

**A Comparison of Leg Health, Carcass Traits, and Muscle Disorders in
Strains of Broiler Chickens Selected for Distinct Growth Rates**

by

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ABSTRACT

A COMPARISON OF LEG HEALTH, CARCASS TRAITS, AND MUSCLE DISORDERS IN STRAINS OF BROILER CHICKENS SELECTED FOR DISTINCT GROWTH RATES

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The growth rate of broiler chickens has greatly increased over the past 60 years because of intense genetic selection, improvements in nutrition, and changes to management strategies. However, this increase in performance has resulted in some undesirable consequences that affect the welfare and meat quality of fast-growing broiler chickens. Thus, there has been an increasing interest in the use of slower-growing (SG) strains to mitigate problems associated with fast growth. In this context, the objectives of this thesis were to evaluate leg health, carcass traits, and incidence of muscle myopathies in 2 conventional (CONV) and 12 SG strains classified into 3 categories (FAST, MODERATE, and SLOW), based on their similarities of growth rate. All birds were raised under similar and controlled conditions and evaluated at similar target weights (TW) of 2.1 kg (TW 1) and 3.2 kg (TW 2). Results indicate that growth differences between categories affected most of the variables evaluated (e.g., carcass traits, incidence of myopathies, bone traits, and mobility) although some differences were also found among strains selected for a similar growth rate. Overall, CONV birds had greater breast yield than SG birds, which was accompanied by a greater incidence of myopathies. However, strain F, categorized as FAST, exhibited similar or greater breast yield and incidence of muscle myopathies compared to CONV birds, which was consistently higher than all of the other SG strains. At both TWs, CONV birds had shorter legs, yet similar or greater tibial breaking strength and ash content compared to the

SG categories. At TW 2, CONV birds exhibited indicators of poorer leg strength than SLOW birds, although the BW of these categories was similar. However, at both TWs, CONV birds showed indicators of similar walking ability (based on total obstacle crossing) compared to FAST birds, despite the lower BW of CONV vs. FAST birds. The large breast of CONV birds, combined with their shorter legs relative to their heavy body weight, might contribute to their reduced mobility. Altogether, these results indicate that not only differences in genetic potential for growth, but also differences in body conformation influence leg health and meat quality of broiler chickens.

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LIST OF ABBREVIATIONS

ADG: Average daily gain

BCO: Bacterial chondronecrosis with osteomyelitis

BW: Body weight

CONV: Conventional

D: Dark period

FAST: Fastest slow-growing

FG: Fast-growing

FPD: Footpad dermatitis

HB: Hock burns

L: Light period

LTL: Latency-to-lie

LW: Live weight

MOD: Moderate slow-growing

SG: Slower-growing

SLOW: Slowest slow-growing

SM: Spaghetti meat

TBS: Tibial breaking strength

TD: Tibial dyschondroplasia

TW: Target weight

WB: Wooden breast

WS: White striping

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Chapter 1: Introduction

The global demand of poultry meat has significantly increased over the past decades, with per capita supply increasing from 3 kg in 1963 to 15 kg in 2013 (FAO, 2020). The growing preference for chicken meat can be attributed to its high nutrient profile, variety of cut-up portions for simple preparation, lack of religious restrictions, and fewer environmental impact of chicken production compared to pork or beef production (Petracci et al., 2015; Clune et al., 2017). In response to the high market demand for chicken meat, substantial advances have been made in the poultry industry, resulting in strains of broiler chickens specialized in early growth and rapid muscle accretion (Petracci et al., 2015; Barbut, 2019). These strains, commonly referred to as conventional, commercial, or fast-growing (**FG**) chickens, are characterized by improved productive performance, high average daily gain, and early processing age compared to their unselected counterparts (Zuidhof et al., 2014). Despite the contribution of nutrition, management strategies, veterinary care, and housing conditions, it has been estimated that 85-90% of the increase in growth performance of FG birds is attributed to genetic selection (Havenstein et al., 1994a, 2003a).

In addition to the growing preference for chicken meat, a change in consumer behaviour has been observed over the past years. While poultry sold as whole birds have drastically decreased, the preference for cut-up portions and further processed products have significantly increased to meet the growing interest in easy preparation and fully prepared products (Barbut, 2020). The high demand for cut-up portions and preference for breast meat in Western countries have influenced not only the growth rate but also the body conformation of FG birds, resulting in heavier birds with large breasts compared to unselected birds at a similar age (Barbut, 2015).

The impact of the selection of broiler chickens for growth on body weight (**BW**) and conformation has been demonstrated by Havenstein et al. (1994b, 2003b) in that at a similar age and feeding regimen, FG birds from a 2001 strain were approximately 5-fold heavier and exhibited breast yield about twice as high as compared to an unselected strain from 1957. This rapid growth and improved feed efficiency of FG birds has decreased the rearing period and costs of production, due to reduction in labour, feed, water, and electricity, greatly contributing to the success of the broiler industry (Griffin and Goddard, 1994). However, several undesirable consequences have been linked to the accelerated growth of broiler chickens, including lameness and muscle myopathies, which have economic and welfare implications (Julian, 1998; Petracci et al., 2015; Kierończyk et al., 2017).

Lameness is a multifactorial disorder that impairs birds' locomotion and has been associated with fast growth (Julian, 1998; Sørensen et al., 1999; Kierończyk et al., 2017). Because lameness can prevent affected birds from accessing feeders and drinkers and possibly cause pain, lame birds are considered to have poor welfare (Bradshaw et al., 2002). The early and rapid increase in BW on an immature skeleton, combined with the large breast muscle, is thought to play a role in the high incidence of lameness of FG birds (Julian, 1998; Corr et al., 2003a; Shim et al., 2012b). Moreover, the increase in breast muscle mass and yield were concomitantly accompanied by the emergence of muscle disorders, including wooden breast and white striping (Petracci et al., 2015; Barbut, 2019). These muscle myopathies negatively affect consumer's acceptance, meat quality, and possibly welfare (Kuttappan et al., 2016; Norring et al., 2019). Therefore, different strategies have been adopted to decrease the incidence of both lameness and muscle disorders. Some of these interventions include dietary modifications, feed restriction, incubation conditions, and lighting

programs (Bizeray et al., 2002b; Brickett et al., 2007; Rault et al., 2017; Clark et al., 2017). Due to the lack of fully effective interventions to reduce the incidence of lameness and myopathies, the use of slower-growing (**SG**) strains has been suggested as an alternative to mitigate these disorders, although there is scarce information comparing SG and FG strains when birds are raised under similar conditions and evaluated at a similar BW, making comparisons between strains challenging. In this context, studies comparing a wide range of strains differing in growth are therefore crucial to determine the influence of selection for growth rate on the welfare and meat quality of broiler chickens.

Chapter 2: Literature Review

2.1 Objectives and Overview

The objective of this review is to provide an overview of leg and muscle disorders commonly found in broiler chickens and the economic and welfare implications of these disorders for the poultry industry. The intensification and evolution of the broiler industry, as well as the emphasis on genetic selection for production traits, will be introduced to provide a better understanding of the changes observed in the growth, anatomy, and physiology of FG birds over the past 60 years. Next, the concept of animal welfare will be discussed to provide a link between selection for growth and birds' welfare. Afterwards, the use of alternatives strategies to decrease lameness and modern muscle myopathies will be discussed. Lastly, selection for robustness and the use of SG strains as an attempt to improve broiler chickens' welfare and meat quality are reviewed, including limitations of the use of SG strains, gaps in the literature, and future directions.

2.2 The Evolution of the Broiler Industry

Human population growth and changes in eating habits, with an increasing global preference for animal products, have contributed to the growth and evolution of the poultry industry (Szöllősi et al., 2014). Chicken meat is the most consumed meat in Canada and the USA, and recent data suggest this trend will continue (FAO, 2020; OECD/FAO, 2020). As a result, the broiler industry has developed into an organized and competitive sector, using specialized strains selected for rapid muscle accretion and growth (Siegel et al., 2009). Selection for production traits in broiler chickens is a fairly recent event (about 80 years ago) relative to the domestication of chickens, which is a result of a long process that commenced over 8,000 years ago during the Neolithic period (Schmidt et al., 2009; Siegel et al., 2009; Lyimo et al., 2014). Chickens were domesticated predominantly

from the red junglefowl (*Gallus gallus*) originating from South East Asia but also including to a lesser extent the *Gallus sonneratii* from South-West India and *Gallus lafayetii* from Sri Lanka (reviewed by Lyimo et al., 2014).

The dispersion of chickens to Europe and Africa occurred mainly as a result of human migration (Tixier-Boichard et al., 2011). Although chicken domestication was possibly initiated for entertainment, religious, and cultural purposes (Wood-Gush, 1959), later chickens began to be used as a food source, with emphasis on egg-laying in the early stages of selection (Wood-Gush, 1959; Schmidt et al., 2009). However, in the 20th century, the advent of industrial agriculture and intensive farming triggered the selection of specialized birds for either meat or egg production (Wood-Gush, 1959; Schmidt et al., 2009). Chickens' general eating habits, adaptability to different environments, promiscuity, and early maturity all contributed to their domestication and widespread use of chickens as a food source (Siegel et al., 2009).

The intensification of broiler production was in part motivated by the “Chicken of Tomorrow” Committee in 1945, in which birds were selected based on characteristics related to improved productive performance and growth (Elfick, 2012). Dual-purpose chickens (raised for both meat and eggs), commonly raised in small-scale farming, were gradually replaced by specialized and efficient strains for meat production raised in commercial poultry houses (Siegel et al., 2009; Elfick, 2012). This focus on productive performance resulted in more efficient birds, which required less food and time to reach a market weight compared to unselected birds (Griffin and Goddard, 1994). Although improvements in nutrition, health, housing, and management play a role in the continued success and evolution of the broiler industry, genetic selection has been considered the main contributing factor to the significant improvements in growth rate and

efficiency observed in broiler chickens (Havenstein et al., 1994a, 2003a). To determine the role of genetic selection and nutrition on the productive performance of broiler chickens, two studies were conducted comparing one strain from 1957 with a commercial strain from 1991 (Havenstein et al., 1994a) and 2001 (Havenstein et al., 2003a). In both studies, the unselected strain from 1957 and the commercial strains from 1991 or 2001 were fed either the representative diet from 1957 or a commercial diet from each respective year (1991 or 2001). Irrespective of the diet, the commercial strain of broiler chickens was over three times heavier than the unselected strain from 1957, suggesting that genetics is the major contributor to the performance of FG strains of broiler chickens. The effects of genetic selection for production traits are not limited to growth and feed efficiency. Besides the substantial increase in BW and muscle accretion, FG birds have a different body conformation, with greater carcass and breast yield compared to unselected birds (Havenstein et al., 1994b, 2003b).

As a result of the intensification of the broiler industry, significant changes in productive traits, and a growing demand for chicken meat, the growth performance of broiler chickens increased over 400% over the past 60 years, with the global production of meat chickens increasing over ten times in the same period (Zuidhof et al., 2014; FAO, 2020). However, the emphasis on early and rapid growth has been linked to many disorders that may have negative impacts on the welfare of broiler chickens.

2.3 Definition of Animal Welfare

When determining how selection for growth can influence the welfare of broiler chickens, it is important to define animal welfare. Welfare is a broad term that encompasses the physical and mental well-being of the animal (Brambell Committee, 1965). According to the World

Organization for Animal Health, good animal welfare is present if the animal is “healthy, comfortable, well-nourished and not suffering from an unpleasant state, such as fear, pain, and distress” (OIE, 2010). Welfare can be scientifically assessed using scientific evidence and unbiased measurements (Bradshaw et al., 2002). The scientific term of animal welfare refers to characteristics intrinsic to the animal, rather than something that is given to them (Broom, 1988; Bradshaw et al., 2002). There is still divergence among scientists on the definition of animal welfare, as several scientific approaches to describe animal welfare vary according to ethical concerns, values, and emphasis (Keeling et al., 2011). A widely accepted framework to conceptualize animal welfare was proposed by Fraser (2008). This framework encompasses the most common approaches and viewpoints about animal welfare, named affective state, natural living, and biological functioning, with the study of affective state being the most accepted approach among animal welfare scientists to determine the animal’s state (Duncan, 2005; Keeling et al., 2011)

The concept of affective state considers the animal’s perception, reaction, emotions, and subjective experience to their environment, management, and rearing conditions provided (Fraser, 2008). It takes into account an animal’s physical and psychological status and considers the absence of negative emotions (i.e., fear, pain, frustration, and in some species, boredom and depression) as an indicator of good welfare (Mellor, 2016). The affective state approach also considers the association of positive emotional states, such as pleasure and excitement, with welfare (Fraser, 2008; Mellor, 2014). Short-term experiences of negative affective states influence survival by directing animal behaviour to obtain sustaining primary resources (those that are survival-related), reducing the exposure to danger and harmful conditions, and enabling recovery from injuries,

leading to responses that increase survival rates (Duncan 2005; Mellor, 2016). On the other hand, a positive affective state can fulfill secondary needs (such as maintenance of health) that promotes long-term fitness. For example, dust-bathing, previously considered a need-driven behaviour in domestic fowl that led to frustration and negative feelings if prevented, may be performed in “opportunity situations”, conditions in which all the animal’s essential needs are met, and the performance of the behaviour results in a state of pleasure when the opportunity is present (Widowski and Duncan, 2000). Because affective states cannot be directly measured, animal behaviour can be used as a proxy to assess and quantify the affective state of the subject (Gonyou, 1994; Duncan, 2005).

The natural living viewpoint encompasses the ability of an animal to perform a wide variety of behaviour commonly expressed in its species and live a “natural” life with access to natural environments (e.g., outdoor access) (Fraser, 2008). Environments that enable the expression of natural behaviour are known to promote animal welfare by reducing suffering and frustration (Gonyou, 1994). In fact, the performance of some behaviours plays a crucial role in the animal well-being due to their value for the animal (Duncan, 1998), even if the function of that behaviour is not relevant to animals raised under human management (Gonyou, 1994). An example of a behavioural need is foraging behaviour, which is performed even by chickens provided with *ad libitum* access to feed (Dawkins, 1989). Due to the high motivation to express behavioural needs, an environment that inhibits their performance is associated with frustration and poor welfare (Duncan, 1998). In this case, there is an overlap of two welfare viewpoints, as the inability to perform a highly motivated behaviour (natural living) will negatively influence the animal’s affective state because of frustration

The biological approach highlights the connection between welfare, health, and physiological stressors. It considers the ability of the animal to cope with its environment, grow, reproduce, and satisfy its biological needs as indicators of good welfare (Duncan, 2005; Fraser, 2008). Different coping strategies, including immunological, physiological, and behavioural responses, can be adopted by the animal to adapt to the environment, achieve biological functioning, and reach homeostasis (Fraser, 2008; Broom, 2014). The biological functioning approach also considers the effects of coping strategies on the animal. Therefore, health and physiological stress responses are crucial components of the biological functioning approach (Barnett and Hemsworth, 2009).

The concept referred to as “The Five Freedoms”, described by the Farm Animal Welfare Council, is another widely accepted definition of animal welfare (FAWC, 2009). It encompasses basic pillars to maximize welfare and prevent negative experiences to the animal, considering biological, physiological, and behavioural aspects, and was later expanded to include provisions for each freedom (Webster, 1994; Mellor, 2016).

The five freedoms and provisions include:

- Freedom from thirst, hunger, and malnutrition - By providing access to fresh water and a balanced diet that promotes health and vigor.
- Freedom from discomfort - By providing appropriate environmental housing, shelter, and resting area.
- Freedom from pain, injury, and disease - By preventing or providing rapid diagnosis of diseases followed by treatment.

- Freedom from fear and distress - By providing conditions and handling that prevent mental suffering.
- Freedom to express normal behaviour - By providing housing and facilities that contains enough space to allow a variety of behaviour repertoire.

Moving beyond the Five Freedoms, the concept of animal welfare has also focused on providing positive experiences rather than merely avoiding negative experiences and providing basic needs, as emphasized in previous definitions. The concept of “life worth living”, proposed by the Farm Animal Welfare Council in Great Britain (2009), states that the lifetime of the animal, including its manner of death, should be considered to determine the animal’s quality of life, which can range from a life not worth living to a good life, with the life worth living being intermediate. This concept highlights that both positive and negative experiences should be taken into account, with more positive experiences than negative experiences to ensure the animal’s quality of life.

Considering the broad concepts of animal welfare mentioned above, the genetic selection of broiler chickens focused on growth rate has been linked to profound changes in birds’ behaviour, physiology, and anatomy, which can directly or indirectly affect the welfare of FG chickens.

2.4 The Impact of Selection for Growth on the Welfare of Broiler Chickens

2.4.1 Resource Allocation Theory

The link between selection for production traits and poor welfare has been reported not only in broiler chickens but also in other poultry and livestock species (Huber, 2018), which has been partially attributed to the emphasis on the allocation of finite resources towards growth and production, compromising resources available for other biological functions and processes. This

concept, called “Resource Allocation Theory” states that negative associations (trade-offs) may occur in an organism as a consequence of limited resources (e.g., energy, nutrients, and time) being distributed among competing traits, including maintenance, ontogenic growth, production, and reproduction (Rauw et al., 1998). In the case of availability and abundance of resources, the traits sharing the resources will be positively correlated, whereas the scarcity of resources will lead to competition among traits, resulting in a negative correlation (Rauw et al., 1998).

As demonstrated in many species, the allocation of the resources can be influenced by artificial selection (Rauw, 2008), with the allocation of resources increasing for the selected trait, while decreasing the resources available for other competing traits (Rauw et al., 1998). Active selection for a trait of interest, such as early growth, will lead to disproportional allocation of resources for that particular trait, affecting the ability to properly distribute resources to other processes and demands, such as coping with disease and stress (Siegel and Dunnington, 1997; Rauw et al., 1998). In broiler chickens, selection has been mainly focused on rapid BW increase and the intensity of selection has been high and practiced over many generations, whereas in other livestock such as cattle and pigs, the selection has been less intensive, with fewer generations included as their generation interval is significantly longer (Rauw et al., 1998). While natural selection aims to maintain the equilibrium of many traits to optimize fitness, which allows the organism to slowly adapt to the changes in a wide range of environments, selection that occurs too rapidly and/or leads to substantial changes, may impair the ability of the organism to respond to such changes, disrupting the biological balance and proper allocation of resources (Dunnington, 1990)³. Thus, rapid changes in production traits likely decrease the ability of broilers to cope with these changes, increasing the propensity for maladies and disorders (Dunnington, 1990).

2.4.2 Heart Conditions

The accelerated growth rate of FG birds has been linked to heart failure, with ascites and sudden death syndrome being considered the most prevalent heart conditions in broiler chickens (Julian, 1998).

Ascites is a result of a mismatch between oxygen demand and supply to the tissues. Because fast growth requires a higher metabolic demand for oxygen than slower growth, there is an increase in pulmonary arterial pressure and blood flow to provide the amount of oxygen needed to keep up with growth (Julian, 1998). However, avian lungs are fixed in the thoracic cavity, only allowing a limited dilation of blood capillaries to cope with the increased blood flow (Julian, 1998). In order to increase oxygen supply, there is an increase in the number of red blood cells, which makes the blood more viscous, causing greater resistance to flow (Julian, 1998). The increase in blood flow to supply the high oxygen demand increases the blood pressure needed to pump the blood through the lungs, causing pulmonary hypertension (Acar et al., 1995; Julian, 1998). As a result of this increased workload, there is an enlargement in the right ventricle, which will continue to increase if pulmonary hypertension persists, causing many pathophysiological events, subsequently leading to a right ventricular failure (Julian, 1998; Gupta, 2011). The increase in blood pressure causes leakage of plasma fluids out of the vessels accumulating within the peritoneal space, resulting in ascites (Julian, 1998; Olkowski, 2007; Gupta, 2011). Pulmonary hypertension leading to ascites has been linked to the relatively inefficient cardio-respiratory system of FG birds that is unable to meet the high oxygen demand required to support the accelerated growth (Gupta, 2011). However, other environmental factors increase the susceptibility of birds to ascites, including high altitudes and low brooding temperatures (Bessei, 2006). Because ascites is a chronic disorder, its

development is gradual (Bessei, 2006; Olkowski, 2007). Thus, affected birds may suffer for a prolonged time before they die (Bessei, 2006).

