SD (30 kg/m^2) had the best performance parameters. Birds raised at higher SD, between 50-60 kg/m^2, had poorer feather cover and cleanliness, presence of FPD, reduced mobility and increased chronic stress. Behavioural changes in the high and low SD treatments may be of concern for bird welfare. In the low SD, birds exhibited more aggression, and performed less comfort and maintenance behaviours, but they were most active and performed exploratory behaviours. In the high SD, more birds were observed resting, experienced more disturbances from other birds, and performed more feather pecking and preening which may be indicative of frustration and stress from reduced activity. A similar percentage of birds were observed standing at high SD as the birds in the other treatments because of restricted floor space at 11 weeks reducing the ability to rest comfortably. Birds in the middle densities (40 and 50 kg/m^2) performed more comfort and maintenance behaviours, however, these are low incidence behaviours and may not accurately suggest a positive affective state.

In conclusion, the data presented in this thesis demonstrates that turkey hen performance, health, and behaviour are negatively impacted by high SD (60 kg/m^2). However, the data also demonstrate negative effects on bird welfare at both high and low SD of 60 and 30 kg/m^2.

Although economic return is greatest with higher SD, a balance between income (poult and feed cost vs income only), production, health, and welfare parameters are important when developing SD guidelines to ensure the welfare of birds is maximized without compromising economic return. Additionally, this economic analysis did not consider management and labour costs, equipment damage, or potential carcass condemnations, thus not accurately depicting the true effects of high SD on economic return. The data outlined in this thesis demonstrates that the age the birds are reared to is an important factor when forming SD recommendations for commercial production. Therefore, further research that encompasses a multidisciplinary approach assessing the effects of SD in turkey hens raised to older ages will be beneficial. By assessing all three factors of performance, health, and behaviour, it is evident that a moderate density of around 40 to 50 kg/m^2 is more beneficial to turkey hens raised to 11 weeks of age as well as the producer as it ensures efficient production, health, and wellbeing. However, this SD recommendation is based on maintaining air quality (CO_2 and ammonia) at or below allowable levels by managing ventilation and room temperatures.

5.0 Literature Cited

Appleby, M. C., J. A. Mench, and B. O. Hughes. 2004. Poultry Behaviour and Welfare. CABI, Washington, DC, USA.

Aviagen. 2015a. Nicholas Select Commercial Performance Objectives. Accessed October 2018. Available at https://www.aviagenturkeys.us/uploads/2021/01/14/Nicholas Comm Perf Obj Select.pdf.

Aviagen. 2015b. Management Guidelines for Raising Commercial Turkeys. Accessed October 2018. Available at https://www.aviagenturkeys.us/performance-objectives.

Ayer, L., K. Greaves-Lord, R. R. Althoff, J. J. Hudziak, G. C. Dieleman, F. C. Verhulst, and J. van der Ende. 2013. Blunted HPA axis response to stress is related to a persistent dysregulation profile in youth. Biol. Psychol. 93:343–351.

Balloun, S. L. 1958. The problem of "lame" chickens and turkeys. Iowa State Univ. Vet. 20:93–95.

Barnes, T. M., and K. A. Greive. 2017. Topical pine tar: History, properties and use as a treatment for common skin conditions. Australas. J. Dermatol. 58:80–85.

Bartz, B. M., K. A. Anderson, E. O. Oviedo-Rondon, K. Livingtson, and J. L. Grimes. 2020. Effects of stocking density on large white, commercial tom turkeys reared to 20 weeks of age: 1. growth and performance. Poult. Sci. 99:5582–5586.

Beaulac, K., H. L. Classen, S. Gomis, K. S. Sakamoto, T. G. Crowe, and K. Schwean-Lardner. 2019. The effects of stocking density on turkey tom performance and environment to 16 weeks of age. Poult. Sci. 98:2846–2857.