Unlike ascites, sudden death syndrome occurs rapidly in birds that were apparently healthy and in good condition; there are generally a few seconds from the first sign of the syndrome to death (Bessei, 2006). However, birds that die from sudden death syndrome frequently have long-term signs of cardiac rhythm disturbances (Olkowski, 2007). Because feed restriction has been shown to reduce the susceptibility of broilers to cardiac rhythm disturbance, this disorder has been linked to fast growth (Olkowski, 2007).

2.4.3 Behaviour

Domestication and selection for growth have been linked to changes in the behaviour of broiler chickens. While the red junglefowl were found to spend about 10% of their day sitting (Dawkins, 1989), FG birds can spend up to 70 - 80% of their day sitting (Bizeray et al., 2000; Bokkers and Koene, 2003; Dixon, 2020), emphasizing their low activity compared to their ancestor. However, the behavioural repertoire of FG broilers and jungle fowl are similar, yet performed in different frequencies (Garnham and Løvlie, 2018).

In agreement with the differences found between FG birds and their ancestor, the red jungle fowl, previous studies demonstrated a decrease in activity and time spent perching, walking, and scratching, yet an increase in time spent lying, sitting, eating, and drinking in FG birds compared to SG strains (Bizeray et al., 2000; Bokkers and Koene, 2003; Lichovnicková et al., 2017; Dixon, 2020); these studies provide evidence of a similar behavioural repertoire but different time budget between SG and FG chickens (Bokkers and Koene, 2003). The impacts of growth rate on

behaviour of broiler chickens have been detected in birds as young as 2 to 3 days, when there is a small discrepancy in BW between FG and SG birds, indicating the influence of genetic factors on behaviour of chickens selected for different growth rates (Bizeray et al., 2000).

Indeed, it has been hypothesized that selection for faster growth has decreased the frequency of energy-consuming behaviours in order to prioritize resources for desirable production traits (Rauw et al., 1998; Bizeray et al., 2000). Therefore, the decrease in energetically costly behaviours (e.g., running and prolonged walking) observed in FG birds may occur in response to the high demand of resources directed towards growth and BW gain, leading to prolonged time resting and low activity levels. Experimentally increasing the pectoral mass of birds through the use of front packs resulted in a disproportional increase in energetic costs associated with locomotion compared to carrying similar loads on their back (Tickle et al., 2013). Because selection for growth has increased BW and also breast muscle mass and yield, changes in locomotion, and general activity levels may have occurred to cope with this modification in body conformation (Tickle et al., 2018).

Although the ability to perform species-specific behaviour is considered to be one of the pillars of animal welfare (Fraser, 2008; Bergmann et al., 2017), determining to which extent the changes in behaviour observed in broiler chickens affect their welfare may be difficult, as birds selected for accelerated growth may be less motivated to perform energetically costly behaviour (Bizeray et al., 2000). Behavioural needs are equivalent to psychological needs, leading to suffering and frustration if not performed (Weeks and Nicol, 2006). Thus, physical or environmental limitations preventing the birds from engaging in motivated species-specific behaviours can lead to frustration and decreased welfare (Bergmann et al., 2017). A study by Bokkers and Koene (2004) examined the impacts of growth rate on motivation and physical ability to gain access to a food reward after

a similar period of feed restriction in FG and SG birds. The researchers found that SG birds walked faster and had a shorter latency to start walking compared to FG birds (Bokkers and Koene, 2004). In addition, FG birds showed more preening than SG strains, possibly due to their inability to walk during the test on the runway test, leading to frustration. The authors concluded that while walking was mostly determined by motivation in lighter birds, physical ability appeared to be the major determinant factor for walking in heavier birds.

Locomotor activity declines, whereas time spent lying increases as broilers age (Weeks et al., 2000; Bokkers and Koene, 2004; Dixon, 2020). Although changes in behaviour are expected to occur in response to developmental transitions (Spear, 2004), broiler chickens are usually processed at 5 to 6 weeks of age, much earlier than when they would be expected to reach sexual maturity at approximately 20 weeks of age (Lewis et al., 2007). While the decrease in walking behaviour has been observed in both SG and FG birds, this decline is more evident in FG birds, in part due to the limitations in space as the birds grow heavier (Bokkers and Koene, 2003; Kjaer and Mench, 2009) or exacerbation of skeletal disorders that can lead to lameness, which will be discussed in section 2.4.5 (Julian, 1998; Bradshaw et al., 2002; Shim et al., 2012b). Interestingly, the reduction in activity and locomotion can aggravate skeletal problems as physical loading plays a crucial role in normal bone development and maintenance (Kjaer and Mench, 2009). Moreover, reduced locomotor activity is associated with other health implications, since more time spent lying or sitting increases birds' susceptibility to potentially painful contact dermatitis (e.g., hock burns and footpad dermatitis), especially if environmental conditions are suboptimal with poor litter quality (Bassler et al., 2013; De Jong et al., 2014).

2.4.4 Contact Dermatitis

Contact dermatitis is a skin condition commonly found in broiler chickens (Haslam et al., 2007; Bassler et al., 2013). These skin lesions can affect the feet (footpad dermatitis), hocks (hock burns), or breast (breast blisters). In severe cases, contact dermatitis is assumed to cause pain, impacting the welfare of affected birds (Haslam et al., 2007; Bassler et al., 2013). Therefore, the incidence and severity of contact dermatitis (especially hock burns and footpad dermatitis) are routinely determined either at the farm or slaughter plants as animal-based measures and indicators of rearing conditions and welfare (Ask, 2010; Bassler et al., 2013).

Footpad dermatitis (**FPD**) is also referred to as ammonia burns as the lesions are mainly caused by the extended period in contact with poor litter containing high moisture, ammonia, and other litter chemicals (Berg, 2009). The lesions are characterized by an initial discoloration affecting the plantar region of the feet in mild cases. Hyperkeratosis and necrosis of the epidermis can gradually occur followed by ulcerations, inflammation, and degeneration of the affected subcutaneous tissues in more severe cases (Ekstrand et al., 1997). Footpad dermatitis can become a gateway for bacteria, increasing the susceptibility to secondary infections (e.g., *Escherichia coli*) (Bessei, 2006).

Despite the contribution of environmental conditions (e.g., temperature, humidity, and ventilation), stocking density, feed composition, and litter quality, genetic selection influences the incidence of contact dermatitis (Ask, 2010). Numerous studies demonstrated a higher incidence of FPD and hock burns (**HB**) in FG birds compared to SG birds (Kjaer et al., 2006; Wilhelmsson et al., 2019; Dixon, 2020). The genetic factor associated with the propensity to develop FPD may be related to a dysregulation in biotin uptake, deposition, or utilization (Kjaer et al., 2006), as this

vitamin plays a role in the skin integrity of poultry species (Shepherd and Fairchild, 2010). Alternatively, the higher incidence of FPD in FG birds may be due to an increase in pressure on the footpads when walking or standing as the BW increases (Wylie, 1999; Mayne, 2005). However, while a genetic correlation was found between BW and HB with an increase in HB as the BW increased, this relationship has not been found between FPD and BW (Kjaer et al., 2006; Ask, 2010). Nevertheless, genetic selection mainly focused on increased BW gain, without considering FPD into the selection criteria, will likely increase the risk of broilers developing this contact dermatitis (Ask, 2010).

Rapid deterioration of litter quality, with an accelerated increase in moisture content, is considered the primary risk factor for FPD (Shepherd and Fairchild, 2010). Because FG birds grow at a faster rate compared to SG birds (Havenstein et al., 2003a; Zuidhof et al., 2014; Wilhelmsson et al., 2019; Dixon, 2020), litter deterioration may occur more rapidly in response to the accelerated accumulation of excreta as the birds grow. This rapid decrease in litter quality, combined with suboptimal environmental conditions (e.g., poor ventilation and high relative humidity and ammonia concentration) and prolonged time in contact with litter may increase birds' propensity to develop FPD.

Hock burns are closely related to FPD (Kjaer et al., 2006), being characterized by brown or black lesions on the plantar surface of the hocks of broiler chickens. The severity of HB may vary, with inflammation and skin ulceration being observed in more severe cases (Hepworth et al., 2010). As previously stated, increasing BW tends to increase the time spent sitting and decrease locomotion (Bokkers and Koene, 2003). Therefore, the higher propensity of FG birds to develop HB is likely attributed to their heavy BW and prolonged time sitting on the litter, as the weight of the bird

centered on their hocks while sitting (Dixon, 2020). The incidence of contact dermatitis may also influence walking ability, since severe lesions of FPD can result in unsteady walking (Hester, 1994) and partially contribute to the impaired locomotion observed in FG birds (Kjaer et al., 2006). In fact, both HB and FPD have been found to affect objective assessments of lameness, suggesting the contribution of contact dermatitis on the walking ability and leg strength of broiler chickens (Caplen et al., 2014). Even though birds' walking ability is affected by several different factors, selection for growth is often considered to increase birds' propensity to develop leg disorders and lameness (Bradshaw et al., 2002; Kierończyk et al., 2017).

2.4.5 Lameness

There is a growing body of scientific evidence suggesting a link between selection focusing on growth performance and the incidence of lameness (Julian, 1998; Bradshaw et al., 2002; Shim et al., 2012c; Kierończyk et al., 2017, Dixon, 2020). The terms “lameness” and “leg weakness” are commonly used to refer to several disorders resulted from infectious and non-infectious origins that may affect birds' bones, muscle, joints, tendons, skin, or nervous system (Bradshaw et al., 2002). For the purpose of this review, the term lameness will be used to describe impaired locomotion and leg disorders that reduce the welfare of affected birds.

Lame birds commonly show a wobbling walk and altered gait, frequently squatting or sitting while walking (Julian, 1984, 1998). Moreover, lameness affects birds' behaviour, with lame birds spending less time walking and standing than sound birds (Weeks et al., 2000; Bradshaw et al., 2002; Norring et al., 2019). Lameness is assumed to cause pain based on studies where lame birds showed a preference for food containing the analgesic drug carprofen and then had improvements in their walking ability followed administration of the analgesic (McGeown et al., 1999; Danbury

et al., 2000). In addition, lameness can make accessing the feeder and drinker difficult for affected birds, potentially leading to hunger, dehydration, and weight loss (Bradshaw et al., 2002).

The relationship between skeletal disorders and lameness has been well studied in broiler chickens. However, even apparently healthy broiler chickens based on gross and histological examinations show some potential signs of lameness and/or poor leg strength, with frequent sitting and prolonged squatting while walking. Therefore, it is unclear if the changes observed in lame birds occur in response to pain or discomfort in bones, tendons, ligaments, or muscles (Julian, 1998). Because lameness can be reduced by slowing the growth of broiler chickens, especially during the first 2 weeks of life, it has been suggested that the heavy and rapidly reached BW of FG birds, supported by a still immature skeletal system (Julian, 1998), likely causes lameness. While broiler chickens reach the market weight of 2.0 to 2.1 kg as early as 5 weeks of age, bone maturity occurs much later at 23 to 27 weeks of age (Rath et al., 2000; Sherlock et al., 2010). Moreover, in a typical production cycle of 6 weeks, long bones involved in body support, such as femur and tibia, increase approximately 4-fold in length, while the mid-shaft diameter grows 3- to 5-fold (Applegate and Lilburn, 2002), in comparison to a > 65-fold increase in BW in the same interval in FG birds (Aviagen, 2014). This rapid increase in BW relative to the slower bone development and maturation has been suggested to predispose FG birds to bone disorders that can cause lameness (Julian, 1998; Angel, 2007; Shaw et al., 2010).

Alternatively, bone structure rather than bone dimensions may be compromised, providing inadequate support for the rapid increase in BW. Poor bone quality as indicated by high porosity, and low mineral content, density, and breaking strength, has been reported in FG birds (Corr et al., 2003b; Williams et al., 2004; Shim et al., 2012a), likely as a result of selection focused on the

growth rate, leaving fewer resources available for other metabolic processes (Tallentire et al., 2016). Despite the negative association between walking ability and growth rate, bone health and skeletal integrity have been incorporated into breeding programs over the past couple of decades, with the aim to reduce bone disorders associated with lameness in broiler chickens (Angel, 2007; Whitehead, 2007).

To investigate the relationship between growth rate and leg strength, Neeteson-van Nieuwenhoven et al. (2013) evaluated several generations of broiler chickens from 1996 to 2012. Although growth rate was negatively associated with leg strength in each year, over time the authors observed an overall increase in leg health, indicating the possibility of improving bone health without greatly compromising BW gain. The incorporation of bone health in breeding programs, along with improvements in nutrition, health, and management strategies (Bradshaw et al., 2002; Fleming, 2008; Nääs et al., 2012; Kierończyk et al., 2017) have contributed to a reduction in bone disorders (McKay et al., 2000). However, globally, moderate to severe lameness (gait score equal or greater than 3) still affects 14 to 30 % of broiler chickens (Sanotra et al., 2003; Bassler et al., 2013; Kittelsen et al., 2017; Vasdal et al., 2018), indicating that leg health is an ongoing problem that must be addressed to improve the welfare of broiler chickens (Bradshaw et al., 2002). Most skeletal disorders resulting in lameness are related to long bone deformities, commonly involving the growth plate. The most common skeletal disorders associated with lameness include tibial dyschondroplasia, bacterial chondronecrosis with osteomyelitis, and valgus and varus deformities (Bradshaw et al., 2002).

Tibial dyschondroplasia (**TD**) is characterized by an abnormal accumulation of a non-vascularized mass of cartilage below the growth plate and extending into the metaphysis (Julian, 1998). The

lesion is often found in the proximal tibiotarsus although occurrence in other long bones has been reported (Julian, 1998). Tibial dyschondroplasia is a result of a failure in mineralization and disturbance in normal bone development (Bradshaw et al., 2002). Severe lesions can cause bone deformity or fracture, increasing the propensity for lameness, carcass downgrading, or condemnation (Julian, 1998; Bradshaw et al., 2002). Some studies suggest a failure of blood vessels to invade the growth plate as a primary cause of TD (Duff, 1984; Lynch et al., 1992). The pressure of the heavy BW of FG chickens on the growth plate blood vessels and prolonged time spent sitting may contribute to the failure in vascularization observed in TD lesions (Thorp, 1988; Bradshaw et al., 2002). Another study suggested that the lesions occur due to a failure in chondrocyte hypertrophy, preventing normal vascularization (Poulos et al., 1978). The peak of incidence of TD occurs at 3 weeks of age (Bradshaw et al., 2002), which coincides with a period where a 20-fold growth in BW is observed. This rapid and early increase in BW is likely involved in the incidence of TD in FG chickens.

Bacterial chondronecrosis with osteomyelitis (**BCO**) is also referred to as femoral head necrosis, long bone necrosis, bacterial chondronecrosis, and proximal femoral degeneration. It has been estimated that over 1% of broiler chickens are affected by BCO after 5 week of age (reviewed in Wideman, 2016). This disorder, first reported in 1972, is considered the major cause of lameness in commercial broilers, commonly affecting birds from 14 - 70 days of age, with higher incidence occurring around 35 days of age (McNamee and Smyth, 2000). Bacterial chondronecrosis with osteomyelitis is mainly caused by *Staphylococcus aureus*, although the involvement of other pathogenic bacteria such as *E. coli*, and *Enterococcus* spp. have been reported (McNamee and Smyth, 2000; Wideman et al., 2014). The pathogenesis of the disorder is not completely

understood, however the bacterial translocation from blood to exposed cartilage has been demonstrated (Wideman et al., 2014). The proximal femur and tibiotarsus are commonly affected due to the susceptibility of fracture and damage in enlarged growth plates, exposing the cartilage to bacterial infection (McNamee and Smyth, 2000). Stress and immunosuppression have also been associated with outbreaks of BCO, likely due to an increase in permeability and disruption of the gastrointestinal barrier, which facilitates bacterial translocation infection (McNamee and Smyth, 2000). The use of vitamin D3 and its metabolites have shown successful protective efficacy against BCO when birds are under stress, probably due to their capacity in decreasing immunosuppression and improving resistance to stress-related disorders (Huff et al., 2000; Wideman et al., 2014). The prolonged time spent sitting observed in FG birds may compress the major arteries supplying the femora and tibiae (Wideman, 2016). Interestingly, the insufficient blood supply to epiphyseal and physeal cartilage has been considered to be the most likely primary cause of osteochondrosis in different species (e.g., pigs, horses, dogs) (Ytrehus et al., 2007). Fast growth has been linked to poor mineralization of chondrocytes in the enlarged growth plates, which are more susceptible to the development of microfractures in response to mechanical stress (Wideman, 2016). Therefore, the extended time spent sitting combined with the heavy BW of broiler chickens may cause arterial compression, resulting in insufficient blood flow and likely higher susceptibility to develop BCO (Wideman, 2016).

Valgus and varus deformities cause lateral or medial angulation of the distal end of the tibiotarsus, resulting in deviation of the inferior part of the limb (Leterrier and Nys, 1992). Because valgus and varus angulation disorders show clinical, morphological, and anatomical differences, these deformations likely result from different etiologies (Leterrier and Nys, 1992; Shim et al., 2012a).

Varus deformations commonly appear between 5 to 15 days of age, mainly affecting the right limb and commonly associated with tendon displacement. The deformation is characterized by an inward rotation of the femora and medial angulation of the tibiotarsi and metatarsi (Leterrier and Nys, 1992). Valgus deformations occur more frequently than varus deformations. The deformity is often bilateral and gradually progresses with age, appearing between 2 to 7 weeks of age. Affected birds show angulation of tibiotarsi and lateral deviation of metatarsi, with a displacement of the gastrocnemius tendon occurring in some cases (Leterrier and Nys, 1992). In moderate and severe cases, these deformities may influence birds' locomotion, with affected birds having difficulty walking, a tendency to waddle or hobble, and frequent sitting while walking (Julian, 1984). In more severe cases, affected birds walk on their hocks. Moreover, valgus and varus angulation can result in carcass condemnation due to lesions or fractures caused by the deformities (Julian, 1984). Because valgus and varus angulations are heritable, genetic selection against these disorders has been successful in reducing their incidence (Mercer and Hill, 1984; Akbas et al., 2009). Moreover, based on a study that investigated leg disorders in birds selected for fast and slow-growth, a higher incidence was found in the former group, suggesting the deformity may be associated with growth potential (Shim et al., 2012a).

Despite the relevance of bone integrity to walking ability, lameness can also result from pain (Corr et al., 1998; McGeown et al., 1999; Caplen et al., 2013), biomechanical issues associated with body conformation, or both (Corr et al., 2003b). The large breast muscle of FG birds is thought to shift the centre of gravity cranially, likely altering the gait of FG birds (Paxton et al., 2014). Corr and colleagues conducted two studies to investigate the morphology of the musculoskeletal system (Corr et al., 2003b) and locomotion (Corr et al., 2003a) of FG and SG birds fed *ad libitum* or feed

restricted and culled at similar BW of 2.4 kg. Fast-growing birds fed *ad libitum* had larger breasts (total and relative to the BW), yet shorter legs and heavier thighs (total and relative to the BW) likely resulting in greater forces required by the short limbs to move the body compared to SG birds. In addition, FG birds fed *ad libitum* had lower bone ash content, possibly indicating poorer mineralization and weaker bones. These morphological changes in the musculoskeletal system were accompanied by several gait alterations. Fast-growing birds fed *ad libitum* walked slower, with lower cadences (steps per minute), shorter and wider steps, and longer periods with their feet in contact with the ground compared to the other groups of birds (Corr et al., 2003a). The researchers suggested that these gait alterations were likely used as a strategy to cope with the apparent instability in response to the changes in the aforementioned morphological traits. This altered gait appears to be inefficient, rapidly tiring the birds and likely contributing to the low activity levels observed in FG birds. In addition to the potential implications on walking ability, the musculoskeletal morphology of FG birds, resulting from selection focused on rapid growth and large breast muscle has been considered to influence the development of muscle disorders that have negative effects on the meat quality and potentially welfare of broiler chickens.

2.4.6 Modern Muscle Myopathies

The improvements in productive traits observed in broiler chickens have not been achieved without consequences. In the past ten years, concomitant with the continuous increase in growth rate and breast muscle mass, muscle abnormalities have been observed in broiler chickens (Petracci et al., 2019; Barbut, 2019). The negative effects of these muscle disorders on meat quality, appearance, nutritional profile, and consumer acceptance have been clearly demonstrated (Petracci et al., 2019). Although the precise etiology and underlining mechanisms are yet to be elucidated,

selection for increased BW gain and breast yield are considered to play a role in the incidence of modern myopathies, so-called wooden breast, white striping, and spaghetti meat (Petracci et al., 2015; Barbut, 2019).

Wooden breast (**WB**), also referred to as woody breast mostly affects the *Pectoralis major* and occasionally the *Pectoralis minor*. This myopathy is characterized by a distinct hardened texture, bulged areas, and pale appearance affecting different regions of the breast fillet (Petracci et al., 2019). Wooden breast can be classified from mild to severe, depending on the proportion of the breast affected and severity of the myopathy (Petracci et al., 2019). The lesion appears focally at earlier stages and gradually progresses to a diffuse and more severe fibrotic phase as the birds grow (Papah et al., 2017). In commercial processing plants, incidences as high as 60% have been observed (Xing et al., 2020), whereas under experimental conditions incidences above 95% have been recorded in some studies (Tijare et al., 2016; Bodle et al., 2018), while other studies found much lower values (Trocino et al., 2015; Gratta et al., 2019; Dixon, 2020). This discrepancy in results is likely due to the subjective evaluation, scoring systems adopted, and contribution of environmental factors to the incidence of WB. In fact, non-genetic factors (e.g., nutrition, incubation, management strategies) may play a major role in the development of both WB and white striping as suggested by Bailey et al. (2015, 2020). Analysis of proximal composition (i.e., moisture, protein, fat, and ash content) revealed that WB-affected breast fillets have greater fat and moisture, yet lower protein content compared to normal fillets (Soglia et al., 2016; Cai et al., 2018).

White striping (**WS**) is the most common muscle myopathy reported in broiler chickens. Recent findings reported that varying degrees of WS can affect over 90% of breast fillets (Kuttappan et al., 2017; Petracci et al., 2019; Che et al., 2020). This muscle abnormality is easily identified,

characterized by the presence of white striations parallel to the muscle fibres mainly present on the breast (*Pectoralis major*), but also found on the thigh, tenders (*Pectoralis minor*), and drumstick (Kuttappan et al., 2013a; c; Petracci et al., 2019). Microscopic examinations show that the white striations are a result of an accumulation of lipids and connective tissue (Kuttappan et al., 2013a). The incidence of WS negatively impacts consumer acceptance, which tends to decrease as the severity of the WS lesions increases (Kuttappan et al., 2012c).

Compared to normal breast meat, WB and WS-affected breasts exhibit heavier and thicker fillets, higher pH, and poorer water holding capacity as indicated by lower marinated uptake, higher drip loss, and cooking loss (Mudalal et al., 2015; Tijare et al., 2016; Cai et al., 2018; Xing et al., 2020). The lower cooking yield of WB-affected breasts may be caused by the accumulation of adipose and connective tissue to substitute for injured muscle fibres (Cai et al., 2018). Histological and pathological examinations have shown that WB and WS myopathies are characterized by fibre myodegeneration and regeneration, fibrosis, variability in fibre size and shape, lipidosis, interstitial inflammation, and mononuclear cell infiltration (Kuttappan et al., 2013c; Sihvo et al., 2014; Soglia et al., 2016). The detailed etiology and causes of WB and WS are still unknown. However, a number of investigators have suggested that hypoxia may trigger the development of these myopathies, likely due to a decrease in blood supply to the tissues (Sihvo et al., 2018; Lilburn et al., 2019; Barbut, 2019; Hosotani et al., 2020). Indeed, genetic selection for accelerated growth and large breast muscle has likely led to alterations in muscle fibre size and structural and metabolic traits (Petracci et al., 2015; Velleman and Clark, 2015).

The increase in the size of existing fibres (hypertrophy) is the main mechanism responsible for muscle growth post-hatch, as the increase in the total number of muscle fibres (hyperplasia) is

essentially completed at hatch (Petracci et al., 2015). The increase in fibre size is linked to a decrease in capillarization, which may lead to inadequate oxygen and nutrients supplied to the muscle cells and impaired removal of metabolic products, potentially affecting fibre functionality (MacRae et al., 2006; Branciarri et al., 2009). Previous studies revealed that FG birds have larger fibre diameters, lower capillary density, and higher intercapillary distance compared to unselected birds (MacRae et al., 2006; Velleman and Clark, 2015). The enlarged muscle fibres combined with insufficient vascularization and capillary supply, may result in metabolic stress in FG birds due to the increase in diffusion distance for oxygen, metabolites, and waste products, consequently affecting meat quality (MacRae et al., 2006).

Breast fillets affected by spaghetti meat (**SM**) condition exhibit altered structural integrity leading to the separation of muscle fibres primarily found on the cranial surface of the *Pectoralis major* muscle. This muscle abnormality may be accompanied by the presence of varying degrees of WS (Petracci et al., 2019). The effects of SM myopathy resemble the changes observed with the aforementioned muscle disorders. A study by Baldi et al. (2018) comparing the impacts of WS and SM revealed that these myopathies were linked to several changes in meat quality and histological traits compared to normal fillets. The researchers reported that breast fillets exhibiting SM myopathy were heavier and thicker yet had lower protein and higher moisture content than normal breast fillets. Moreover, SM myopathy was associated with extensive myodegeneration of the *Pectoralis major*, poor fibre uniformity, infiltration of inflammatory cells, lower protein solubility, and extra myofibrillar water in the superficial section of affected fillets, leading to reduced water holding capacity compared to normal fillets. The same authors concluded the effects in meat quality resulting from SM are more pronounced compared to breast muscle solely affected by WS.

The development of SM defect is likely associated with the formation of intracellular spaces in the muscle, probably as a result of alterations in connective tissue within the perimysial compartments and inadequate support to muscle fibre growth (Baldi et al., 2018). In fact, An et al. (2010) reported that FG broiler chickens had a thinner perimysium compared to white leghorns (traditional laying hens).

Recent findings suggest that the aforementioned muscle myopathies may not only influence muscle, metabolic, and meat quality traits, but also the welfare of affected birds. Some birds exhibiting WB are unable to stand up from dorsal recumbency and are reluctant to move (Papah et al., 2017). In addition, WB-affected birds exhibit tissue pathology linked to painful conditions in humans, suggesting the welfare of birds may be compromised (Papah et al., 2017). This agrees with the results of Norring et al. (2019), who reported higher gait scores and fewer movements while lying down in birds with WB lesions compared to non-affected birds. Kawasaki et al. (2016) found that birds exhibiting any degree of WB myopathy were unable to fully lift and close their wings, likely due to a reduction in extensibility of the muscle. Even though behaviour was not assessed in the Kawasaki study, the inability to fully lift the wings could prevent the birds from performing motivated species-specific behaviour, potentially leading to frustration, and further compromising birds' welfare.

As emphasized in this review, the effects of genetic selection for growth rate on walking ability, lameness, and meat quality attributes have been confirmed by a number of investigators. However, the role of factors such as management practices and nutrition have also been widely investigated due to their contribution to the disorders mentioned above.

2.5 Rearing Conditions

Commercial poultry houses shelter thousands of birds under the same environmental conditions that if not strictly controlled, can lead to detrimental effects on live performance, health, and welfare of broiler chickens (Bessei, 2006). Therefore, it is essential to provide optimal conditions in poultry houses by adopting best management practices.

2.5.1 Temperature

Temperature has long been recognized as having an influence on welfare, especially considering the reduced thermoregulatory capacity of modern strains of broiler chickens and reduced heat dissipation, likely due to the extra heat production associated with protein synthesis (Deeb and Cahaner, 2002; Tixier-Boichard, 2020). In commercial poultry houses, due to the high number of birds, accelerated growth rate, and continuous heat production, broiler chickens are susceptible to heat stress as they grow heavier, which can affect their behaviour, physiology, immune status, performance, and welfare (Sandercock et al., 2001; Bessei, 2006).

Although the effects of rearing temperature on the incidence of muscle myopathies have not been thoroughly investigated, heat stress is recognized to have deleterious effects on meat quality. For example, acute heat stress has been associated with undesirable alterations in meat quality traits in broiler chickens, producing pale, soft, and exudative (**PSE**) meat (Barbut, 1997). Acute stress can occur under normal rearing conditions or during transportation and pre-slaughter holding, potentially causing musculoskeletal damage and deleterious effects on muscle membrane integrity and functionality of meat proteins (Sandercock et al., 2001). Moreover, a recent study conducted to investigate the prevalence and risk factors associated with WB, WS, and SM in Canada reported

a higher prevalence of SM during the summer compared to the winter, whereas the higher prevalence of severe scores of WB was observed in the spring in comparison to the fall, indicating seasonal effects on the presence and severity of these myopathies (Che et al., 2020). Therefore, the adoption of efficient ventilation systems and adequate temperature throughout the production cycle, during transportation, and pre-slaughter procedures are essential to mitigate and prevent the negative impacts of heat stress on broiler performance, welfare, and meat quality.

2.5.2 Stocking Density

Similar to temperature, the impacts of stocking density on broiler production have been thoroughly investigated. However, inconsistencies in literature have been found when determining the effects of stocking density on broiler growth performance and welfare. Numerous studies demonstrated that higher stocking densities were associated with decreased locomotion, BW, feed intake, and poorer welfare as a result of higher incidence of skin lesions, leg problems, mortality, elevated ammonia levels, and litter moisture content (Kristensen and Wathes, 2000; Hall, 2001; Meluzzi and Sirri, 2009; Abudabos et al., 2013). However, other studies have suggested little effect of stocking density on broiler performance. For example, when comparing five different target stocking densities (30, 34, 38, 42, and 46 kg/m²), Dawkins et al. (2004), reported no effect of stocking density on mortality, FPD, HB, leg deformities, and ammonia concentration. They concluded that overall environmental and housing conditions have a greater influence on broiler welfare and productivity than stocking density, suggesting that if temperature, ammonia levels, and litter quality are regularly monitored and controlled, the negative impacts of stocking density can be alleviated.

2.5.3 Ventilation

Improved ventilation has been shown to attenuate the effects of high stocking density on the growth rate, suggesting that impaired heat dissipation due to the high number of birds may be a major contributor to poor welfare in elevated stocking densities (Bessei, 2006). These findings are in line with McLean et al. (2002), who reported increased time spent panting in birds kept at 34 and 40kg/m² as compared to birds reared at 28 kg/m², indicating that thermal comfort was decreased as stocking density increased and ventilation was not improved. Indeed, an increase in stocking density has been associated with increased litter temperature, likely as a result of higher nitrogen and moisture levels in the litter, favouring microbial activity and heat production (Bessei, 2006).

The increased amount of feces and moisture in the litter due to elevated stocking densities, combined with poor litter management and inadequate ventilation can lead to elevated levels of ammonia. Ammonia is an irritant gas that has adverse effects in poultry species, including increased susceptibility to respiratory diseases, irritation of mucous membrane in the eyes and respiratory system, increased incidence of contact dermatitis, and poorer productive performance (Kristensen and Wathes, 2000). Therefore, adequate ventilation is crucial to reduce the humidity in poultry houses resulting from birds' respiration and litter moisture, facilitate air circulation and air renewal, maintain ideal temperature range by removing heat excess produced in the facility, and remove the ammonia formed as a by-product of litter fermentation. (De Moura et al., 2010)

2.6 Nutrition

A balanced diet that provides all the required nutrients is essential to promote skeletal development and growth (Waldenstedt, 2006). Therefore, deficiencies or excess of nutrients can lead to bone abnormalities and impaired walking ability in broiler chickens (Williams et al., 2000). Due to the contribution of diet to bone development, the manipulation of growth by altering the nutrient density and restricting feed consumption has also been studied as a strategy to improve bone quality in broiler chickens (Kierończyk et al., 2017).

The use of lower nutrient diets improves leg health by reducing early growth rate, due to the accelerated bone development in this stage. For example, in a study conducted to investigate the effects of diet density on leg health and growth rate, Brickett et al. (2007) reported a reduction in early growth in the group fed the low-nutrient diet (i.e., lower protein content and energy) and a better gait score at 11 and 18 days of age than birds fed a standard diet. However, no difference in gait score was observed at 25 and 32 days of age, probably due to the accelerated growth rate in both treatments, as a positive correlation was found between gait score and BW. These results may indicate a limited effect of the diet in regulating growth and disorders associated with fast growth, likely due to the contribution of genetics on BW (Shim et al., 2012b).

A study by Fanatico et al. (2008) investigated the effects of two dietary nutrient levels (low-nutrient and standard diet) and two genotypes (FG and SG) on performance, livability, and leg health (as measured by gait score, bone mineral density, and incidence of TD). The low-nutrient diet only increased the bone mineral content of SG birds, which also exhibited better gait scores than FG birds, though the low-nutrient diet significantly improved gait scores in both strains.

However, the authors reported that the incidence of TD was only affected by strain, with a higher incidence of this disorder found in FG vs. SG birds. Nevertheless, other nutritional strategies have been implemented to decrease the incidence of TD and other bone disorders in broiler chickens. Besides a decrease in TD observed as a result of genetic selection against this disorder, dietary supplementation of different sources of vitamin D (e.g., 25 hydroxycholecalciferol and 1,25 dihydroxycholecalciferol) has been considered an efficacious strategy to decrease the prevalence and severity of TD in FG strains (Roberson and Edwards, 1994; Rennie and Whitehead, 1996). In addition to reducing the incidence of TD, the supplementation of 25 hydroxycholecalciferol has also been used to prevent lameness caused by BCO (Huff et al., 2000; Wideman, 2016).

Feed restriction has also been successful in modulating growth rates and improving bone quality. When comparing the effects of quantitative (limited vs. *ad libitum* access to feed) and qualitative (control vs. diluted feed) in a 2 x 2 factorial, Nielsen et al. (2003a) reported an increase in activity in the feed-restricted group. However, the authors pointed out that the benefits of increased activity levels and potential improvements in leg health are overshadowed when considering the well-known welfare implications of chronic hunger associated with feed restriction (Nielsen et al., 2003a).

Because muscle myopathies are influenced by the accelerated growth of FG birds, the modulation of growth through diet has been studied as an attempt to decrease these muscle abnormalities. The effects of feed restriction (from 13 to 21 days of age) on the incidence of muscle disorders was investigated by Trocino et al. (2015), who found no effect of feeding regimen on WB. However, feed-restricted birds tended to exhibit a higher incidence of WS compared to the *ad-libitum* group.

The authors concluded that the increase in WS may be attributed to the compensatory growth after the period of restriction, which likely caused a rapid increase in breast muscle growth rate, resulting in fibre damage in the feed-restricted group. In addition to feed restriction, other nutrients have been studied due to their relevance in muscle formation and prevention of other muscle disorders (Cruz et al., 2017; Bodle et al., 2018). However, most studies showed limited effects of dietary interventions and nutrient modulation to reduce the incidence and severity of WB and WS, suggesting that other environmental or genetic factors may be involved with these muscle abnormalities.

2.7 Use of Slower-Growing Birds: Previous Findings and Limitations

With the increase in consumption of poultry meat as well as growing public concerns about the welfare and meat quality of FG birds, there has been considerable interest in the use of SG birds. In 2016, SG birds made up 25 to 30% of broiler production in the Netherlands, while in France and the UK, SG birds represented 15 and 7% of broiler slaughtered, respectively (Thornton, 2017). In addition, several major companies have pledged to adopt the standards established by the Better Chicken Commitment, which specifies the use of some SG strains as one of the strategies to improve the welfare of broiler chickens (Better Chicken Commitment, 2021).

The comparisons between FG and SG birds have been performed in several studies comparing behaviour, leg health, meat quality, and overall welfare, as previously described in this review. In general, remarkable differences between strains differing in growth rate have been reported. In terms of behaviour, SG birds are more active, spend more time walking, and less time sitting compared to FG birds (Bizeray et al., 2000; Bokkers and Koene, 2003; Dixon, 2020). The negative

correlation between growth rate and leg health is well known in broiler chickens (Julian, 1998). Thus, as expected, SG birds are commonly reported to exhibit better walking ability, improved bone health, and lower incidence of leg disorders than FG birds (Shim et al., 2012a; b; Dixon, 2020; Singh et al., 2021). As the muscle myopathies discussed earlier have only recently been investigated, there are not as many studies comparing their incidence in SG and FG strains. However, there is strong evidence that the rapid muscle accretion and large breasts of FG birds are involved in these disorders (Kuttappan et al., 2013a; Petracci et al., 2015; Cai et al., 2018; Barbut, 2019). Thus, the use of SG birds is expected to decrease the incidence and severity of WB, WS, and SM. In fact, a study comparing 3 FG strains and a 1950's strain revealed that WB myopathy was only present in FG birds, which was detected as early as 2 weeks of age and gradually became more severe as the birds aged (Chen et al., 2019). However, mild muscle lesions without any overt clinical signs of WB myopathy were observed in the 1950's strain, suggesting the presence of subclinical muscle disease prior to the 'discovery' of this myopathy. Thus, the use of SG chickens has been suggested to decrease the incidence of muscle myopathies. An additional potential advantage of the use of SG birds is the possibility to mitigate chronic hunger in the parent stock (broiler breeders) of FG birds, as these birds are intensively feed restricted to prevent health problems related to the exacerbated increase in BW, while maintaining good reproductive performance (Decuyper et al., 2006).

The term "fast-growing" chicken commonly refers to birds raised in intensive production systems and exhibiting high productive performance, fast growth rate (> 50 g/d), high breast yield, rapid muscle accretion, and early slaughter age (2.5 kg of BW reached in approximately 40 days) (Doğan et al., 2019; Mancinelli et al., 2020). On the contrary, the term "slow-growing" encompasses a

heterogeneous group of birds selected for reduced growth rate, lower breast yield, and extended time to reach market weight in comparison to FG birds (Fanatico et al., 2008; Doğan et al., 2019; Mancinelli et al., 2020). The growth rate of SG birds varies according to the strains used, with strains often taking 52 to 81 days to reach the market weight compared to 35 to 42 days in FG birds (Dixon, 2020).

Slower-growing birds are commonly raised in alternative rearing systems that may not represent conventional and intensive rearing conditions. In organic rearing systems, providing outdoor access is considered to be crucial to improving animal welfare and the quality of the product (Mancinelli et al., 2020). When comparing the adaptation to organic rearing systems in eight strains differing in growth rate, Castellini et al. (2016) observed better feather condition, lower incidence of skin lesions (i.e., footpad dermatitis and breast blisters), less time spent resting, more exploratory behaviour, and greater activity levels in SG birds vs. FG birds. There was also a negative correlation between adaptation and average daily weight gain, which combined with the differences in welfare indicators mentioned above led the researchers to conclude that despite their better productive performance, FG birds did not adapt well to organic systems, with birds having average daily gain < 50 g/d showing better adaptation to organic systems. These findings are in line with a recent work by Mancinelli et al.(2020), in which birds with faster growth rates showed indicators of poorer welfare and worse adaptation to organic systems compared to strains selected for lower growth rates. Although these results indicate relevant differences between FG and SG birds, intensive rearing conditions, characterized by indoor systems with high stocking densities, as well as controlled temperature and lighting programs (Bessei, 2006), greatly differ from organic

and free-range rearing systems, which only represent a small proportion of broiler production (Mottet and Tempio, 2017).

Dixon (2020) compared FG and SG strains in a research setting that simulated commercial conditions. Overall, like the findings obtained in organic systems, SG birds were more active and had improved welfare as indicated by lower mortality and incidence of skin lesions, as well as better walking ability compared to FG birds, suggesting that SG birds consistently show better welfare-related outcomes in a wide range of environments. In addition, a larger proportion of lower scores (less severe lesions) of WB and WS was found in SG birds, suggesting the deleterious effects of selection for fast growth rate and high breast yield on muscle development and meat quality (Dixon, 2020). However, this study tested one SG strain and three FG strains, making generalizations about the welfare of SG birds difficult due to the large variation in growth rates and individual characteristics of SG strains (Castellini et al., 2016). In addition, Dixon (2020) and other researchers used gait score system to assess walking ability in FG and SG strains. This system, first described by Kestin et al. (1992), assesses birds' walking ability using a 6-point scale, ranging from 0 (no detectable abnormality) to 5 (birds unable to walk). The practicality of this method, which does not require any special tools, equipment, or apparatus, justifies its common use in research and commercial settings to assess walking ability in broiler chickens. However, the subjectivity of this method can make it difficult to compare the walking ability between FG and SG birds, as differences in body conformation and motivation to walk, often reported among strains differing in growth rate, may not necessarily be related to lameness.