Beaulac, K., and K. Schwean-Lardner. 2018. Assessing the effects of stocking density on turkey tom health and welfare to 16 weeks of age. Front. Vet. Sci. 5:1–12.

Bilgili, S. F., and J. B. Hess. 1995. Placement density influences broiler carcass grade and meat yields. J. Appl. Poult. Res. 4:384–389.

Brown, E. J., and A. Vosloo. 2017. The involvement of the hypothalamo- pituitary-adrenocortical axis in stress physiology and its significance in the assessment of animal welfare in cattle. Onderstepoort J. Vet. Res. 84:1–9.

Buchholz, R. 1997. Male dominance and variation in fleshy head ornamentation in wild turkeys. J. Avian Biol. 28:223–230.

Buchwalder, T., and B. Huber-Eicher. 2003. A brief report on aggressive interactions within and between groups of domestic turkeys (Meleagris gallopavo). Appl. Anim. Behav. Sci. 84:75–80.

Buchwalder, T., and B. Huber-Eicher. 2004. Effect of increased floor space on aggressive behaviour in male turkeys (Meleagris gallopavo). Appl. Anim. Behav. Sci. 89:207–214.

Buchwalder, T., and B. Huber-Eicher. 2005. Effect of group size on aggressive reactions to an introduced conspecific in groups of domestic turkeys (Meleagris gallopavo). Appl. Anim. Behav. Sci. 93:251–258.

Buijs, S., L. Keeling, S. Rettenbacher, E. van Poucke, and F. A. M. Tuyttens. 2009. Stocking density effects on broiler welfare: Identifying sensitive ranges for different indicators. Poult. Sci. 88:1536–1543.

Buijs, S., L. J. Keeling, and F. A. M. Tuyttens. 2011a. Using motivation to feed as a way to assess the importance of space for broiler chickens. Anim. Behav. 81:145–151.

Buijs, S., L. J. Keeling, C. Vangestel, J. Baert, and F. A. M. Tuyttens. 2011b. Neighbourhood analysis as an indicator of spatial requirements of broiler chickens. Appl. Anim. Behav. Sci. 129:111–120.

Buijs, S., E. Van Poucke, S. Van Dongen, L. Lens, J. Baert, and F. A. M. Tuyttens. 2012. The influence of stocking density on broiler chicken bone quality and fluctuating asymmetry. Poult. Sci. 91:1759–67.

Canadian Food Inspection Agency. 2014. Canadian carcass poultry grading program – inspection manual for graded poultry. Gov. Canada. Accessed October 2018. Available at http://www.inspection.gc.ca/food/meat-and-poultry-products/program-changes/inspection-manual-for-graded-poultry/eng/1394632245049/1394632377400?chap=4.

Carlile, F. S. 1984. Ammonia in poultry houses: a literature review. Worlds. Poult. Sci. J. 40:99–113.

Cengiz, Ö., B. H. Köksal, O. Tatll, Ö. Sevim, U. Ahsan, A. G. Üner, P. A. Ulutaş, D. Beyaz, S.

Büyükyörük, A. Yakan, and A. G. Önol. 2015. Effect of dietary probiotic and high stocking density on the performance, carcass yield, gut microflora, and stress indicators of broilers. Poult. Sci. 94:2395–2403.

Chicken Farmers of Canada. 2019. Chicken Farmers of Canada Annual Report. Accessed March 2021. Available at https://www.chickenfarmers.ca/wp-content/uploads/2020/04/Annual-report-2019-—-E-Web2.pdf.

Clark, S., G. Hansen, P. McLean, P. Bond, W. Wakeman, R. Meadows, and S. Buda. 2002. Pododermatitis in Turkeys. Avian Dis. 46:1038–1044.

Classen, H. L., C. Riddell, F. E. Robinson, P. J. Shand, and A. R. Mccurdy. 1994. Effect of lighting treatment on the productivity, health, behaviour and sexual maturity of heavy male turkeys. Br. Poult. Sci. 35:215–225.