In order to overcome the subjectivity of the gait score assessment, other methods have been used to assess leg health in broiler chickens, including the latency-to-lie test developed by Weeks et al. (2002). This test is based on the concept that sound birds will stand for a longer period than lame birds to avoid contact with shallow water, which is considered to be an aversive and novel stimulus. This test was recently used by Singh et al. (2021) to compare differences in latency- to-lie in one FG and one SG strain. In agreement with the results obtained from the gait score assessment by Dixon (2020), SG birds stood longer in water than FG birds, indicating better leg health in the former (Singh et al., 2021). However, this study tested a limited number of strains, which has its limitations as mentioned above.

Another limitation of studies comparing FG and SG birds is the differences in BW between strains. Bird welfare and the incidence of muscle disorders are well known to be negatively affected by an increase in BW as the birds grow (Kestin et al., 2001; Kuttappan et al., 2017; Dixon, 2020). In some studies, FG birds were heavier than SG birds when variables of interest were evaluated (Bizeray et al., 2000; Bokkers and Koene, 2003; Shim et al., 2012b; Lichovniková et al., 2017); therefore, differences in BW rather than potential for growth may have influenced the results. For example, Kestin et al. (2001) found better walking ability in SG birds *vs.* FG birds. However, when BW was considered, differences between strains disappeared. Conversely, other studies reported poorer walking ability in FG birds compared to SG birds even when both strains were evaluated at a similar BW (Corr et al., 2003b; Dixon, 2020; Rayner et al., 2020; Singh et al., 2021), suggesting that genetic growth potential, rather than merely differences in BW may influence the differences in locomotion found among strains.

From the findings described above, one can conclude that previous work evaluating FG and SG strains, in general, pointed towards better welfare-related outcomes in the latter. However, the limitations mentioned indicate that further research is needed to empirically determine the effects of selection for rapid and early growth on the welfare and meat quality of broiler chickens while controlling the potential confounding factors.

2.8 Selection for Growth: Have We Reached Biological Limits?

In the 20th century, the poultry industry focused on the use of quantitative genetics to select for productive traits for accelerated growth (broiler chickens) or high egg production (laying hens) (Siegel et al., 2009). As a result, specialized breeds reached productive performance that far exceeded their ancestor, the red junglefowl (Griffin and Goddard, 1994; Siegel et al., 2009). Molecular studies have suggested that intensive selection could lead to a loss of genetic diversity (reviewed in Tixier-Boichard, 2020). However, despite the intensive selection for production traits, broiler chickens show greater genome variation than layers, likely due to the high number of breeds involved in the selection process of the former (Tixier-Boichard, 2020). These findings suggest that selection for increased growth in broiler chickens can probably continue without compromising their genetic variation. However, considering the aforementioned disorders and their potential welfare implications, one can wonder if selection focused on growth should be continuously carried out without incorporating other traits to mitigate the emerging problems commonly found in broiler chickens.

Due to the large populations used for developing selection criteria in broiler chickens combined with possible interactions between genes and recombination between alleles, a selection plateau

will likely not be reached any time soon in FG birds (Tixier-Boichard, 2020). The biological limits to selection, however, may occur when selection is affecting fitness traits and potentially the survival of individuals, which is mainly caused by unfavourable correlated responses (Tixier-Boichard, 2020). Although the selection plateau for growth will likely not be reached due to lack of genetic variation, this plateau will eventually be reached likely due to biological limitations in response to selection for growth (Tallentire et al., 2018). Therefore, a balanced allocation approach is crucial to maintain good biological functioning (e.g., reproduction, immunity, growth) and improve fitness.

As described in this review, the modification of the environment has been widely used as a strategy to mitigate possible deleterious effects of a desired trait (i.e., rapid growth) on the welfare of broiler chickens (Bessei, 2006). However, these often have limited efficacy to overcome biological limitations resulting from unfavourable correlated responses. Breeding programs have incorporated new traits in the selection criteria, including leg health and oxygen saturation, aiming for a decrease in the occurrence of skeletal and heart disorders associated with fast growth (McKay et al., 2000; Kapell et al., 2012). However, the high occurrence of metabolic disorders reported in broiler chickens suggests that these problems continue to be a concern to the poultry industry. In addition, there is an increasing consumer awareness on welfare (Clark et al., 2016) and meat quality of FG chickens (Petracci et al., 2015). In this context, there is a growing interest in the use of more robust birds as a strategy to mitigate the disorders found in FG birds and meet the growing demand for products that promote better welfare.

Slower-growing birds are generally thought to be more robust, likely due to their longer time for development and balanced allocation of resources among competing traits compared to FG birds. These birds are commonly raised in alternative systems such as free-range and organic production due to their better welfare outcomes and adaptation to diverse rearing conditions (Nielsen et al., 2003b; Mancinelli et al., 2020).

Despite the improved welfare and potentially lower incidence of muscle disorders, the use of SG birds is associated with more resource use and greater environmental impacts. Slaughtering SG birds at 56 days, with a BW of 2.2 kg compared to FG birds slaughtered at 33 days at a similar BW, represents a 27% increase in total feed energy consumption (Tallentire et al., 2018). However, the use of lower-nutrient diets and inclusion of alternative ingredients should be investigated to determine performance differences in FG and SG strains. The cost-efficiency of animal welfare in different broiler production systems was evaluated by Gocsik et al. (2016), who reported better welfare index scores in SG birds raised in alternative systems compared to FG birds raised under conventional rearing conditions. Meanwhile, as expected, raising FG birds in a conventional system was associated with the lowest production cost. These results suggest the complexity of using more robust birds, while simultaneously maintaining the competitiveness and productivity of broiler production. In pigs, the incorporation of robustness traits in breeding strategies resulted in profits similar to those obtained from selection solely focusing on production traits (Knap, 2009). The same author also pointed out the possibility of selecting for robustness and production simultaneously, resulting in animals with high production levels but also improved livability and health. Considering that leg disorders and muscle myopathies have economic and welfare implications, the evaluation of morbidity, mortality, processing losses, carcass condemnations,

meat quality, and a life cycle analysis would be useful to compare the cost benefits associated with the use of SG strains. In this context, sustainable breeding strategies must be adopted to incorporate robustness and production traits, while considering the efficiency and sustainability of the broiler industry.

2.9 Research Objectives

This Ph.D. thesis aims to investigate the impact of growth rate on welfare and meat quality of broiler chickens, focusing on leg health and incidence of muscle disorders in 14 strains (2 FG and 12 SG), differing in genetic potential for BW gain. The strains tested in this study were fed a similar diet and raised under similar conditions to prevent the potential influence of diet and environmental factors on the outcome variables measured throughout the study. The study was conducted under experimental conditions that intended to simulate commercial conditions (e.g., indoor system, high stocking density, controlled temperature, lighting, and ventilation, and similar diet).

It was hypothesized that the lower growth rate of SG strains would be associated with better leg health and meat quality, yet lower carcass yields compared to FG strains. This thesis is part of a multidisciplinary project conducted over eight production cycles, in which behavioural, physiological, and health parameters were collected and reported in other associated studies.

2.10 Specific Objectives

The hypothesis of this thesis was tested in three studies divided into two research objectives focusing on differences in the incidence of muscle disorders and leg health among the strains.

2.10.1 Objective 1

Investigate the effect of growth rate on carcass traits and muscle disorders of broiler chickens.

- *Chapter 4:* to examine the impacts of growth rate on carcass characteristics and yields and incidence and severity of wooden breast and white striping.

2.10.2 Objective 2

Investigate the effect of growth rate on leg health and walking ability of broiler chickens.

- *Chapter 5:* to determine differences in tibial morphology, breaking strength, and mineral content as indicators of bone status, health, and development among strains selected for different growth rates.
- *Chapter 6:* to investigate the effects of growth rate on leg strength and walking ability assessed through objective mobility tests (i.e., latency-to-lie and group obstacle test). Litter quality and the incidence of contact dermatitis (i.e., FPD and HB) were also investigated due to their potential impacts on birds' welfare and walking ability.

Chapter 3: General Methodology

This chapter provides a brief and general methodology of the multidisciplinary project included in this thesis, which investigated productive performance, carcass traits, muscle disorders, meat quality, behaviour, and physiology of FG and SG strains. The complete methodology of the study (including incubation conditions, animal husbandry and housing) is described by Torrey et al. (2021). As mentioned in Chapter 2, this thesis encompassed carcass traits and muscle myopathies, bone characteristics, and leg health of different strains of broiler chickens. For the specific methodology related each study, refer to Chapters 4, 5, and 6.

3.1 Hatching and Husbandry

Animal care and use were reviewed and approved by the University of Guelph's Animal Care Committee (Animal Utilization Protocol 3746) and followed the Canadian Council for Animal Care's Guidelines (CCAC, 2009).

In short, the study encompassed eight trials conducted at the Arkell Poultry Research Station (Guelph, ON, Canada). Each trial represented a typical broiler production cycle, from incubation and hatch to slaughter, with 5 - 7 strains tested per trial. In each trial, fertile eggs from each strain (2 FG and 12 SG) were incubated simultaneously under standardized conditions at the federally inspected facility at Arkell Poultry Research Station. In total, 7,216 birds were reared over eight trials in a single room, containing 28 floor pens (160 cm × 238 cm; width × length) with an expected final stocking density of 30 kg/m².

The room was divided into four blocks to account for micro-climate differences that were detected in pilot studies (Figure 3.1). With the exception of strains G and M, each strain was tested in up to three trials, with four pens per trial representing each block of the room, totaling 12 pens per strain. Due to the low availability of fertile eggs in one trial, strain G was tested in four production cycles, with two pens represented in each of two trials totaling four pens, while the remaining eight pens were equally divided into two trials (4 pens per trial). Strain M was tested in two trials (8 pens, 4 pens per trial) due to the limited availability of fertile eggs.

In each pen, a total of 44 birds were placed. The birds were vent sexed at hatch to maintain sex balance, with 22 males and 22 females per pen. The group weight of each pen was obtained to keep a similar initial BW across the pens of each strain. From each pen, 12 birds (6 males and 6 females), used as focal birds, were individually weighed, wing tagged, and marked with livestock paint for identification purposes. These focal birds were used to assess behavioural, health, meat quality, and physiological parameters described in other studies to be published. All birds received vaccines against infectious bronchitis, coccidiosis, and Marek's disease (Torrey et al., 2021).

In total, 164 groups of birds were reared in 28 pens throughout the study. Each pen contained five nipple drinkers and a hanging round feeder. The pens were enriched with a 30 cm high raised platform attached to a ramp with 25° incline, a hanging round scale (diameter: 50.8 cm), one quarter of a mineral PECKstone (Protekta, Lucknow, Ontario, Canada), and a hanging nylon rope tied to strips of polyester as an oral enrichment (Figure 3.2). Softwood shavings were used as bedding and were removed and replaced at the end of each trial. Birds had *ad libitum* access to a 3-phase (starter, grower, and finisher), all-vegetable, antibiotic-free diet formulated for slow-growth. The feed type (grower, finisher) was switched when strains reached a similar feed intake

compared to FG birds. Light intensity was kept at 20 lux. On the first three days, the lighting schedule was maintained at 23 h of light (L) and 1 h of dark (D) to allow birds to locate feed and water. Thereafter, a 16L:8D photoperiod was used, with one continuous dark period. At placement, room temperature was maintained at 32°C and gradually decreased as the birds aged, reaching 21°C at 5 wk of age.

Both FG and SG were processed at two target weights (TWs) based on their breeder's expected time to reach 2.1 kg (TW 1) and 3.2 kg (TW 2). Due to the differences in growth rate between FG and SG strains, birds were processed at different ages, with half of the pens of each strain being processed at each TW. At TW 1 and TW 2, FG strains were 34 d and 48 d of age, respectively, whereas SG strains were 48 d and 62 d. These processing dates were intended to allow us to evaluate the response variables when the birds had similar BW (2.1 and 3.2 kg) and similar age (48 d).

3.2 Categorization of Strains Based on Growth Rate to TW 2

Strains were categorized into four groups based on their realized growth rate to TW 2 (48 d for FG and 62 d for SG strains, respectively, Table 3.1, previously published by Torrey et al., 2021) in order to facilitate statistical analyses. The 14 strains were categorized as Conventional (CONV; strains B and C; $ADG_{0-48} = 66.0$ to 68.7 g/d), fastest slow-growing (FAST; strains F, G, I, and M; $ADG_{0-62} = 53.5$ to 55.5 g/d), moderate slow-growing (MOD; strains E, H, O, and S; $ADG_{0-62} = 50.2$ to 51.2 g/d) and slowest slow-growing (SLOW; strains D, J, K, and N; $ADG_{0-62} = 43.6$ to 47.7 g/d).

Table 3.1: Average daily gain (ADG)¹ of strains tested throughout trials. Strains are listed by breeder's estimated days to reach 2.1 kg, estimated, and realized ADG (g/d) to Target Weight 1 (approximately 2.1 kg) and Target Weight 2 (approximately 3.2 kg). This data has been published by Torrey et al. (2021).

Strain	Category	Estimated days to reach 2.1 kg	Estimated ADG, g/d	Realized ADG, g		Category
				Target wt 1	Target wt 2	
B	CONV	36	58.33	54.03	68.70	CONV
C	CONV	37	56.76	55.26	66.01	CONV
F	FAST	43	48.84	53.08	55.29	FAST
G	FAST	44	47.73	47.40	53.54	FAST
I	FAST	45	46.67	47.10	54.65	FAST
M	FAST	50	42.00	51.97	55.46	FAST
E	MOD	42	50.00	53.27	50.83	MOD
H	MOD	44	47.73	47.86	51.22	MOD
O	MOD	40	52.50	47.78	50.15	MOD
S	MOD	51	41.18	45.57	50.61	MOD
D	SLOW	50	42.00	42.44	45.56	SLOW
J	SLOW	47	44.68	42.73	47.73	SLOW
K	SLOW	49	42.86	39.31	43.58	SLOW
N	SLOW	50	42.00	39.82	44.06	SLOW

¹ Strains were categorized into CONV, FAST, MOD, and SLOW based on realized Average Daily Gain to Target Weight 2. This categorization facilitated analyses and comparisons among categories, enabling generalizations based on similar growth rates.

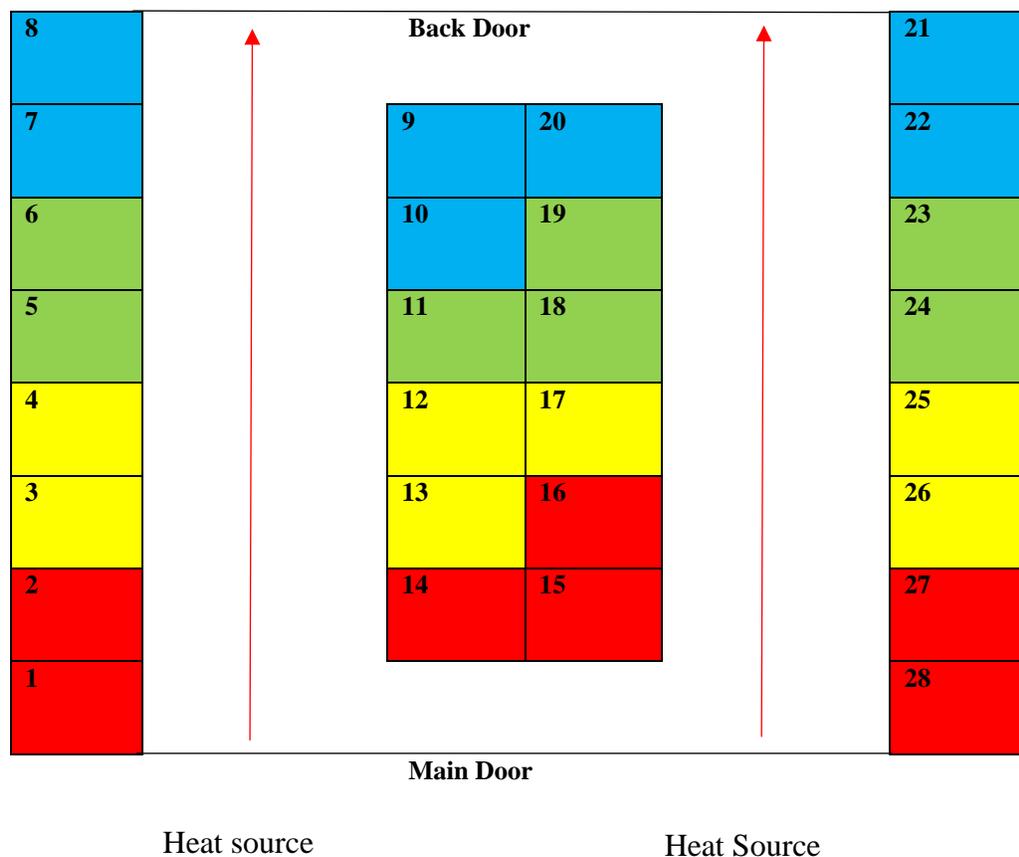


Figure 3.1: Layout of room and block design used throughout the trials. A total of 4 blocks (differentiated by colour) were formed based on temperature gradient from the main door to the back door that influenced pen microclimate temperature. Each pen is approximately 3.83 m². Each block had 7 pens located in different parts of the room (left, and right walls, and in the middle of the room). The following pens were included in each block: block 1: pens 1,2,14,15,16,27,28; block 2: pens 3,4,12,13,17,25,26; block 3: pens 5,6,11,18,19,23,24; block 4: pens 7,8,9,10,20,21,22. In each trial, the strains tested were represented in one pen within each block (4 pens per strain per trial).



Figure 3.2: Pens and enrichments. Each pen was enriched with a ramp with 25° incline to a platform 30 cm above the floor, a hanging scale, PECKStone mineral block (cut into $\frac{1}{4}$ of its original size) and hanging (blue) polyester cloth on one side of the wall. Water lines were located in the back of the pen (not represented in the picture because they were lowered prior to bird placements). Wood shavings were added as a bedding material. The birds were fed with an all-vegetarian, antibiotic-free diet, provided in the round pan feeder placed in each pen. During the first week, brown paper with scattered food was provided to facilitate location of feed and water. On the top of the walls, pens were separated by a wooden frame attached to a net to prevent birds from flying into adjacent pens.

Chapter 4: In Pursuit of a Better Broiler: Carcass Traits and Muscle Myopathies in Conventional and Slower-Growing Strains of Broiler Chickens¹

4.1 Abstract

Selection for accelerated growth rate and high breast yield in broiler chickens have been associated with an increase in myopathies, including wooden breast (WB) and white striping (WS). To investigate effects of growth rate on carcass traits and incidence of myopathies, 14 strains were evaluated, encompassing 2 conventional (CONV; strains B and C: $ADG_{0-48} > 60$ g/d) and 12 slower-growing (SG) strains. The latter were categorized based on growth rate: FAST (strains F, G, I, and M; $ADG_{0-62} = 53-55$ g/d), MOD (strains E, H, O and S; $ADG_{0-62} = 50-51$ g/d), and SLOW (strains D, J, K, and N; $ADG_{0-62} < 50$ g/d). In a randomized incomplete block design, 7,216 mixed-sex birds were equally allocated into 164 pens (44 birds/pen; 30 kg/m²), with each strain represented in 8-12 pens over 2-3 production cycles. From each pen, 4 males and 4 females were processed at 2 Target Weights (TWs) based on their expected time to reach 2.1 kg BW (TW 1: 34 d for CONV; 48 d for SG strains) and 3.2 kg BW (TW 2: 48 d for CONV; 62 d for SG strains). Weights and yields for the carcass, breast, drumsticks, thighs, and wings were obtained; breast fillets were assessed to determine the presence and severity of WB and WS. At both TWs, breast yield was higher as growth rate increased ($P < 0.001$), with CONV having greater breast yield than other categories. Strain F had the greatest breast yield at both TWs ($P < 0.001$) within the FAST category. At TW 2, CONV had the greatest incidence of WB and WS ($P < 0.001$). However, within

¹ A version of this chapter has been published in Poultry Science. Santos, M. N., D. Rothschild, T. M. Widowski, S. Barbut, E. G. Kiarie, I. Mandell, M. T. Guerin, A. M. Edwards, and S. Torrey. 2021. In pursuit of a better broiler: Carcass traits and muscle myopathies in conventional and slower-growing strains of broiler chickens. *Poult. Sci.* In press. <https://doi.org/10.1016/j.psj.2021.101309>

FAST, strain F had the greatest incidence of myopathies ($P < 0.001$) at both TWs, exhibiting values as high or greater than CONV birds. The incidence of WB and WS in strains with differing growth rates but high breast meat yield suggests that the latter may play a major role in the occurrence of these myopathies.

Keywords: chicken meat; meat yield; myopathies; processing traits; slow-growth.

4.2 Introduction

There has been a significant increase in preference for chicken meat globally, with world per capita consumption rising over 250% over the past 50 years (FAO, 2020). Chicken is now the most consumed meat in North America and the second most consumed meat worldwide after pork (FAO, 2020). The increase in demand for chicken meat is mainly due to its high nutrient content, lack of religious or cultural restrictions, affordability, convenience and simple preparation for a variety of individual cut-up portions (Barbut, 2015; Wideman, 2016; Petracci et al., 2019). To meet the growing demand, selection criteria have been adopted to focus on strains with fast, early growth and accelerated muscle accretion (Petracci et al., 2015). These strains selected for rapid growth, commonly referred to as conventional or fast-growing strains, can reach over 2 kg body weight in about 35 days, whereas unselected strains from the 1940's require over 100 days to reach the same target weight (Siegel et al., 2009). This represents an increase of over 400% in the growth rate of broiler chickens in the past 60 years (Zuidhof et al., 2014) with genetic selection being considered the major contributor to this improvement (Havenstein et al., 2003a). Besides differences in growth and body weight, genetic selection has also been associated with changes in body composition and processing traits, with conventional broiler strains having greater carcass and breast yields compared to unselected strains (Havenstein et al., 2003b).

Selection for high breast yield can be attributed to the increased demand for further processed products and cut-up portions rather than the whole carcass and to the increase in preference for breast meat in Western markets (Petracci et al., 2015). Because breast meat is one of the most valuable cuts of the carcass, strategies to improve the quality and appearance of the breast are relevant to primary breeding companies and producers to avoid economic losses associated with condemnations and rejections (Cruz et al., 2017). However, the increase in growth rate and breast yield in fast-growing strains has been accompanied with muscle disorders including wooden breast and white striping, which are two major myopathies reported for conventional strains of broiler chickens (Kuttappan et al., 2012c; Petracci et al., 2019). These disorders have posed a growing concern to producers and retailers due to their high incidence and significant economic impacts to the poultry industry (Kuttappan et al., 2012c; Cai et al., 2018; Petracci et al., 2019).

Wooden breast (**WB**), also referred to as woody breast, was first described by Sihvo et al. (2014) and is characterized by a distinct hardness that can affect different regions of the *Pectoralis major* (Cai et al., 2018). Affected breasts can also exhibit bulging and pale areas that can be associated with the presence of exudate and hemorrhage in severely affected fillets (Barbut, 2019). Wooden breast can be classified as mild, moderate, or severe and different degrees of this myopathy have been reported in many countries where fast-growing broiler chickens are raised (Kuttappan et al., 2012a; Sihvo et al., 2014; Cruz et al., 2017; Ferreira et al., 2020). Recent industry reports have suggested that about 20% of the breast fillets are affected by WB, although a wide range of incidence has been reported in different flocks and studies (as reviewed by Barbut, 2019 and Petracci et al., 2019).

White striping (**WS**) was first described in 2009 and is characterized by white striations parallel to the muscle fibres in breast fillets (Kuttappan et al., 2009). Similar to WB, WS is classified according to the severity of the lesions, which can vary from mild to severe (Kuttappan et al., 2012c). Microscopic analyses suggested that the white striations are a result of fat and connective tissue infiltration in the muscle (Kuttappan et al., 2013a). Recent studies have shown that the incidence of different degrees of WS in modern strains of broiler chickens can surpass 90% (Kuttappan et al., 2017; Petracci et al., 2019; Che et al., 2020).

The effects of WB and WS on meat quality, nutritional value, and technological properties of raw and cooked meat have been well documented (Trocino et al., 2015; Mudalal et al., 2015; Soglia et al., 2016; Dalgaard et al., 2018). In addition, breast fillets with severe WB and WS may require sorting at processing plants, due to the negative impacts of these meat abnormalities on consumer acceptance and meat processing (Kuttappan et al., 2012b; Petracci et al., 2019). The economic losses caused by these meat defects have been estimated to cost over \$1 billion per year in the USA alone (Barbut, 2019).

In addition to their impact on meat quality, muscle myopathies are linked to degeneration and regeneration of muscle fibres, necrosis, hypoxia, and infiltration of inflammatory cells (Petracci et al., 2019; Hosotani et al., 2020). Furthermore, recent findings suggest these muscle disorders may alter bird behaviour and have potential welfare implications for broiler chickens (Kawasaki et al., 2016; Norring et al., 2019). In this context, strategies to mitigate the incidence of these myopathies have been studied. Some of these interventions include modulation of growth (Kuttappan et al., 2012a; Gratta et al., 2019), dietary alterations (Kuttappan et al., 2012b; Cruz et al., 2017), manipulation of incubation temperature and embryonic development (Clark et al., 2017), reduced

age at slaughter (Kuttappan et al., 2017), and reduction of breast yield through genetic selection or nutrition (Bailey et al., 2015; Alnahhas et al., 2016; Cruz et al., 2017; Bailey et al., 2020). Because these strategies have demonstrated limited or no effects on the incidence of WB and WS, the use of slower-growing strains has been suggested as an alternative to alleviate these disorders (Petracci et al. 2019). However, there is little information comparing the incidence of muscle myopathies in slower-growing and conventional broiler chickens raised under similar conditions.

Thus, in this study, we aimed to investigate the differences in carcass traits and the incidence of myopathies among 14 strains of broiler chickens (2 conventional and 12 slower-growing) raised indoors under similar conditions. We hypothesized that slower-growing strains of broiler chickens would have lower meat yields and lower incidence of myopathies compared to conventional strains.

4.3 Materials and Methods

4.3.1 Hatching, Housing, and Rearing

All procedures in this study were reviewed and approved by the University of Guelph's Animal Care Committee (AUP#3746) and followed the Canadian Council for Animal Care's guidelines (CCAC, 2009). This paper is part of a large multidisciplinary study that included the measurement of different variables to assess productivity, behaviour, meat quality, and welfare of conventional and slower-growing strains of broiler chickens. The detailed description of the incubation conditions, animals, housing, and husbandry was reported by (Torrey et al., 2021).

Briefly, a total of eight trials were conducted at the Arkeil Poultry Research Station (Guelph, ON, Canada), with each trial representing a typical production cycle for broiler chickens, from

incubation to slaughter. Fertile eggs from 14 strains (2 conventional and 12 slower-growing) obtained from breeding companies located in North America were incubated and hatched in one federally inspected facility under similar conditions. All the birds were reared in a single room with 28 floor pens (160 cm × 238 cm; width × length) divided into 4 blocks based on location, to account for micro-climate differences at the pen level. The birds from each strain were raised under similar conditions in up to 3 trials, with 4 pens per trial, located in each block of the room, totalizing 12 pens per strain. Due to the availability of fertile eggs and project logistics, strain M was only tested in two trials, totaling eight pens. Details about the methodology and strains tested in each trial are described elsewhere (Torrey et al., 2021). Chicks were vent sexed at hatch, and each strain had equal numbers of males and females, with each pen containing 44 birds (22 males and 22 females). The group weight for each pen was obtained prior to placement and 12 birds (6 males, 6 females) per pen were wing tagged, individually weighed, and painted with livestock paint to differentiate males and females. These birds were used as focal birds for behavioural, physiological, bone quality, and meat quality assessments described in other associated studies. All the birds were vaccinated against Marek's disease, coccidiosis, and infectious bronchitis (Torrey et al., 2021).

Over the eight trials, 164 pens were used. Each pen contained a round feeder (diameter: 33.75 cm) and 5 nipple drinkers. The pens were enriched with the following items: a platform raised 30 cm above the litter attached to a 25° ramp, a mineral PECKstone (Protekta, Inc., Lucknow, Ontario, Canada), a hanging round scale (diameter: 50.8 cm) and hanging nylon ropes containing polyester strips. Soft wood shavings were used as bedding material and replaced in every trial. Pens were separated by solid white plastic walls to prevent visual contact between birds located in different

pens. Lighting schedule was maintained at 23 h of light (L):1h on the dark (D) from day 1 to day 3. From day 4 onward, a photoperiod of 16L:8D was provided, with lights turned on at 06:00 h and turned off at 22:00 h. The light intensity was kept at 20 lux throughout the trials. Room temperature was 32°C at placement and gradually decreased to 21°C at 5 wk of age. The birds had *ad libitum* access to an all-vegetable, antibiotic-free diet, formulated based on nutrient requirements for a slower-growing broiler (Torrey et al., 2021). The diet was provided in 3 phases (starter, grower, and finisher); diet was transitioned when slower-growing strains reached a similar feed intake to conventional birds. Starter, grower, and finisher diet were prepared as fine crumble, coarse crumble, and short pellet, respectively.

Due to the effects of body weight (**BW**) on the variables evaluated in our study, conventional and slower-growing strains were processed at 2 target weights (**TWs**) based on the breeder estimated age to reach 2.1 kg liveweight (**TW 1**) and 3.2 kg liveweight (**TW 2**). At TW 1, conventional and slower-growing strains were 34 and 48 d of age, respectively. At TW 2, conventional and slower-growing strains were 48 and 62 d of age, respectively. On day 34 and 48, the group size for the pens not processed was reduced to maintain a commercial stocking density of 30 kg/m².

4.3.2 Processing Measurements

On the day before processing, eight focal birds (4 females and 4 males) per pen from those to be processed were individually weighed and labelled with a colored zip tie on one of their legs. These labeled birds were part of the 12 wing-tagged focal birds and represented approximately 18% of the initial pen population (prior to removal of birds to maintain stocking density of 30 kg/m²) to encompass a representative sample size, while considering the logistics of the project (e.g., transportation, capacity of the processing plant, and labour). The colored labels enabled the

identification of each pen and differentiation of selected birds from the remaining birds being processed. The group weight was obtained to determine the final BW for each pen processed. The feeders were removed from each pen at 23:00 h the night prior to processing. Birds had free access to water until loading. The next morning at 06:00 h, the birds were placed into crates and transported 35 min to a provincially inspected processing plant. Birds were hung on a shackle line and electrically stunned in a brine-water bath (25V and 120 Hz for 10 s), and then bled for 90 s. Following mechanical defeathering and manual evisceration, the carcasses were air-chilled at 4°C for 5 h and transported in coolers, to the university, where the carcasses were kept overnight at 4°C.

Ready-to-cook carcass weight (after removal of the viscera, feet, and head) was obtained and carcass yield was expressed as the percentage of the live weight (**LW**) obtained the day before processing. Trained butchers manually deboned the carcasses and weights for the skinless and boneless breast muscle (*Pectoralis major* and *Pectoralis minor*), wings, drumsticks, and thighs were obtained. The yields of the carcass portions are reported as the percentage of the ready-to-cook carcass weight and of the LW.

During processing, some of the samples were over scalded. Because the birds were processed in a commercial processing plant, it is unclear why over-scalding occurred more often in some trials and processing than others, as no pattern was observed regarding season, strains, BW, and age of birds showing over-scalded breasts. Color of the skinless breast fillets was measured at 3 different locations (cranial, medial, and caudal) using Minolta CR-400 with Spectra QC-400 software (Folio Instruments, Kitchener, ON) to identify possibly over-scalded samples, following the CIE L*a*b* system, in which L* represents lightness, a* represents redness, and b* represents yellowness.

Samples showing L^* values equal or greater than 59 were removed from the WS assessment (Sirri et al., 2011; da Silva-Buzanello et al., 2019). This value was used as a cut-off point to be conservative in our analysis, resulting in the exclusion of light breast samples potentially due to over-scalding that could interfere the assessment of WS.

4.3.3 Assessment of Wooden Breast and White Striping Myopathies

Following deboning, the presence and severity of WB and WS were assessed. To keep the evaluation consistent, the breast fillets were evaluated by a single trained researcher throughout all the trials. The researcher was trained in palpation for WB and visual assessment of WS (described below) by a field expert from a breeding company. In addition, each breast sample in each trial was evaluated twice at random to ensure consistency in the scoring scheme adopted and good intra-observer reliability. The values were compared using PROC FREQ in SAS[®] version 9.4 (SAS Institute Inc., Cary, NC), with the “agree” option included to calculate weighted kappa statistics. For WB and WS, weighted kappa statistics (K_w) were 0.811 and 0.845, respectively, indicating almost perfect Kappa agreement ($K_w > 0.80$) for both variables. The breast fillets were palpated and scored for WB levels, using a 4-point scale modified from Cruz et al. (2017) and classified as: 0 (normal) - no hardness or paleness detected on the surface of the breast fillet; 1 (moderate light) - hardness detected in up to one third of the breast fillet, present in cranial and/or caudal regions; 2 (moderate severe) - hardness affecting up to two thirds of the breast fillet; and 3 (severe) hardness detected throughout the fillet, commonly associated with presence of hemorrhage and exudate on the surface of the fillet. Similarly, breast fillets were visually examined and scored for WS using the 4-point classification scheme previously described by Kuttappan et al. (2016), in which 0 (normal) represents breast fillets with no visible white lines; 1 (moderate)

represents breast fillets showing thin white lines (< 1 mm thick); 2 (severe) represents breast fillets exhibiting large and noticeable white lines (1-2 mm thick); and 3 (extreme) includes breasts fillets presenting thick white lines (> 2 mm) that commonly merge with other lines and cover a major surface of the breast fillet.

Due to the low proportion of the severe score (3) for WB and WS, scores 2 and 3 were combined and classified as moderate-severe which represents more evident muscle defects that may result in rejection or condemnation. The total incidence of breast fillets exhibiting WB or WS and moderate-severe scores of WB or WS were calculated and reported as a percentage of total breasts examined. Average severity index was obtained separately for WB and WS using the following calculation for each strain: $((n_0 \times 0) + (n_1 \times 1) + (n_2 \times 2) + (n_3 \times 3)) / (n_0 + n_1 + n_2 + n_3)$, where n represents the number of breast fillets in each score. The average mean myopathy score of WB and WS for each strain could range from 0 (all fillets receiving scores of 0) to 3 (all fillets receiving scores of 3). Therefore, greater values are associated with greater severity of myopathies.

4.3.4 Statistical Analyses

To simplify the analysis of differences related to growth rate, the strains were grouped into 4 categories based on their average daily gain (ADG) to TW 2 (48 and 62 d for conventional and slower-growing strains, respectively): Conventional (**CONV**; strains B and C; $ADG_{0-48} = 66.0$ to 68.7 g/d); fastest slower-growing (**FAST**; strains F, G, I, and M; $ADG_{0-62} = 53.5$ to 55.5 g/d); moderate slower-growing (**MOD**; strains E, H, O, and S; $ADG_{0-62} = 50.2$ to 51.2 g/d); and slowest slower-growing (**SLOW**; strains D, J, K, and N; $ADG_{0-62} = 43.6$ to 47.7 g/d).

Data were analyzed as an incomplete block design, with pen as the experimental unit, using Generalized linear mixed models (GLIMMIX) in SAS[®], version 9.4 (SAS Institute Inc.). Category, strain nested within category, sex, target weight and the interactions were included as main effects to compare the variables tested at a similar BW. Trial and block nested within trial were included as random effects. Contrast statements were used to test the differences among categories and strains within categories. Pairwise comparisons were corrected using Tukey adjustment to explore multiple comparisons and differences between categories, strains, and sex. Differences between sex and the interactions of sex with category or strain (within category) are not provided in data tables but are described in the results section with respective P-values if significant. Assumptions of the models were checked by analyzing the residuals to check for normality using quantile-quantile plots and Shapiro-Wilk test. Linearity and homogeneity were assessed using boxplots and studentized residuals. The Gaussian distribution was used for those variables that met all the model assumptions. For carcass traits (absolute and relative values), lognormal distribution was required to meet the model assumptions. The relationships between the incidence of breasts affected by myopathies (WB or WS) and the carcass traits for each strain category were investigated using Spearman's rank correlation coefficients. Correlation coefficients were classified as weak ($r_s < |0.35|$), moderate ($r_s |0.35| \leq r_s < |0.67|$), or strong ($r_s \geq |0.68|$) (Bohrer et al., 2018). For all tests performed, statistical significance was considered at $P < 0.05$.

4.4 Results

The differences in variables measured in this study are described as comparisons between categories (CONV, FAST, MOD, SLOW) and comparisons of strains within categories at both TWs. Significant interactions between category and sex or strain (within category) and sex are

described in the results below for each variable evaluated. As expected, category affected all the variables evaluated. However, some variables differed within category, suggesting distinct criteria in strains selected for similar growth rate. Differences in processing traits between categories are presented in Table 4.1 and differences among strains (within category) are presented in Tables 4.2 - 4.5. The incidence, severity, and average scores of WB and WS are presented in Figures 4.1-4. 6. The incidence of each score (0 to 3) of WB and WS by category is provided in Figure 4.7 (descriptive statistics).

4.4.1 Processing Traits

4.4.1.1 Live Weight

Category affected LW ($P < 0.001$) at both TWs (Table 4.1). However, an interaction between target weight and category ($P < 0.001$; Table 4.1) was found, demonstrating that BW at each target weight depended on category. At TW 1, FAST and MOD categories were heavier than CONV and SLOW, and SLOW was heavier than CONV. At TW 2, SLOW was lighter than the other categories and MOD was lighter than FAST, while CONV was similar to FAST and MOD. Males were heavier than females across all the categories ($P < 0.001$).

Overall, there were no differences in BW between strains within the same category (Tables 4.2, 4.3, and 4.5), except among MOD birds, where strain E was 300 g heavier than strain S (Table 4.4; $P = 0.007$) at TW 1. However, at TW 2, this difference disappeared. The absolute weights for each category in both target weights are presented in Table 4.1 and differences between strains within category are presented in Tables 4.2 - 4.5.

4.4.1.2 Carcass Yield

At both TWs, SLOW had the lowest carcass yield ($P < 0.001$; Table 4.1), with no difference in carcass yield among the other categories. There was no interaction between target weight and category ($P = 0.319$) on carcass yield. Overall, males had greater carcass yields than females ($71.8 \pm 0.15\%$ vs. $71.2 \pm 0.15\%$; $P = 0.005$). No interactions between category and sex ($P = 0.790$) or category and strain ($P = 0.161$) were found. Carcass yields differed ($P < 0.05$) among FAST (Table 4.3) and SLOW (Table 4.5) strains with an interaction between strain within category and target weight present ($P = 0.018$). Within FAST strains (Table 4.3), strain F had greater carcass yields than strains G and M at TW 1, with strain I being similar to the other FAST strains. (Table 4.3). At TW 2, strain F had greater carcass yield than the other FAST strains. Among SLOW strains (Table 4.5), strain D had lower carcass yield than strains J and K at TW 1, whereas at TW 2, carcass yield for strain D was similar to strain K, but lower than strains J and N.

4.4.1.3 Breast Yield

Breast yield was impacted by category, strain, target weight and sex ($P < 0.001$). Among categories, a significantly higher breast yield was observed as the growth rate increased, with $CONV > FAST > MOD > SLOW$, which was consistent at both TWs, despite the differences in age, BW, and carcass weight (Table 4.1). While females had greater breast yields relative to the carcass weight than males ($28.2 \pm 0.11\%$ vs. $27.7 \pm 0.11\%$; $P < 0.001$), a category by sex interaction was present ($P = 0.035$). Sex did not affect breast yield in CONV and MOD strains, but females had greater breast yields than males for FAST and SLOW strains.

Breast yield was affected by strain ($P < 0.001$) in all categories (Tables 4.2 - 4.5). For CONV strains, breast yield was greater in strain C vs. B at both TWs (Table 4.2). For FAST strains, strain

F had the greatest breast yield at both TWs (Table 4.3). However, at TW 1, strain M had the lowest breast yield and strains I and G had similar and intermediate breast yields. At TW 2, this difference disappeared and strain M had similar breast yield to strains G and I. For MOD strains, strain E had lower breast yield (relative to both carcass and LW) than strain O at TW 1, which was also observed at TW 2 for the values relative to LW, while breast yield relative to the carcass weight was similar among the MOD strains (Table 4.4). For SLOW, strain D had the lowest breast yield at both TWs (Table 4.5).

4.4.1.4 Thigh Yield

Similar to breast yield, thigh yield differed among categories, following growth rates ($P < 0.001$; Table 4.1). However, an opposite pattern was observed, with lower thigh yields as growth rate increased. At TW 1, CONV and FAST had lower thigh yield (relative to the carcass weight) than SLOW, while at TW 2, CONV had the lowest thigh yield (relative to carcass weight and LW). Although at TW 1 thigh yield relative to the LW was not significant among the categories, it followed a similar numerical pattern to the values relative to carcass weight. A decrease in thigh yield relative to carcass weight was observed from TW 1 to TW 2 (15.9 ± 0.10 vs. $15.5 \pm 0.09\%$; $P = 0.062$) with no interactions present between target weight and category ($P = 0.513$) or target weight and strain ($P = 0.755$).