Coleman, J. W., and A. T. Leighton. 1969. The effect of population density on the production of market turkeys. Poult. Sci. 48:685–693.

Collins, L. M., and D. J. T. Sumpter. 2007. The feeding dynamics of broiler chickens. J. R. Soc. Interface 4:65–72.

Cravener, T. L., W. B. Roush, and M. M. Mashaly. 1992. Broiler production under varying population densities. Poult. Sci. 71:427–433.

Crespo, R., G. Y. Ghazikhanian, and C. I. Hall. 2002. Avulsion of the common retinaculum in meat turkeys. Avian Dis. 46:245–248.

Dalton, H. A., B. J. Wood, and S. Torrey. 2013. Injurious pecking in domestic turkeys: development, causes, and potential solutions. Worlds. Poult. Sci. J. 69:865–876.

Davami, A., M. J. Wineland, W. T. Jones, R. L. Ilardi, and R. A. Peterson. 1987. Effects of population size, floor space, and feeder space upon productive performance, external appearance, and plasma corticosterone concentration of laying hens. Poult. Sci. 66:251–257.

Dawkins, M. S. 1990. From an animal's point of view: Motivation, fitness, and animal welfare. Behav. Brain Sci. 13:1–61.

Dawkins, M. S. 2018. Stocking density: can we judge how much space poultry need?Pages 227–242 in Advances in Poultry Welfare. Woodhead Publishing, Oxford, UK.

Dawkins, M., C. A. Donnelly, T. A. Jones, and M. Dawkins. 2004. Chicken welfare is influenced more by housing conditions than by stocking density. Nature 427:342–344.

Denbow, D. M., A. T. Leighton, and R. M. Hulet. 1984. Behavior and growth parameters of large white turkeys as affected by floor space and beak trimming. I. Males. Poult. Sci. 63:31–37.

Dozier, W. A., J. P. Thaxton, S. L. Branton, G. W. Morgan, D. M. Miles, W. B. Roush, B. D. Lott, and Y. Vizzier-Thaxton. 2005. Stocking density effects on growth performance and processing yields of heavy broilers. Poult. Sci. 84:1332–1338.

Duggan, G., T. Widowski, M. Quinton, and S. Torrey. 2014. The development of injurious pecking in a commercial turkey facility. J. Appl. Poult. Res. 23:280–290.

Duncan, I. J. H. 1998. Behavior and behavioral needs. Poult. Sci. 77:1766–1772.

Duncan, I. J. H., E. R. Beatty, P. M. Hocking, and S. R. I. Duff. 1991. Assessment of pain associated with degenerative hip disorders in adult male turkeys. Res. Vet. Sci. 50:200–203.

Duncan, I. J. H., and D. G. M. Wood-Gush. 1972. An analysis of displacement preening in the domestic fowl. Anim. Behav. 20:68–71.

Erasmus, M. A. 2017. A review of the effects of stocking density on turkey behavior, welfare, and productivity. Poult. Sci. 96:2540–2545.

Erasmus, M. A. 2018. Welfare issues in turkey production.Pages 263–292 in Advances in Poultry Welfare. West Lafayette, US.

Estevez, I. 2007. Density allowances for broilers: where to set the limits? Poult. Sci. 86:1265–1272.

Feddes, J. J. R., E. J. Emmanuel, and M. J. Zuidhof. 2002. Broiler performance, body weight variance, feed and water intake, and carcass quality at different stocking densities. Poult. Sci. 81:774–779.

Feddes, J. . J. R., and Z. J. Licsko. 1993. Air quality in commercial broiler housing. Can. Agric. Eng. 35:147–150.

Forkman, B., and L. Keeling. 2009. Assessment of Animal Welfare Measure for Layers and Broilers. Welfare Quality Reports no. 9. Cardiff, UK.

Fraser, D. 2008. Chapter 8: Affective States. Pages 1–161 in Understanding Animal Welfare: the science in its cultural context. Wiley-Blackwell Publishing, Oxford, UK.