Overall, males had greater thigh yield than females (15.8 ± 0.09 vs. $15.5 \pm 0.09\%$; $P = 0.027$) with no interactions between category and sex ($P = 0.748$) or strain and sex ($P = 0.668$). Within categories, only FAST strains differed in thigh yield. Strain F had lower thigh yield relative to carcass weight than strains G and I strains at TW 2, with no strain differences at TW 1 (Table 4.3).

4.4.1.5 Drumstick Yield

Drumstick yield was influenced by category, following a similar pattern as thigh yield, with higher drumstick yield associated with lower growth rate ($P < 0.001$; Table 4.1). At TW 1, CONV strains had drumstick yields relative to carcass weight similar to FAST, yet lower than MOD and SLOW strains. At TW 2, drumstick yield relative to the carcass weight was different among all the categories, with $CONV < FAST < MOD < SLOW$, with a similar pattern observed for the yield relative to the LW, although CONV did not differ from FAST, but was lower than MOD and SLOW ($P < 0.001$; Table 4.1) There was a decrease in drumstick yield relative to carcass yield from TW 1 to TW 2 (13.4 ± 0.05 vs. $13.0 \pm 0.49\%$; $P < 0.001$) with no interactions present between category and target weight ($P = 0.798$) or strain and target weight ($P = 0.911$). For FAST birds (Table 4.3), strain F had the lowest drumstick yield at TW 1, with the same pattern observed at TW 2 for the drumstick yield relative to the LW, whereas drumstick yield relative to carcass weight for strain F was lower than yields for strains G and M, yet similar to strain I. For SLOW birds (Table 4.5), strain D had greater drumstick yield relative to carcass weight than strains J and N at both TWs.

While males had greater drumstick yields than females ($13.6 \pm 0.08\%$ vs. $12.9 \pm 0.04\%$; $P < 0.001$), a strain by sex interaction ($P = 0.004$) was present. For CONV birds, strain B males had greater drumstick yields relative to carcass weight than strain B females, with no sex differences for drumstick yields in strain C. For FAST strains, F females had lower drumstick yield compared to other FAST females, while among FAST males, strain did not affect drumstick yield. For MOD category strains, males had greater drumstick yields relative to carcass weight than females for strains E, H and S with no sex differences in drumstick yield for strain O birds. Drumstick yields

(relative to carcass weight) for SLOW strains females did not differ, while strain D males had greater drumstick yields than strains J and N males (data not shown).

4.4.1.6 Wing Yield

Category affected wing yield at both TWs ($P < 0.001$; Table 4.1). At TW 1, CONV and FAST had similar wing yields (relative to the carcass weight) that were lower than MOD and SLOW. A similar pattern was observed for wing yield relative to the LW, but CONV had the lowest yield, while MOD and SLOW had the highest wing yield, with FAST being intermediate. At TW 2, a significant higher wing yield was observed as the growth rate decreased (CONV < FAST < MOD < SLOW). Wing yield decreased as birds aged (TW 1 = $10.8 \pm 0.05\%$, TW 2 = $10.1 \pm 0.04\%$; $P < 0.001$) with no interactions present between target weight and category ($P = 0.599$) or target weight and strain ($P = 0.128$). Sex did not affect wing yield ($P = 0.370$) with no category by sex interaction present ($P = 0.290$).

Wing yield was affected by strain within categories ($P < 0.001$) at both TWs. For FAST birds (Table 4.3), strain F had lower wing yield (relative to carcass weight) than strain M at TW 1, while strains G and I were similar and did not differ from strains F and M, but this difference disappeared at TW 2 (Table 4.3). For SLOW birds (Table 4.5), no differences in wing yield were observed at TW 1, but at TW 2, strain J had lower wing yield (relative to the carcass weight) than strains D and K. A strain by sex interaction was present ($P < 0.040$) involving FAST and SLOW strains. For FAST birds, strain differences in wing yield (relative to the carcass weight) were found for females, but not for males. For SLOW birds, strain J males had greater wing yield (relative to carcass weight) than strain J females while for the remaining SLOW strains, females and males had similar wing yields (data not shown).

4.4.2 Incidence and Severity of Myopathies

4.4.2.1 Total Incidence of WB and WS

The incidence of WB differed by category, target weight, strain, and sex ($P < 0.001$). Overall, the incidence of WB increased from TW 1 to TW 2 ($28.2 \pm 1.81\%$ of breast fillets scored *vs.* $40.5 \pm 1.86\%$ of breast fillets scored; $P < 0.001$) and males had greater incidence of WB than females ($41.9 \pm 1.77\%$ of breast fillets scored *vs.* $26.8 \pm 1.76\%$ of breast fillets scored, $P < 0.001$). However, including BW as a covariate eliminated differences in WB incidence between TW ($P = 0.249$) and between sexes ($P = 0.155$).

A category by target weight interaction was found ($P = 0.048$; Figure 4.1). At both TWs, CONV strains had the greatest incidence of WB. However, at TW 1, WB incidence for FAST exceeded MOD and SLOW, with no differences in WB incidence between FAST and MOD strains at TW 2.

For CONV birds, strain C had a greater incidence of WB than strain B at TW 1, but this difference disappeared at TW 2 (Figure 4.2). For FAST birds, strain F had the greatest incidence of WB at both TWs (Figure 4.2). When using BW as a covariate, CONV birds still had the greatest incidence of WB compared to other categories and strain F had the greatest incidence of WB among FAST strains (data not shown).

Similar to WB incidence, category, target weight, strain, and sex affected the total incidence of WS ($P < 0.001$). When BW was included as a covariate, the effect of sex disappeared ($P = 0.410$). While incidence of WS increased as birds grew from TW 1 to TW 2 ($16.6 \pm 2.04\%$ of breast fillets scored *vs.* $31.4 \pm 2.27\%$ of breast fillets scored, $P < 0.001$), a target weight by category interaction

was observed ($P = 0.018$; Figure 4.1). Breasts from CONV and FAST had similar incidence of WS at TW 1, which was greater than MOD and SLOW. However, at TW 2, CONV had the highest and SLOW had lowest incidence of WS, while MOD and FAST had intermediate values. When differences in BW were considered, the effect of target weight on WS disappeared. In contrast, category ($P = 0.036$) and strain ($P < 0.001$) still influenced the incidence of WS, with CONV having the greatest percentage of WS across categories. Strains C and F still had more breasts exhibiting WS among CONV and FAST strains, respectively, while there were no differences among the strains in other categories on WS incidence (data not shown). Strain F had the greatest incidence of WS among FAST strains at both TWs (Figure 4.2). WS incidence did not differ among CONV, MOD strains or SLOW strains.

4.4.2.2 Moderate-Severe WB and WS

While breast fillets affected by moderate to severe scores differed by category ($P < 0.001$; Figure 4.3) at both TWs, incidence increased from TW 1 to TW 2 ($15.1 \pm 1.37\%$ vs. $25.1 \pm 1.41\%$ of breast fillets scored; $P < 0.001$). However, there was a TW by category interaction ($P < 0.001$) for moderate-severe WB due to a similar incidence in CONV and FAST at TW 1, while at TW 2 CONV had twice the incidence of moderate-severe WB than FAST.

Males had a greater incidence of moderate-severe WB than females ($25.9 \pm 1.36\%$ vs. $14.4 \pm 1.35\%$ of breast fillets scored; $P < 0.001$), but sex interacted with category ($P = 0.008$). Males had a greater incidence of moderate-severe WB than females in all the categories, except among SLOW birds, which were not affected by sex (data not shown). Incidence of moderate-severe WB was greatest in strain C for CONV birds and strain F for FAST birds (Figure 4.4). Incidence of moderate-severe WB was similar among strains for MOD birds and SLOW birds.

Incidence of moderate-severe WS increased from TW 1 to TW 2 (6.1 ± 1.27 vs. $12.2 \pm 1.32\%$ of breast fillets scored; $P < 0.002$). A TW by category interaction ($P = 0.005$, Figure 4.3) in moderate-severe WS was due to similar incidence of moderate-severe scores for CONV and FAST at TW 1, while incidence of moderate-severe scores for WS was greater in CONV vs. FAST at TW 2. Overall, males had greater incidence of moderate-severe WS than females (males: $11.2 \pm 1.13\%$ of breast fillets scored; females: $7.1 \pm 1.11\%$ of breast fillets scored; $P = 0.001$). Strain did not affect the incidence of moderate-severe WS in CONV strains at TW 1, but strain C had more moderate-severe WS than strain B at TW 2 (Figure 4.4). Strain F had a greater percentage of moderate-severe WS compared to other FAST strains at both TWs.

4.4.2.3 Average Scores for WB and WS

The average WB score increased from TW 1 to TW 2 (0.50 ± 0.037 vs. 0.79 ± 0.039 ; $P < 0.001$). A TW by category interaction ($P = 0.001$; Figure 4.5) was due to similar average WB scores for CONV and FAST at TW 1, while average WB score was greater in CONV vs. FAST at TW 2.

Males had a greater average score for WB than females (0.81 ± 0.038 vs. 0.48 ± 0.04 ; $P < 0.001$). However, there was an interaction between sex and category ($P < 0.001$), where average WB score was similar among females for MOD and SLOW, whereas among males MOD was greater than SLOW.

Strain C had a greater average WB score than strain B for CONV birds at both TWs (Figure 4.6). Strain F had the greatest average WB score compared to other FAST strains at both TWs (Figure 4.6). For SLOW birds, strain N had greater average WB score than strain D (Figure 4.6).

Average WS score increased from TW 1 to TW 2 (TW 1: 0.25 ± 0.030 , TW 2: 0.50 ± 0.034 ; $P < 0.001$). A TW by category interaction ($P = 0.001$; Figure 4.5) was due to similar average WS scores between CONV and FAST, which exceeded similar values between MOD and SLOW at TW 1 while at TW 2, average WS score was higher as the growth rate increased, with $CONV > FAST > MOD > SLOW$. Males had greater average WS scores than females (0.48 ± 0.030 vs. 0.27 ± 0.030 ; $P < 0.001$). A sex by category ($P = 0.004$) interaction was due to similar average WS scores between CONV and FAST females which exceeded similar values between MOD and SLOW females, while for males, average WS scores were significantly higher as the growth rate increased, with $CONV > FAST > MOD > SLOW$ (data not shown).

Strain did not affect average WS score for CONV birds at TW 1, yet strain C had greater average WS scores than strain B at TW 2 (Figure 4.6). Strain F had a greater percentage of severe WS compared to other FAST strains at both TWs.

4.4.3 Correlation among Myopathies and Carcass Traits

The relationships between the incidence of myopathies and processing traits (relative to the carcass weight) are shown in Table 4.6. Live weight was weakly to moderately positively correlated ($P < 0.010$) with the incidence of WB and WS for all categories of strains. Similarly, for all categories, there was a positive weak to moderate correlation ($P < 0.030$) between carcass yield and incidence of WB, whereas carcass yield was weakly to strongly positively correlated ($P < 0.028$) with WS, depending on the category. Breast yield was also weakly to moderately positively correlated ($P < 0.001$) with the incidence of WB and WS across all categories. In contrast, there was a weak negative ($P < 0.020$) correlation between thigh yield and the incidence of WB for the FAST category ($P = 0.007$), while no significant correlations were found in the other categories.

Drumstick yield was weakly and negatively correlated ($P = 0.012$) with the incidence of WB in the FAST group, whereas a negative weak correlation ($P < 0.003$) was found between drumstick yield and the incidence of WS in the CONV and FAST categories. Among FAST, MOD, and SLOW categories, wing yield and the incidence of WB were weakly to moderately negatively correlated ($P < 0.038$), whereas there was a negative weak to moderate correlation ($P < 0.007$) between wing yield and WS incidence for FAST and MOD groups.

4.5 Discussion

Selection for improved growth performance in fast-growing strains of broiler chickens has been associated with changes in body composition and development of muscle disorders including WB and WS (Petracci et al., 2015; Barbut, 2019). Therefore, the use of slower-growing strain has been suggested as an alternative to decrease these myopathies (Petracci et al., 2019). However, there is little information on the incidence of myopathies in slower-growing strains with different growth rates and carcass traits than fast-growing strains. Therefore, this study investigated the differences in processing traits (weight and yield) and the incidence and severity of WB and WS in 14 strains of broiler chickens representing a wide range of growth rates. These strains were divided into 4 categories (CONV, FAST, MOD, SLOW) based on the similarity of growth rates.

Slower-growing strains were hypothesized to have different carcass yields and composition along with fewer myopathies compared to fast-growing broiler chickens. Our findings demonstrate differences among categories for all carcass traits and the incidence and severity of muscle abnormalities. However, we also found differences within categories, indicating remarkable differences among strains, despite their similar growth rate.

4.5.1 Processing Traits - Absolute Weight and Percentage Yield

The differences in BW among the categories were reflected in the weight differences for carcass and cuts (absolute weights for breast, thigh, drumstick, and wing). Therefore, only the differences in yield are discussed below. If not specified, differences described for yields are applicable for values relative to both the carcass and LW.

4.5.1.1 Effect of Category

Previous studies have found that increased BW is associated with increased incidence and severity of myopathies (Kuttappan et al., 2012a; Alnahhas et al., 2016). Therefore, the present study slaughtered birds at two time points (TW 1 and TW 2) based on their expected growth to allow comparisons at a similar BW for both conventional and slow growing strains. While birds were expected to reach approximately 2.1 and 3.2 kg BW, at TW 1 and TW 2 respectively, there were strain differences in BW at both TWs due to differences in ADG (43.6 to 68.7 g/d).

At TW 1, MOD and FAST were heavier than CONV and SLOW strains, with SLOW being heavier than CONV. The lighter BW for SLOW strains was expected based on classification due to reduced growth rate ($ADG < 50\text{g/d}$). However, CONV strains were lighter than the expected target weight of 2.1 kg due to project logistics which necessitated an early processing date (~ 2 days before reaching 2.1 kg BW). The lower than expected LW at slaughter age for CONV birds may have been due to feeding an identical diet to all the birds in the study, which was formulated for slower growth. A pilot study (Santos et al., 2018) found this same diet produced lower BW vs. birds fed a standard, conventional diet. However, at TW 2, CONV had a similar BW compared to MOD and FAST strains, while SLOW strains were still lighter.

The lower BW for CONV and SLOW strains at TW 1 resulted in lighter carcass, breast, thigh, drumstick and wing weights compared to FAST and MOD strains. While BW and carcass weights for CONV birds did not differ from those of MOD and FAST strains at TW 2, CONV birds had greater breast weights compared to other categories. Regardless, at both TWs, higher growth rate resulted in greater breast yield and lower thigh, drumstick, and wing yields. These differences among categories of strains suggest that genetic selection influences not only growth rate but also carcass parts and processing yields. This is in agreement with Fanatico et al. (2008) and Singh et al. (2021), in which a fast-growing strain of broiler chicken had greater breast yield, yet lower wing and leg yields compared to a slower-growing strain. Selection for greater breast yield in fast-growing strains of broiler chickens is mainly due to the preference for and greater cost of this portion of the carcass in Western countries (Barbut, 2019).

We expected to find differences among strains with different growth rates in breast and carcass yields. Havenstein and colleagues (2003b) found greater breast and carcass yields in a modern (from 2001) strain of broiler chickens compared to an unselected (from 1957) strain. Despite differences in growth rates and carcass composition among categories in the present study, CONV strains had similar carcass yield compared to FAST and MOD, whereas SLOW had the lowest carcass yield at both TWs. These results indicate that selection for carcass yield has been successful in slower-growing strains with ADG > 50 g/d, despite their reduced efficiency and increased time to reach the market weight compared to CONV birds (Torrey et al., 2021). Greater carcass yields for conventional and some slower-growing strains can be attributed to a shift from consumption of the whole carcass to cut-up portions, with selection for maximum yield for main carcass parts

and edible components (e.g., leg and breast) and minimal offal yield (e.g., head, neck, viscera and giblets) (Brake et al., 1993; Petracci et al., 2015).

4.5.1.2 Effect of Strain

While strains were categorized based on their ADG to aid analyses, we found differences among strains within each category in carcass traits and composition. Some of these differences persisted as the birds aged, however others were age-dependent and were not consistent as the birds grew. Changes in carcass yield and composition with age have been reported in both conventional and slower-growing strains (Brake et al., 1993; Havenstein et al., 1994a; Young et al., 2001). The differences found within categories suggest that despite similar growth and efficiency, strains from the same category may have undergone distinct selection criteria that resulted in differences in carcass traits and yield. Differences in yield from broiler chickens selected for similar growth performance have been observed in other studies. López et al. (2011) reported similar LW at d 42, yet differences in breast and carcass yield in two strains selected for distinct emphasis on yield maximization. Mehaffey et al. (2006) also found differences in breast yield among five commercial genotypes of broiler chickens despite their similar LW at 7 wk.

4.5.1.3 Effect of Sex

As expected, sex affected weights of all carcass components evaluated, with males being heavier than females irrespective of target weight. The effect of sex on BW and processing traits have been well-documented and is mainly attributed to differences in growth performance between male and female broiler chickens (Shim et al., 2012c). Similarly, sex affected the yield of carcass components, with females having lower yields for carcass, drumsticks, and thighs, yet greater yields for breasts than males. However, breast yield was not affected by sex for CONV and MOD

categories. The greater breast yield of females compared to males has been reported in previous studies in both fast and slower-growing strains (Havenstein et al., 2003b; López et al., 2011). In contrast, Hussein et al. (2019) reported similar breast yields in females and males for a fast-growing strain of broiler chicken, which corroborates our findings, suggesting uniformity and selection for maximization of breast yield in both sexes. In general, the effects of sex on carcass yield and composition in the present study are in agreement with results in past studies evaluating different strains of broiler chickens (Brake et al., 1993; Young et al., 2001; López et al., 2011; Shim et al., 2012c).

4.5.2 Incidence and Severity of Myopathies

4.5.2.1 Effect of Category

Similar to carcass traits, myopathies were influenced by category, with CONV birds having greater incidence, average scores, and severity for WB and WS than the remaining categories, especially at TW 2. These results are in agreement with those reported by Dixon (2020), who found a greater incidence of WB and WS in fast-growing strains compared to slower-growing birds. Even though the presence of these myopathies has not been thoroughly investigated in slower-growing birds, the association between accelerated growth rate and greater incidence of muscle abnormalities has been demonstrated in fast-growing strains of broiler chickens (Kuttappan et al., 2012a; Lorenzi et al., 2014). These results suggest that muscle from fast-growing birds may not be accompanied by adequate supporting cells and tissues, leading to muscle damage and myopathies (Wilson et al., 1990b; Kuttappan et al., 2013a).

Although the underpinning causes for development of WB and WS have yet to be elucidated, previous studies suggest that the development of breast muscle in modern strains of broiler

chickens may be associated with insufficient supply of blood and oxygen to the muscle (Sihvo et al., 2018; Petracci et al., 2019). Since muscle fibre formation in chicks is complete at hatch, post-hatch muscle growth is attributed to the enlargement of myofibers (Clark and Velleman, 2017). Fast-growing strains of broiler chickens have greater myofiber diameters in the *Pectoralis major* compared to laying hens (MacRae et al., 2006). Selection for increased breast size and yield has been associated with increase in breast thickness in the cranial region of the breast, which is speculated to have larger myofibers than the caudal region (Clark and Velleman, 2017). This enlargement of muscle fibres post-hatch has been linked to poor circulation due to the reduced space available for capillaries and blood supply, leading to tissue hypoxia and reduced transportation of nutrients to the muscle (Sihvo et al., 2018; Petracci et al., 2019). These effects increase the potential for muscle fibre damage and metabolic stress (MacRae et al., 2006; Clark and Velleman, 2017; Sihvo et al., 2018). In fact, breast muscle tissues from WB-affected birds, which also commonly exhibit WS, often have increased levels of biomarkers associated with oxidative stress and muscle degeneration (Abasht et al., 2016). In our study, we found that CONV birds had the greatest plasma concentrations of aspartate transaminase (AST), lactate dehydrogenase (LDH), and creatine kinase (CK) compared to the other categories (unpublished data). However, strain F (FAST) had the greatest level of these enzymes among FAST birds, exhibiting values as high as CONV birds, with AST, LDH, and CK being 1.75 to 1.85, 2 to 2.5 and 3.9 to 5.8-fold greater than the remaining FAST birds, respectively (unpublished data). CONV strains and strain F also had the greatest breast yield and incidence of myopathies among all strains. While we have not yet analyzed the correlations between these enzymes and carcass traits, these findings are in line with Kuttappan et al. (2013b) who found differences in serum biochemical

profile between normal and severe WS- affected chickens, with the latter exhibiting greater levels of AST, LDH, and CK. In agreement with these findings, a study conducted as part of our large project compared the plasma attributes of one conventional (B) and four slower-growing strains (D, E, H, and M) at 48 d. The authors reported greater plasma concentrations of AST, LDH, and CK in strain B compared to the remaining slower-growing strains tested (Mohammadigheisar et al., 2020). Elevated concentrations of these enzymes could be due to muscle damage, causing disruption of the sarcolemma and leakage of such enzymes into the plasma or serum (Kuttappan et al., 2013b).

Besides the impacts of WB and WS on meat quality, consumer acceptance, and biochemical profile, recent studies have suggested these myopathies may also compromise the behaviour, health, and welfare of broiler chickens. In fact, a study conducted to investigate the aetiology of WB disorder found that some birds showing this abnormality were unable to right themselves when accidentally falling on their back (dorsal recumbency) (Papah et al., 2017). In addition, birds affected by WB appeared reluctant to general movement. The authors emphasized that WB-affected birds exhibited tissue pathology that has been associated with painful conditions in humans. Even though pain assessment was not conducted in that study, the researchers suggested that WB disorder may have welfare implications in broiler chickens (Papah et al., 2017). In another study, Gall et al.(2019) reported that WB was not only associated with dorsal recumbency, but also with pulmonary disease and mortality. In fact, 71% of the late mortality (last 16 d of a rearing period of 56 d) were heavy males, with evaluation of gross lesions revealing that 68% of the dead birds (culls and found dead) exhibited WB and concurrent pulmonary disease. The authors concluded that besides the well-known adverse effects of WB on meat quality, this disorder also

brings welfare concerns due to the inability of some of the affected birds to stand after falling onto their backs, which can lead to respiratory distress and even death if the birds are not righted quickly. Similarly, Norring et al. (2019) reported behavioural differences, with less movements while lying down and poorer gait scores at different ages in WB-affected birds as compared to non-affected counterparts, indicating impaired walking ability for the former group. The authors concluded that the occurrence of lameness observed in broiler chickens may be partially associated with the incidence of WB. However, the heavier BW and greater breast yield for WB-affected birds may have contributed to both the occurrence of the WB myopathy and poorer gait score (Norrning et al., 2019). Even though the walking ability was not investigated by De Almeida Mallmann et al. (2019), they observed a reduced femur diameter, and calcium, and phosphorus percentages, yet greater breast weight and fillet thickness in WB-affected birds compared to unaffected birds, suggesting a possible relationship between WB disorder and bone mineralization. In addition, elevated levels of metabolites that are often associated with inflammation, tissue injury, and pain have been found in breast muscle tissue samples from WB-affected birds (Abasht et al., 2016).

A recent study suggests similarities between type 2 diabetes and WB, indicating a possible dysregulation of lipid and glucose metabolism in WB cases (Lake and Abasht, 2020). Wooden breast-affected birds have also shown signs of gas disturbance in a study by Lake et al.(2020), who reported higher partial pressure of CO₂ and total CO₂, yet lower pH, partial pressure of O₂, and O₂ saturation in blood samples of males exhibiting WB compared to unaffected birds. The authors suggested that WB-affected birds may have respiratory acidosis as indicated by the low pH and high partial pressure of CO₂. Respiratory acidosis may be attributed to the increase in CO₂

production, insufficient respiratory gas exchange (cardiopulmonary insufficiency), or both, suggesting an elevated metabolic rate that is not properly supported by adequate systems (e.g., cardiovascular and respiratory systems). Other researchers reported that birds with any degree of WB myopathy were incapable of fully lifting their wings, which has been suggested as a clinical symptom and potential method for detection of WB in live birds (Kawasaki et al., 2016). Although not yet investigated, the inability to fully lift the wings, possibly because of reduced extensibility of the degenerated muscle as suggested by Kawasaki et al. (2016), could potentially prevent or reduce behaviours the birds may be motivated to perform, such as wing flapping. The inability of some WB affected birds to right themselves from dorsal recumbency has been speculated to be attributed to the damage of the *Pectoralis major* muscle, preventing the effective use of wing-assisted movements needed to turn the birds back onto their legs (Gall et al., 2019). All together, these findings suggest that the modern myopathies commonly reported in fast-growing strains of broiler chickens may have significant welfare implications and should be further investigated.

At the heavier processing weight, the incidence of WS (53 to 80%) found in the present study for CONV strains was similar to those reported by Dixon (2020) for birds at 42 d (63 to 78%) but lower than other recent studies that reported almost all (> 90%) breast fillets from fast-growing strains exhibited some degree of WS (Kuttappan et al., 2017; Che et al., 2020). The percentages of WB-affected breasts in CONV birds in the present study at TW 1 (36 to 70%) and TW 2 (70 to 84%) were greater than values found in some previous studies (Trocino et al., 2015; Gratta et al., 2019; Dixon, 2020), but similar to those reported elsewhere (Cruz et al., 2017; Kuttappan et al., 2017; Bodle et al., 2018). The discrepancy in the incidence of myopathies may be due to subjective evaluation (palpation and visual) of myopathies. In addition, genetics along with non-genetic

factors (e.g., diet, incubation, age, growth rate) influence carcass yield, composition, and incidence of WB and WS in broiler chickens (Kuttappan et al., 2012b; Kuttappan et al., 2013a; Clark et al., 2017; Cruz et al., 2017; Kuttappan et al., 2017). The birds used in the present study were incubated and reared under similar environmental conditions, with a standardized stocking density and no outdoor access. A controlled and similar environment allowed us to investigate differences among strains under standardized housing and management. However, different strains may have different responses to the conditions provided. Therefore, the incubation and environmental conditions in the present study may have affected the strains differently, with some strains performing better than others as a result. In fact, ideal incubation conditions may vary according to many factors, including the breeder genetics and age, yolk size and egg composition, eggshell properties, length, and temperature of storage (reviewed in Oviedo-Rondón et al., 2020). There is evidence that modified or suboptimal incubation conditions may affect the incidence of myopathies in poultry (Oviedo-Rondón et al., 2020). A recent study by Nyuiadzi et al. (2020) demonstrated that chickens from eggs exposed to a short, cold temperature (15°C for 30 minutes) during the last stages of incubation, had slightly lower incidence of WS compared to the control-incubated group when birds were reared in optimal (control) temperatures. However, when exposed to early cold rearing temperatures, the incidence of WS in those males from the cold-incubated group was higher than males from the control-rearing group, suggesting a possible interaction between incubation and post-hatch conditions on the incidence of myopathies. In addition, all birds were fed the same three-phase diet that may not be suitable to optimize the growth and productive performance for conventional birds, but enabled the comparisons under standardized conditions.

The influence of diet on the incidence of myopathies has been reported in earlier studies (Cruz et al., 2017; Sachs et al., 2019; Livingston et al., 2019). Although most of the aforementioned studies only tested fast-growing strains, the standard diet used in our study may have affected slow and fast-growing strains differently based on their specific nutrient requirements. Furthermore, the diet formulated for slower-growth in our study resulted in a reduction in BW in CONV birds, but only at the end of the production cycle (42 d) as demonstrated in a study previously conducted by our research team (Santos et al., 2018). Due to the positive correlation between BW and the incidence of myopathies (Kuttappan et al., 2013b; Alnahhas et al., 2016; Petracci et al., 2019), an effect of diet would be expected as a result of the reduced BW in CONV birds. However, CONV birds reached the expected BW of 3.2 kg at TW 2, suggesting that the impacts of diet on BW might have occurred in earlier stages of growth. Interestingly, Torrey et al. (2021) reported some differences in ADG to TW 1 in some strains compared to the breeder's expected growth rate, suggesting that strains may have responded differently to the diets and rearing conditions, which may have influenced the results observed in the present study. Therefore, considering the heterogeneous group of birds evaluated, the conditions the birds were incubated, reared, and fed may have affected the incidence and severity of myopathies observed among strains. However, considering the number of strains investigated in this study, optimizing diet, incubation, and rearing conditions for each strain was not feasible in our research facility.

Despite the contribution of growth rate to the incidence of muscle disorders, an increase in breast yield was also associated with greater incidences of myopathies. In contrast, as drumstick, thigh and wing yield increased, the presence of WB and WS decreased. Correlation analysis supported these findings, with breast yield positively correlated and the percentage yield of drumstick, thigh

and wing negatively correlated with the myopathies in some categories of strains. These findings are in agreement with many studies that found increased growth rate and greater breast meat yield were critical factors in the development of muscle disorders in modern strains of broiler chickens (Kuttappan et al., 2012a; Kuttappan et al., 2013a; Petracci et al., 2013; Lorenzi et al., 2014; Mudalal et al., 2015; Alnahhas et al., 2016; Kuttappan et al., 2017). However, the possibility of increasing breast muscle yield while reducing the genetic propensity to develop myopathies, has been reported in fast-growing birds as a result of a balanced breeding program (Bailey et al., 2015, 2020).

Lake et al. (2020) recently reported greater *Pectoralis major* in WB-affected birds, confirming the association between high breast yield and the development of myopathies, possibly due to a reduction in capillary density and overstretching of myofibers (Kuttappan et al., 2013c; Dalle Zotte et al., 2017). However, because WB lesions can be detected as early as 1 wk of age (Papah et al., 2017), the hypothesis that WB is caused by overstretching of myofibers may not accurately and fully explain the onset of the disorder (Lake et al., 2020). Despite the potential contribution of breast yield on the incidence of WB and WS, other alternative explanations for this relationship should not be discarded. As an example, recent studies suggest that breast muscle hypertrophy may be a symptom rather than the cause of myopathies, similar to the hypertrophy of organs (e.g., kidneys, heart, liver) observed in some diabetic complications in mammals (Lake et al., 2019; Lake and Abasht, 2020).

Besides the presence of WB and WS, the incidence of spaghetti meat, characterized by the loss of integrity and separation of muscle fibre bundles, has also been observed in broiler chickens, affecting up to 20% of the birds at processing age (Petracci et al., 2021). Although not investigated

in this present study, the incidence of this disorder should be determined in future studies comparing slow and fast-growing strains of broiler chickens due to the negative effects of this myopathy on meat quality. Furthermore, due to the effects of WB and WS on meat quality, nutritional profile, histological traits, and gene expression (Velleman and Clark, 2015; Mudalal et al., 2015; Dalle Zotte et al., 2017), other studies should investigate these variables in strains selected for distinct growth rates to provide a better understanding of the changes associated with muscle disorders.

4.5.2.2 Effect of Strain

We found differences among strains within the same category, despite their similar production performance and LW at both processing ages. Strain C, within the CONV category and strain F, within the FAST category had a greater incidence and greater average scores for muscle disorders than other strains within their categories. The incidence and severity of myopathies in strain F were similar to those found in strain C and even greater than the values for strain B, despite the slower growth rate for strain F compared to the CONV birds. These two strains also had the greatest breast yield among all the strains tested, suggesting that despite the irrefutable role of ADG on the incidence of muscle disorders, breast yield may also be a major contributor to the high incidence of WB and WS.

Other groups have investigated the incidence of myopathies in strains selected for similar growth rates but different breast yield. Even though this association was not consistently found (Trocino et al., 2015), selection for greater breast yield has been linked with greater incidence of myopathies even when growth rates are similar (Petracci et al., 2013; Lorenzi et al., 2014). In fact, a study conducted to determine the genetic parameters of WS found that this myopathy is more genetically

correlated to the development of the breast muscle than to the increase in BW (Alnahhas et al., 2016). The association between high breast yield and the incidence of muscle disorders has been speculated to be the result of a mismatch between increased fibre size and inadequate capillary development which can lead to insufficient oxygen and nutrients supplied to muscle cells and impaired removal of lactic acid from the muscle, resulting in muscle damage (Petracci et al., 2015; Chen et al., 2019; Kuttappan et al., 2013a). In fact, Hoving-Bolink et al. (2000) found that lower capillary density was associated with higher breast muscle yield but not with body weight, suggesting that selection for increased percentage of breast muscle may lead to diminished oxygen supply to the breast muscle tissue and possibly detrimental effects on meat quality.

4.5.2.3 Effects of Sex, Target Weight, and Body Weight

Sex and target weight also influenced the incidence of muscle disorders; males and birds processed at TW 2 had the greatest values for WB and WS. Overall, females had higher breast yield than males, yet lower incidence of WB and WS compared to males. Both growth rate and breast yield have been shown to influence the occurrence of myopathies in broiler chickens (Kuttappan et al., 2012a; Alnahhas et al., 2015; Kuttappan et al., 2017). Therefore, the lower occurrence of breast fillets affected by myopathies in females might be a result of their lower BW, lower average daily gain, and lighter breast fillets compared to males. Other studies have suggested WB and WS are mainly affected by non-genetic factors (Bailey et al., 2015, 2020; Kuttappan et al., 2017). Interestingly, the effects of target weight and sex disappeared with the inclusion of BW in the statistical model, whereas the differences among categories and strains remained. Since males were heavier than females and birds processed at TW 2 were heavier than those processed at TW 1, the effects of sex and target weight on myopathies incidence are most likely due to the

differences in BW. Similar findings were reported by Kuttappan et al. (2013a), who suggested that the greater percentage of breast fillets affected by WS in males may be the result of their heavier carcass weights and thicker breast fillets compared to females processed at a similar age. Alternatively, differences in gene expression between sexes may have contributed to the higher susceptibility of males to myopathies, as suggested by Brothers et al. (2019), who found almost 200 genes upregulated in males that may influence metabolic processes involved in the occurrence of WB disease.

Despite the initial plan to process the birds at the same BW to allow comparisons between strains by weight, this was not possible due to the large differences in growth rate among strains, availability of the processing plant, and logistics for the project. Since differences in BW may affect the percentage and severity of breast fillets affected by myopathies, this factor should be controlled for in future studies. However, because the effects of categories and strains remained even when the differences in BW were considered, genetics appears to play a crucial role in the incidence of muscle disorders in broiler chickens.

4.6 Conclusion

In conclusion, our results indicate that processing traits and the incidence and severity of WB and WS are affected by strain, primarily dependent on growth rates. CONV birds had lower thigh, drumstick, and wing yields, yet greater breast yield and percentage of breast fillets exhibiting muscle disorders compared to the other categories of strains. The greater incidence for WB and WS in strains selected for similar growth rates but greater breast yield suggests that increased breast meat development and yield may be the underpinning factor associated with muscle abnormalities in both fast and slower-growing strains. Further field studies should be conducted

to investigate if the results obtained in our study under experimental conditions can be extrapolated to commercial poultry houses.

Table 4.1: Effect of category on live weight (LW), carcass weight (CW) and cut-up yields of broiler chickens (LS-means \pm SEM) at Target Weights 1 and 2. At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

Variable	Category			
	CONV	FAST	MOD	SLOW
Target Weight 1¹				
Live wt (g)	1,881 \pm 36.9 ^c	2,431 \pm 31.0 ^a	2,348 \pm 28.3 ^a	2,001 \pm 24.1 ^b
Carcass wt (g)	1,327 \pm 27.9 ^b	1,736 \pm 24.0 ^a	1,661 \pm 21.4 ^a	1,373 \pm 17.8 ^b
Breast wt (g)	400.6 \pm 11.08 ^c	483.9 \pm 8.72 ^a	443.3 \pm 7.53 ^b	330.3 \pm 5.62 ^d
Thigh wt (g)	202.9 \pm 4.52 ^c	273.4 \pm 3.99 ^a	266.1 \pm 3.64 ^a	226.9 \pm 3.11 ^b
Drumstick wt (g)	171.3 \pm 2.93 ^c	231.2 \pm 2.58 ^a	224.7 \pm 2.36 ^a	192.5 \pm 2.02 ^b
Wing wt (g)	134.8 \pm 2.66 ^c	183.3 \pm 2.36 ^a	182.5 \pm 2.21 ^a	158.0 \pm 1.91 ^b
Carcass yield (% LW) ²	70.6 \pm 0.42 ^a	71.4 \pm 0.28 ^a	70.8 \pm 0.26 ^a	68.7 \pm 0.26 ^b
Breast yield (% CW) ³	30.2 \pm 0.35 ^a	27.9 \pm 0.22 ^b	26.7 \pm 0.19 ^c	24.0 \pm 0.18 ^d
Breast yield (% LW)	21.3 \pm 0.28 ^a	19.9 \pm 0.17 ^b	18.9 \pm 0.16 ^c	16.5 \pm 0.14 ^d
Thigh yield (% CW)	15.3 \pm 0.27 ^b	15.8 \pm 0.18 ^b	16.0 \pm 0.17 ^{ab}	16.5 \pm 0.18 ^a
Thigh yield (% LW)	10.8 \pm 0.19	11.2 \pm 0.13	11.3 \pm 0.12	11.4 \pm 0.12
Drumstick yield (% CW)	12.9 \pm 0.14 ^c	13.3 \pm 0.10 ^{bc}	13.5 \pm 0.09 ^b	14.0 \pm 0.09 ^a
Drumstick yield (% LW)	9.1 \pm 0.09 ^b	9.5 \pm 0.07 ^a	9.6 \pm 0.06 ^a	9.6 \pm 0.06 ^a
Wing yield (% CW)	10.1 \pm 0.13 ^c	10.6 \pm 0.09 ^c	11.0 \pm 0.09 ^b	11.5 \pm 0.09 ^a
Wing yield (% LW)	7.2 \pm 0.09 ^c	7.5 \pm 0.06 ^b	7.8 \pm 0.06 ^a	7.9 \pm 0.06 ^a
Target Weight 2⁴				
Live wt (g)	3,285 \pm 58.8 ^{ab}	3,417 \pm 42.1 ^a	3,182 \pm 36.4 ^b	2,809 \pm 32.0 ^c
Carcass wt (g)	2,415 \pm 46.4 ^{ab}	2,495 \pm 33.1 ^a	2,314 \pm 28.4 ^b	1,994 \pm 24.4 ^c
Breast wt (g)	814.6 \pm 20.58 ^a	733.2 \pm 12.78 ^b	642.4 \pm 10.37 ^c	503.8 \pm 8.11 ^d
Thigh wt (g)	351.2 \pm 7.14 ^b	384.7 \pm 5.48 ^a	366.1 \pm 4.78 ^{ab}	325.2 \pm 4.23 ^c
Drumstick wt (g)	301.3 \pm 4.70 ^b	322.7 \pm 3.52 ^a	306.7 \pm 3.08 ^b	270.9 \pm 2.71 ^c
Wing wt (g)	226.6 \pm 4.07 ^{bc}	249.0 \pm 3.11 ^a	238.9 \pm 2.75 ^{ab}	217.6 \pm 2.50 ^c
Carcass yield (% LW)	73.5 \pm 0.39 ^a	73.1 \pm 0.28 ^a	72.7 \pm 0.26 ^a	71.0 \pm 0.26 ^b
Breast yield (% CW)	33.7 \pm 0.37 ^a	29.4 \pm 0.21 ^b	27.8 \pm 0.19 ^c	25.3 \pm 0.17 ^d
Breast yield (% LW)	24.8 \pm 0.30 ^a	21.4 \pm 0.18 ^b	20.2 \pm 0.16 ^c	17.9 \pm 0.141 ^d
Thigh yield (% CW)	14.5 \pm 0.24 ^c	15.4 \pm 0.17 ^b	15.8 \pm 0.16 ^{ab}	16.3 \pm 0.17 ^a
Thigh yield (% LW)	10.7 \pm 0.17 ^b	11.2 \pm 0.12 ^a	11.5 \pm 0.11 ^a	11.6 \pm 0.12 ^a
Drumstick yield (% CW)	12.5 \pm 0.13 ^d	12.9 \pm 0.09 ^c	13.3 \pm 0.08 ^b	13.6 \pm 0.09 ^a
Drumstick yield (% LW)	9.2 \pm 0.09 ^c	9.4 \pm 0.06 ^{bc}	9.6 \pm 0.06 ^{ab}	9.7 \pm 0.02 ^a
Wing yield (% CW)	9.4 \pm 0.11 ^d	9.9 \pm 0.08 ^c	10.3 \pm 0.08 ^b	10.9 \pm 0.08 ^a
Wing yield (% LW)	6.9 \pm 0.08 ^d	7.3 \pm 0.06 ^c	7.5 \pm 0.06 ^b	7.7 \pm 0.06 ^a

^{a-d} Different superscripts within the same row represent differences among categories ($P < 0.05$).