Garner, J. P., C. Falcone, P. Wakenell, M. Martin, and J. A. Mench. 2002. Reliability and validity of a modified gait scoring system and its use in assessing tibial dyschondroplasia in broilers. Br. Poult. Sci. 43:355–363.

Genovese, K. J., H. Haiqi, V. K. Lowry, C. L. Swaggerty, and M. H. Kogut. 2006. Comparison of heterophil functions of modern commercial and wild-type Rio Grande turkeys. Avian Pathol. 35:217–223.

Genovese, K. J., H. He, C. L. Swaggerty, and M. H. Kogut. 2013. The avian heterophil. Dev. Comp. Immunol. 41:334–340.

Gentle, M. J., and L. N. Hunter. 1991. Physiological and behavioural responses associated with feather removal in Gallus gallus var domesticus. Res. Vet. Sci. 50:95–101.

Gentle, M. J., V. Tilston, and D. E. F. McKeegan. 2001. Mechanothermal nociceptors in the scaly skin of the chicken leg. Neuroscience 106:643–652.

Gill, D. J., and A. T. Leighton. 1984. Effects of light environment and population density on growth performance of male turkeys: 2. Physiological changes. Poult. Sci. 63:1314–1321.

Goo, D., J. H. Kim, H. S. Choi, G. H. Park, G. P. Han, and D. Y. Kil. 2019. Effect of stocking density and sex on growth performance, meat quality, and intestinal barrier function in broiler chickens. Poult. Sci. 98:1153–1160.

Greene, J. A., R. M. Mccracken, and R. T. Evans. 1985. A contact dermatitis of broilers-clinical and pathological findings. Avian Pathol. 14:23–38.

Gross, W. B., and H. S. Siegel. 1983. Evaluation of the heterophil/lymphocyte ratio as a measure of stress in chickens. Avian Dis. 27:972–979.

Guenthner, E., C. W. Carlson, and J. Emerick. 1978. Copper salts for growth stimulation and alleviation of aortic rupture losses in turkeys. Poult. Sci. 57:1313–1324.

Gunther, P., and W. Bessei. 2006. Behavioural reactions of growing turkeys to different stocking densities.Pages 1–4 in Proc. 12th European Poult. Conf. World’s Poult. Sci. Assoc. Verona, Italy.

Hafez, H. M., N. Hagen, and T. S. Allam. 2016. Influence of stocking density on health condition in meat turkey flocks under field conditions. Pak. Vet. J. 36:134–139.

Hall, A. L. 2001. The effect of stocking density on the welfare and behaviour of broiler chickens reared commercially. Anim. Welf. 10:23–40.

He, S. P., M. A. Arowolo, R. F. Medrano, S. Li, Q. F. Yu, J. Y. Chen, and J. H. He. 2018. Impact of heat stress and nutritional interventions on poultry production. Worlds. Poult. Sci. J. 74:647–664.

Heckert, R. A., I. Estevez, E. Russek-Cohen, and R. Pettit-Riley. 2002. Effects of density and perch availability on the immune status of broilers. Poult. Sci. 81:451–457.

Hocking, P. M., R. K. Mayne, R. W. Else, N. A. French, and J. Gatcliffe. 2008. Standard European footpad dermatitis scoring system for use in turkey processing plants. Worlds. Poult. Sci. J. 64:323–328.

Huff, G., W. Huff, N. Rath, A. Donoghue, N. Anthony, and K. Nestor. 2007. Differential effects of sex and genetics on behavior and stress response of turkeys. Poult. Sci. 86:1294–1303.

Hughes, B. O., N. L. Carmichael, A. W. Walker, and P. N. Grigor. 1997. Low incidence of aggression in large flocks of laying hens. Appl. Anim. Behav. Sci. 54:215–234.

Hughes, B. O., and P. N. Grigor. 1996. Behavioural time-budgets and beak related behaviour in floor-housed turkeys. Anim. Welf. 5:189–198.

Human Farm Animal Care. 2014. Animal Care Standards. Accessed May 2021. Available at http://certifiedhumane.org/wp-content/uploads/2014/01/Std14.Turkeys.1A.pdf.

Hunter, R. R., M. A. Mitchell, and A. J. Carlisle. 2010. Wetting of broilers during cold weather transport: A major source of physiological stress? Br. Poult. Sci. 40:48–49.

Hurnik, J. F., A. B. Webster, and P. B. Siegel. 1985. Dictionary of farm animal behavior. Second. Iowa State University Press, Ames, Iowa.

Jankowski, J., D. Mikulski, M. R. Tatara, and W. Krupski. 2015. Effects of increased stocking density and heat stress on growth, performance, carcase characteristics and skeletal properties in turkeys. Vet. Rec. 176:21.

Jong, I. de, C. Berg, A. Butterworth, and I. Estevéz. 2012. Scientific report updating the EFSA opinions on the welfare of broilers and broiler breeders.

Julian, R. J. 1984. Tendon avulsion as a cause of lameness in turkeys. Avian Dis. 28:244–249.

Julian, R. J. 1998. Rapid growth problems: Ascites and skeletal deformities in broilers. Poult. Sci. 77:1773–1780.

Keeling, L. J. 1994. Inter-bird distances and behavioural priorities in laying hens: the effect of spatial restriction. Appl. Anim. Behav. Sci. 39:131–140.

Kilpinen, O., A. Roepstorff, A. Permin, G. Nørgaard-Nielsen, L. G. Lawson, and H. B. Simonsen. 2005. Influence of Dermanyssus gallinae and Ascaridia galli infections on behaviour and health of laying hens (Gallus gallus domesticus). Br. Poult. Sci. 46:26–34.

Knierim, U. 2013. Effects of stocking density on the behaviour and bodily state of broilers fattened with a target live weight of 2 kg. Berl. Munch. Tierarztl. Wochenschr. 126:149–155.

Kowalski, A., P. Mormede, K. Jakubowski, and M. Jedlinska-Krakowska. 2002. Comparison of susceptibility to stress in two genetic lines of turkey broilers BUT-9 and Big-6. Pol. J. Vet. Sci. 5:145–150.

Krista, L. M., P. E. Waibel, and R. E. Burger. 1965. The influence of dietary alterations, hormones, and blood pressure on the incidence of dissecting aneurysms in the turkey. Poult. Sci. 44:15–22.

Krista, L. M., P. E. Waibel, R. N. Shoffner, and J. H. Sautter. 1969. A study of aortic rupture and performance as influenced by selection for hypertension and hypotension in the turkey. Poult. Sci. 49:405–411.

Leeson, S., and W. D. Morrison. 1978. Effect of feather cover on feed efficiency in laying birds. Poult. Sci. 57:1094–1096.

Leighton, A. T., D. M. Denbow, and R. M. Hulet. 1985. Behavior and growth parameters of large white turkeys as affected by floor space and beak trimming. II. Females. Poult. Sci. 64:440–446.

Li, X. M., M. H. Zhang, S. M. Liu, J. H. Feng, D. D. Ma, Q. X. Liu, Y. Zhou, X. J. Wang, and S. Xing. 2019. Effects of stocking density on growth performance, growth regulatory factors, and endocrine hormones in broilers under appropriate environments. Poult. Sci. 98:6611–6617.

Lindberg, A. C., and C. J. Nicol. 1997. Dustbathing in modified battery cages: Is dustbathing an adequate substitute? Appl. Anim. Behav. Sci. 55:113–128.

Martland, M. F. 1984. Wet litter as a cause of plantar pododermatitis, leading to foot ulceration and lameness in fattening turkeys. Avian Pathol. 13:241–252.

Martrenchar, A., E. Boilletot, D. Huonnic, and F. Pol. 2002. Risk factors for foot-pad dermatitis in chicken and turkey broilers in France. Prev. Vet. Med. 52:213–226.

Martrenchar, A., D. Huonnic, and J. P. Cotte. 2001. Influence of environmental enrichment on injurious pecking and perching behaviour in young turkeys. Br. Poult. Sci. 42:161–170.

Martrenchar, A., D. Huonnic, J. P. Cotte, E. Boilletot, and J. P. Morisse. 1999. Influence of stocking density on behavioural, health and productivity traits of turkeys in large flocks. Br. Poult. Sci. 40:323–331.

Martrenchar, A., J. P. Morisse, D. Huonnic, and J. P. Cotte. 1997. Influence of stocking density on some behavioural, physiological and productivity traits of broilers. Vet. Res. 28:473–480.

Maxwell, M. H. 1993. Avian blood leucocyte responses to stress. Worlds. Poult. Sci. J. 49:34–43.

Maxwell, M. H., and G. W. Robertson. 1998. The avian heterophil leucocyte: A review. Worlds. Poult. Sci. J. 54:168–178.

Mayne, R. K. 2005. A review of the aetiology and possible causative factors of foot pad dermatitis in growing turkeys and broilers. Worlds. Poult. Sci. J. 61:256–267.

Mayne, R. K., R. W. Else, and P. M. Hocking. 2007. High litter moisture alone is sufficient to cause footpad dermatitis in growing Turkeys. Br. Poult. Sci. 48:538–545.

McFarlane, J. M., and S. E. Curtis. 1989. Multiple concurrent stressors in chicks. 3. Effects on plasma corticosterone and the heterophil:lymphocyte ratio. Poult. Sci. 68:522–527.

McFarlane, J. M., S. E. Curtis, R. D. Shanks, and S. G. Carmer. 1989. Multiple concurrent stressors in chicks. 1. Effect on weight gain, feed intake, and behavior. Poult. Sci. 68:501–

509.

McGeown, D., T. C. Danbury, A. E. Waterman-Pearson, and S. C. Kestin. 1999. Effect of carprofen on lameness in broiler chickens. Vet. Rec. 144:668–671.

Miller, K. A., and J. A. Mench. 2005. The differential effects of four types of environmental enrichment on the activity budgets, fearfulness, and social proximity preference of Japanese quail. Appl. Anim. Behav. Sci. 95:169–187.

Moinard, C., P. D. Lewis, G. C. Perry, and C. M. Sherwin. 2001. The effects of light intensity and light source on injuries due to pecking of male domestic Turkeys (Meleagris gallopavo). Anim. Welf. 10:131–139.

Moran, E. T. J. 1985. Effect of toeclipping and pen population density on performance and carcass quality of large turkeys reared sexes separately. Poult. Sci. 64:226–231.

Nagaraja, K. V, D. A. Emery, K. A. Jordan, J. A. Newman, and B. S. Pomeroy. 1983. Scanning electron microscopic studies of adverse effects of ammonia on tracheal tissues of turkeys. Am. J. Vet. Res. 44:1530–1536.

Nation Turkey Federation. 2012. Animal care best management practices. Accessed May 2021. Available at http://www.eatturkey.com/sites/ default/files/welfarm2012.pdf.

National Farm Animal Care Council. 2016. Code of practice for the care and handling of hatching eggs, breeders, chickens, and turkeys. Accessed May 2019. Available at https://www.nfacc.ca/pdfs/codes/poultry_code_EN.pdf.

Newberry, R. C. 1995. Environmental enrichment: Increasing the biological relevance of captive environments. Appl. Anim. Behav. Sci. 44:229–243.

Newberry, R. C. 1999. Exploratory behaviour of young domestic fowl. Appl. Anim. Behav. Sci. 63:311–321.

Nicol, C. J. 1989. Social influences on the comfort behaviour of laying hens. Appl. Anim. Behav. Sci. 22:75–81.

Noll, S. L., M. E. El-Halawani, P. E. Waibel, P. Redig, and K. A. Janni. 1991. Effect of diet and population density on male turkeys under various environmental conditions. Poult. Sci. 70:923-934.

Petek, M., H. Üstüner, and D. Yesilbag. 2014. Effects of stocking density and litter type on litter quality and growth performance of broiler chicken. Kafkas Univ. Vet. Fak. Derg. 20:743–748.

Petherick, C. J., E. Seawright, D. Waddington, I. J. H. Duncan, and L. B. Murphy. 1995. The role of perception in the causation of dustbathing behaviour in domestic fowl. Anim. Behav. 49:1521–1530.

Proudfoot, F. G., H. W. Hulan, and W. F. Dewitt. 1979a. Response of turkey broilers to different stocking densities, lighting treatments, toe clipping, and intermingling the sexes. Poult. Sci. 58:28–36.

Proudfoot, F. G., H. W. Hulan, and D. R. Ramey. 1979b. The effect of four stocking densities on broiler carcass grade, the incidence of breast blisters, and other performance traits. Poult. Sci. 58:791–793.

Puron, D., R. Santamaria, J. C. Segura, and J. L. Alamilla. 1995. Broiler performance at different stocking densities. J. Appl. Poult. Res. 4:55–60.

Puvadolpirod, S., and J. P. Thaxton. 2000. Model of physiological stress in chickens 1. Response parameters. Poult. Sci. 79:363–369.

Ritz, C. W., B. D. Fairchild, and M. P. Lacy. 2004. Implications of ammonia production and emissions from commercial poultry facilities: A review. J. Appl. Poult. Res. 13:684–692.

Ross, P. A., and J. F. Hurnik. 1983. Drinking behaviour of broiler chicks. Appl. Anim. Ethol. 11:25–31.

Rudolph, M. 2008. Influence of stocking density and litter material on pododermatitis in turkeys.

Sarica, M., S. Boga, and U. S. Yamak. 2008. The effects of space allowance on egg yield, egg quality and plumage condition of laying hens in battery cages. Czech J. Anim. Sci. 53:346–353.

Savory, C. J. 1995. Feather pecking and cannibalism. Worlds. Poult. Sci. J. 51:215–219.

Schwean-Lardner, K., D. Anderson, M. Petrik, S. Torrey, T. M. Widowski, K. Jennifer, and B. Gardner. 2013. Code of practice for the care and handling of chickens, turkeys and breeders: review of scientific research on priority issues. Natl. Farm Anim. Care Counc.

Accessed June 2019. Available at https://www.nfacc.ca/resources/codes-of-practice/chickens-turkeys-and-breeders/Poultry_SCReport_Nov2013.pdf.

Sherwin, C. M., and A. Kelland. 1998. Time-budgets, comfort behaviours and injurious pecking of turkeys housed in pairs. Br. Poult. Sci. 39:325–332.

Sherwin, C. M., P. D. Lewis, and G. C. Perry. 1999. Effects of environmental enrichment, fluorescent and intermittent lighting on injurious pecking amongst male turkey poults. Br. Poult. Sci. 40:592–598.

Simitzis, P. E., E. Kalogeraki, M. Goliomytis, M. A. Charismiadou, K. Triantaphyllopoulos, A. Ayoutanti, K. Niforou, A. L. Hager-Theodorides, and S. G. Deligeorgis. 2012. Impact of stocking density on broiler growth performance, meat characteristics, behavioural components and indicators of physiological and oxidative stress. Br. Poult. Sci. 53:721–730.

Siopes, T. D. 2010. Initiation of egg production by turkey breeder hens: Sexual maturation and age at lighting. Poult. Sci. 89:1490–1496.

Sørensen, P., G. Su, and S. C. Kestin. 2000. Effects of age and stocking density on leg weakness in broiler chickens. Poult. Sci. 79:864–870.

Temple, D., X. Manteca, D. Escribano, M. Salas, E. Mainau, E. Zschiesche, I. Petersen, R. Dolz, and E. Thomas. 2020. Assessment of laying-bird welfare following acaricidal treatment of a commercial flock naturally infested with the poultry red mite (Dermanyssus gallinae). PLoS One 15:1–16.

Thaxton, J. P., W. A. Dozier, S. L. Branton, G. W. Morgan, D. W. Miles, W. B. Roush, B. D. Lott, and Y. Vizzier-Thaxton. 2006. Stocking density and physiological adaptive responses of broilers. Poult. Sci. 85:819–824.

Thomas, D. G., V. Ravindran, D. V. Thomas, B. J. Camden, Y. H. Cottam, P. C. H. Morel, and C. J. Cook. 2004. Influence of stocking density on the performance, carcass characteristics and selected welfare indicators of broiler chickens. N. Z. Vet. J. 52:76–81.

Thomas, D. G., J.-H. Son, V. Ravindran, and D. V. Thomas. 2011. The effect of stocking density on the behaviour of broiler chickens. Korean J. Poult. Sci. 38:1–4.

Torrey, S., R. Bergeron, T. Widowski, N. Lewis, T. Crowe, J. A. Correa, J. Brown, H. W.

Gonyou, and L. Faucitano. 2013. Transportation of market-weight pigs: Effect of season, truck type, and location within truck on behavior with a two-hour transport. J. Anim. Sci. 91:2863–2871.

Turkey Farmers of Canada. 2019. Canadian Turkey Stats 1974-2019. Can. Turkey Mark. Agency Accessed March 2021. Available at https://www.turkeyfarmersofcanada.ca/wp-content/uploads/2020/09/Canadian-Turkey-Stats-1974-to-2019-Final.pdf.

Vermette, C., K. Schwean-Lardner, S. Gomis, B. H. Grahn, T. G. Crowe, and H. L. Classen. 2016. The impact of graded levels of day length on turkey health and behaviour to 18 weeks of age. Poult. Sci. 95:1223–1237.

Vestergaard, K. 1982. Dust-bathing in the domestic fowl - diurnal rhythm and dust deprivation. Appl. Anim. Ethol. 8:487–495.

Viegas, S., V. M. Faísca, H. Dias, A. Clérigo, E. Carolino, and C. Viegas. 2013. Occupational exposure to poultry dust and effects on the respiratory system in workers. J. Toxicol. Environ. Health 76:230–239.

Wathes, C. M., M. R. Holden, R. W. Sneath, R. P. White, and V. R. Phillips. 1997. Concentrations and emission rates of aerial ammonia, nitrous oxide, methane, carbon dioxide, dust and endotoxin in UK broiler and layer houses. Br. Poult. Sci. 38:14–28.

Weber Wyneken, C., A. Sinclair, T. Veldkamp, L. J. Vinco, and P. M. Hocking. 2015. Footpad dermatitis and pain assessment in turkey poults using analgesia and objective gait analysis. Br. Poult. Sci. 56:522–530.

Wilkins, L. J., S. N. Brown, A. J. Phillips, and P. D. Warriss. 2003. Cleanliness of broilers when they arrive at poultry processing plants. Vet Rec 153:701–703.

Woźniakowski, G., and E. Samorek-Salamonowicz. 2014. Direct detection of Marek's disease virus in poultry dust by loop-mediated isothermal amplification. Arch. Virol. 159:3083–3087.

Zuidhof, M. J., J. J. R. Feddes, F. E. Robinson, and S. L. Perkins. 1995. Effect of stocking density on air quality and health and performance of heavy torn turkeys. Can. Agric. Eng. 37:109–112.

Zuidhof, M. J., J. J. R. Feddes, F. E. Robinson, and C. Riddell. 1993. Effect of ventilation rate and stocking density on turkey health and performance. J. Appl. Poult. Res. 2:123–129.