¹ Number of birds per category at Target Weight 1: CONV: $n = 64$; FAST: $n = 155$; MOD: $n = 174$; SLOW: $n = 173$.

² Yields calculated relative to live weight obtained one day before processing.

³ Yields calculated as a ratio to the eviscerated carcass weight.

⁴ Number of birds per category at Target Weight 2: CONV: $n = 80$; FAST: $n = 161$; MOD: $n = 186$; SLOW: $n = 187$.

Table 4.2: Mean values (LS-means \pm SEM) for live weight (LW), carcass weight (CW) and cut-up yields among CONV strains of broiler chickens at Target Weights 1 and 2. At Target Weights 1 and 2, CONV strains were 34 and 48 days, respectively.

Variable	Strain	
	B	C
Target Weight 1¹		
Live wt (g)	1,959 \pm 54.4	1,807 \pm 50.1
Carcass wt (g)	1,369 \pm 40.8	1,288 \pm 38.3
Breast wt (g)	393.5 \pm 15.40	407.8 \pm 15.95
Thigh wt (g)	214.5 \pm 6.76	191.9 \pm 6.04
Drumstick wt (g)	174.9 \pm 4.23	167.8 \pm 4.05
Wing wt (g)	137.7 \pm 3.83	132.0 \pm 3.68
Carcass yield (% LW) ²	69.9 \pm 0.67	71.3 \pm 0.68
Breast yield (% CW) ³	28.7 \pm 0.48 ^b	31.7 \pm 0.53 ^a
Breast yield (% LW)	20.1 \pm 0.38 ^b	22.6 \pm 0.43 ^a
Thigh yield (% CW)	15.7 \pm 0.39	14.9 \pm 0.37
Thigh yield (% LW)	10.9 \pm 0.27	10.6 \pm 0.26
Drumstick yield (% CW)	12.8 \pm 0.20	13.0 \pm 0.20
Drumstick yield (% LW)	8.9 \pm 0.14	9.3 \pm 0.14
Wing yield (% CW)	10.1 \pm 0.18	10.2 \pm 0.19
Wing yield (% LW)	7.0 \pm 0.13	7.3 \pm 0.13
Target Weight 2⁴		
Live wt (g)	3,288 \pm 74.5	3,283 \pm 91.1
Carcass wt (g)	2,411 \pm 58.6	2,419 \pm 72.0
Breast wt (g)	780.4 \pm 24.93	850.2 \pm 33.28
Thigh wt (g)	360.7 \pm 9.28	341.9 \pm 10.77
Drumstick wt (g)	303.5 \pm 5.99	299.2 \pm 7.23
Wing wt (g)	234.2 \pm 5.33	219.2 \pm 6.11
Carcass yield (% LW)	73.4 \pm 0.57	73.7 \pm 0.71
Breast yield (% CW)	32.3 \pm 0.44 ^b	35.1 \pm 0.59 ^a
Breast yield (% LW)	23.7 \pm 0.37 ^b	25.9 \pm 0.49 ^a
Thigh yield (% CW)	14.9 \pm 0.30	14.1 \pm 0.35
Thigh yield (% LW)	11.0 \pm 0.22	10.4 \pm 0.26
Drumstick yield (% CW)	12.6 \pm 0.16	12.4 \pm 0.19
Drumstick yield (% LW)	9.2 \pm 0.11	9.1 \pm 0.14
Wing yield (% CW)	9.7 \pm 0.14	9.1 \pm 0.16
Wing yield (% LW)	7.1 \pm 0.11	6.7 \pm 0.12

^{a-b} Different superscripts within the same row represent differences among categories (P < 0.05).

¹ Number of birds per strain at Target Weight 1: Strain B: n = 32; Strain C: n = 32.

² Yields calculated relative to live weight obtained one day before processing.

³ Yields calculated as a ratio to the eviscerated carcass weight.

⁴ Number of birds per strain at Target Weight 2: Strain B: n = 48; Strain C: n = 32.

Table 4.3: Mean values (LS-means \pm SEM) for live weight (LW), carcass weight (CW) and cut-up yields among FAST strains of broiler chickens at Target Weights 1 and 2. At Target Weight 1 and 2, FAST strains were 48 and 62 d of age, respectively.

Variable	Strain			
	F	G	I	M
Target Weight 1¹				
Live wt (g)	2,522 \pm 71.0	2,338 \pm 53.2	2,402 \pm 54.7	2,464 \pm 68.8
Carcass wt (g)	1,851 \pm 56.0	1,659 \pm 40.5	1,726 \pm 42.4	1,716 \pm 51.5
Breast wt (g)	621.2 \pm 24.71 ^a	448.0 \pm 14.76 ^b	462.9 \pm 14.87 ^b	425.8 \pm 16.78 ^b
Thigh wt (g)	274.5 \pm 8.89	271.1 \pm 7.03	276.1 \pm 7.17	271.9 \pm 8.68
Drumstick wt (g)	232.9 \pm 5.77	223.7 \pm 4.44	232.3 \pm 4.61	236.0 \pm 5.77
Wing wt (g)	185.3 \pm 5.27	176.7 \pm 4.04	181.3 \pm 4.15	190.2 \pm 5.35
Carcass yield (% LW) ²	73.4 \pm 0.72 ^a	70.9 \pm 0.56 ^b	71.7 \pm 0.57 ^{ab}	69.6 \pm 0.68 ^b
Breast yield (% CW) ³	33.5 \pm 0.58 ^a	27.0 \pm 0.37 ^b	26.9 \pm 0.38 ^b	24.9 \pm 0.42 ^c
Breast yield (% LW)	24.6 \pm 0.48 ^a	19.2 \pm 0.30 ^b	19.2 \pm 0.31 ^b	17.3 \pm 0.33 ^c
Thigh yield (% CW)	14.8 \pm 0.38	16.3 \pm 0.34	16.0 \pm 0.33	15.8 \pm 0.40
Thigh yield (% LW)	10.9 \pm 0.26	11.6 \pm 0.24	11.5 \pm 0.23	11.0 \pm 0.27
Drumstick yield (% CW)	12.6 \pm 0.20 ^b	13.5 \pm 0.17 ^a	13.5 \pm 0.17 ^a	13.8 \pm 0.22 ^a
Drumstick yield (% LW)	9.2 \pm 0.14 ^b	9.6 \pm 0.12 ^a	9.7 \pm 0.12 ^a	9.6 \pm 0.15 ^a
Wing yield (% CW)	10.0 \pm 0.19 ^b	10.6 \pm 0.16 ^{ab}	10.5 \pm 0.16 ^{ab}	11.1 \pm 0.20 ^a
Wing yield (% LW)	7.3 \pm 0.14	7.6 \pm 0.12	7.5 \pm 0.11	7.8 \pm 0.14
Target Weight 2⁴				
Live wt (g)	3,391 \pm 82.6	3,357 \pm 76.1	3,457 \pm 79.0	3,464 \pm 98.3
Carcass wt (g)	2,579 \pm 67.7	2,453 \pm 59.6	2,487 \pm 61.07	2,461 \pm 75.1
Breast wt (g)	886.1 \pm 30.54 ^a	695.3 \pm 22.22 ^b	692.1 \pm 22.34 ^b	677.7 \pm 27.17 ^b
Thigh wt (g)	367.8 \pm 10.52	394.5 \pm 10.15	396.6 \pm 10.39	380.7 \pm 12.51
Drumstick wt (g)	318.4 \pm 6.95	322.4 \pm 6.36	323.2 \pm 6.47	326.8 \pm 8.18
Wing wt (g)	247.8 \pm 6.16	247.4 \pm 5.63	249.9 \pm 5.76	250.7 \pm 7.20
Carcass yield (% LW)	76.0 \pm 0.66 ^a	73.1 \pm 0.57 ^b	71.9 \pm 0.57 ^b	71.0 \pm 0.71 ^b
Breast yield (% CW)	34.3 \pm 0.52 ^a	28.3 \pm 0.39 ^b	27.8 \pm 0.39 ^b	27.5 \pm 0.48 ^b
Breast yield (% LW)	26.1 \pm 0.44 ^a	20.7 \pm 0.32 ^b	20.0 \pm 0.31 ^b	19.5 \pm 0.38 ^b
Thigh yield (% CW)	14.2 \pm 0.33 ^b	16.1 \pm 0.33 ^a	16.0 \pm 0.33 ^a	15.5 \pm 0.40 ^{ab}
Thigh yield (% LW)	10.8 \pm 0.24	11.7 \pm 0.24	11.5 \pm 0.23	11.0 \pm 0.28
Drumstick yield (% CW)	12.2 \pm 0.17 ^b	13.1 \pm 0.17 ^a	13.0 \pm 0.16 ^{ab}	13.3 \pm 0.21 ^a
Drumstick yield (% LW)	9.3 \pm 0.12 ^c	9.6 \pm 0.12 ^b	9.3 \pm 0.12 ^{ab}	9.4 \pm 0.15 ^a
Wing yield (% CW)	9.5 \pm 0.16	10.1 \pm 0.15	10.1 \pm 0.15	10.2 \pm 0.19
Wing yield (% LW)	7.2 \pm 0.12	7.4 \pm 0.11	7.2 \pm 0.11	7.2 \pm 0.14

^{a-b} Different superscripts within the same row represent differences among categories ($P < 0.05$).

¹ Number of birds per strain at Target Weight 1: Strain F: n = 30; Strain G: n = 47; Strain I: n = 47; Strain M: n = 31.

² Yields calculated relative to live weight obtained one day before processing.

³ Yields calculated as a ratio to the eviscerated carcass weight.

⁴ Number of birds per strain Target Weight 2: Strain F: n = 38; Strain G: n = 48; Strain I: n = 46; Strain M: n = 29.

Table 4.4: Mean values (LS-means \pm SEM) for live weight (LW), carcass weight (CW) and cut-up yields among MOD strains of broiler chickens at Target Weights 1 and 2. At Target Weight 1 and 2, MOD strains were 48 and 62 d of age, respectively.

Variable	Strain			
	E	H	O	S
Target Weight 1¹				
Live wt (g)	2,506 \pm 56.8 ^a	2,321 \pm 53.8 ^{ab}	2,365 \pm 53.8 ^{ab}	2,208 \pm 50.2 ^b
Carcass wt (g)	1,749 \pm 42.5	1,641 \pm 48.9	1,679 \pm 41.0	1,580 \pm 38.6
Breast wt (g)	451.3 \pm 14.42	439.8 \pm 17.21	463.1 \pm 14.87	419.9 \pm 13.49
Thigh wt (g)	284.0 \pm 7.31	257.8 \pm 8.12	272.3 \pm 7.07	251.1 \pm 6.52
Drumstick wt (g)	238.1 \pm 4.69 ^a	224.2 \pm 5.41 ^{ab}	224.2 \pm 4.46 ^{ab}	213.1 \pm 4.24 ^b
Wing wt (g)	194.6 \pm 4.43 ^a	181.5 \pm 5.06 ^{ab}	180.7 \pm 4.13 ^{ab}	173.8 \pm 3.98 ^b
Carcass yield (% LW) ²	69.8 \pm 0.54	70.7 \pm 0.67	71.0 \pm 0.56	71.5 \pm 0.56
Breast yield (% CW) ³	25.8 \pm 0.35 ^b	26.8 \pm 0.45 ^{ab}	27.6 \pm 0.38 ^a	26.6 \pm 0.37 ^{ab}
Breast yield (% LW)	18.0 \pm 0.27 ^b	18.9 \pm 0.36 ^{ab}	19.6 \pm 0.31 ^a	19.0 \pm 0.29 ^{ab}
Thigh yield (% CW)	16.2 \pm 0.33	15.7 \pm 0.39	16.2 \pm 0.34	15.9 \pm 0.33
Thigh yield (% LW)	11.3 \pm 0.23	11.0 \pm 0.27	11.5 \pm 0.23	11.4 \pm 0.23
Drumstick yield (% CW)	13.6 \pm 0.17	13.7 \pm 0.21	13.4 \pm 0.17	13.5 \pm 0.17
Drumstick yield (% LW)	9.5 \pm 0.12	9.7 \pm 0.15	9.5 \pm 0.12	9.6 \pm 0.12
Wing yield (% CW)	11.1 \pm 0.16	11.1 \pm 0.20	10.8 \pm 0.16	11.0 \pm 0.16
Wing yield (% LW)	7.8 \pm 0.12	7.8 \pm 0.14	7.6 \pm 0.11	7.9 \pm 0.12
Target Weight 2⁴				
Live wt (g)	3,285 \pm 74.5	3,135 \pm 73.7	3,252 \pm 73.7	3,059 \pm 69.3
Carcass wt (g)	2,387 \pm 58.0	2,261 \pm 57.2	2,387 \pm 58.0	2,226 \pm 54.1
Breast wt (g)	638.7 \pm 20.42	620.2 \pm 20.59	681.8 \pm 21.78	630.7 \pm 20.15
Thigh wt (g)	385.0 \pm 9.90	349.7 \pm 9.49	372.3 \pm 9.58	360.9 \pm 9.28
Drumstick wt (g)	319.6 \pm 6.31	299.3 \pm 6.29	314.1 \pm 6.20	295.4 \pm 5.83
Wing wt (g)	247.9 \pm 5.64	237.9 \pm 5.68	243.2 \pm 5.54	227.0 \pm 5.17
Carcass yield (% LW)	72.6 \pm 0.57	72.1 \pm 0.59	73.4 \pm 0.57	72.8 \pm 0.57
Breast yield (% CW)	26.8 \pm 0.37	27.4 \pm 0.40	28.6 \pm 0.39	28.3 \pm 0.39
Breast yield (% LW)	19.4 \pm 0.30 ^b	19.7 \pm 0.32 ^{ab}	21.0 \pm 0.32 ^a	20.6 \pm 0.32 ^{ab}
Thigh yield (% CW)	16.1 \pm 0.33	15.4 \pm 0.33	15.6 \pm 0.32	16.2 \pm 0.33
Thigh yield (% LW)	11.7 \pm 0.23	11.1 \pm 0.24	11.4 \pm 0.23	11.8 \pm 0.24
Drumstick yield (% CW)	13.4 \pm 0.17	13.2 \pm 0.17	13.2 \pm 0.17	13.3 \pm 0.17
Drumstick yield (% LW)	9.7 \pm 0.12	9.5 \pm 0.13	9.7 \pm 0.12	9.7 \pm 0.12
Wing yield (% CW)	10.4 \pm 0.15	10.5 \pm 0.17	10.2 \pm 0.15	10.2 \pm 0.15
Wing yield (% LW)	7.5 \pm 0.11	7.6 \pm 0.12	7.5 \pm 0.11	7.4 \pm 0.11

^{a-b} Different superscripts within the same row represent differences among categories ($P < 0.05$).

¹ Number of birds per strain at Target Weight 1: Strain E: n = 48; Strain H: n = 32; Strain O: n = 47; Strain S: n = 47.

² Yields calculated relative to live weight obtained one day before processing.

³ Yields calculated as a ratio to the eviscerated carcass weight.

⁴ Number of birds per strain at Target Weight 2: Strain E: n = 48; Strain H: n = 42; Strain O: n = 48; Strain S: n = 48.

Table 4.5: Mean values (LS-means \pm SEM) for live weight (LW), carcass weight (CW) and cut-up yields among SLOW strains of broiler chickens at Target Weights 1 and 2. At Target Weight 1 and 2, SLOW strains were 48 and 62 d of age, respectively.

Variable	Strain			
	D	J	K	N
Target Weight 1¹				
Live wt (g)	2,025 \pm 56.2	2,072 \pm 47.2	1,942 \pm 44.2	1,967 \pm 44.8
Carcass wt (g)	1,352 \pm 40.3	1,444 \pm 35.3	1,355 \pm 33.1	1,346 \pm 33.0
Breast wt (g)	281.9 \pm 11.03 ^c	380.3 \pm 12.21 ^a	324.9 \pm 10.43 ^{bc}	341.8 \pm 10.98 ^{ab}
Thigh wt (g)	228.0 \pm 7.18	233.3 \pm 6.05	223.1 \pm 5.79	223.7 \pm 5.80
Drumstick wt (g)	198.9 \pm 4.80	197.7 \pm 3.93	189.7 \pm 3.77	184.0 \pm 3.66
Wing wt (g)	158.4 \pm 4.42	160.2 \pm 3.67	160.9 \pm 3.68	152.5 \pm 3.49
Carcass yield (% LW) ²	66.8 \pm 0.64 ^b	69.7 \pm 0.55 ^a	69.8 \pm 0.55 ^a	68.4 \pm 0.54 ^{ab}
Breast yield (% CW) ³	20.8 \pm 0.35 ^c	26.3 \pm 0.36 ^a	24.0 \pm 0.33 ^b	25.5 \pm 0.36 ^{ab}
Breast yield (% LW)	13.9 \pm 0.26 ^c	18.3 \pm 0.29 ^a	16.7 \pm 0.26 ^b	17.4 \pm 0.27 ^{ab}
Thigh yield (% CW)	16.9 \pm 0.42	16.1 \pm 0.33	16.5 \pm 0.34	16.7 \pm 0.35
Thigh yield (% LW)	11.3 \pm 0.28	11.2 \pm 0.23	11.5 \pm 0.23	11.4 \pm 0.23
Drumstick yield (% CW)	14.7 \pm 0.23 ^a	13.7 \pm 0.17 ^b	14.0 \pm 0.18 ^{ab}	13.7 \pm 0.18 ^b
Drumstick yield (% LW)	9.8 \pm 0.15	9.5 \pm 0.12	9.8 \pm 0.12	9.4 \pm 0.12
Wing yield (% CW)	11.7 \pm 0.21	11.1 \pm 0.17	11.9 \pm 0.18	11.3 \pm 0.17
Wing yield (% LW)	7.8 \pm 0.14	7.8 \pm 0.14	8.3 \pm 0.13	7.7 \pm 0.12
Target Weight 2⁴				
Live wt (g)	2,805 \pm 65.1	2,936 \pm 66.5	2,753 \pm 62.7	2,746.1 \pm 62.2
Carcass wt (g)	1,951 \pm 48.7	2,134 \pm 51.9	1,924 \pm 47.0	1,974 \pm 47.9
Breast wt (g)	429.4 \pm 14.07 ^c	594.3 \pm 18.99 ^a	492.7 \pm 15.82 ^{bc}	512.3 \pm 16.37 ^{ab}
Thigh wt (g)	326.8 \pm 8.75	342.2 \pm 8.80	320.6 \pm 8.32	311.9 \pm 8.02
Drumstick wt (g)	274.0 \pm 5.66	281.6 \pm 5.56	266.7 \pm 5.30	261.8 \pm 5.17
Wing wt (g)	221.9 \pm 5.21	215.6 \pm 4.91	220.6 \pm 5.05	212.5 \pm 4.84
Carcass yield (% LW)	69.5 \pm 0.57 ^c	72.7 \pm 0.57 ^a	69.9 \pm 0.55 ^{bc}	71.9 \pm 0.56 ^{ab}
Breast yield (% CW)	22.1 \pm 0.31 ^c	27.8 \pm 0.38 ^a	25.8 \pm 0.35 ^b	25.9 \pm 0.36 ^b
Breast yield (% LW)	15.3 \pm 0.25 ^c	20.2 \pm 0.31 ^a	17.9 \pm 0.28 ^b	18.7 \pm 0.29 ^b
Thigh yield (% CW)	16.8 \pm 0.36	16.0 \pm 0.33	16.8 \pm 0.35	15.8 \pm 0.32
Thigh yield (% LW)	11.7 \pm 0.24	11.7 \pm 0.23	11.7 \pm 0.24	11.4 \pm 0.23
Drumstick yield (% CW)	14.1 \pm 0.17 ^a	13.2 \pm 0.17 ^b	13.9 \pm 0.18 ^{ab}	13.3 \pm 0.17 ^b
Drumstick yield (% LW)	9.8 \pm 0.13	9.6 \pm 0.12	9.7 \pm 0.12	9.5 \pm 0.12
Wing yield (% CW)	11.4 \pm 0.17 ^a	10.0 \pm 0.15 ^b	11.5 \pm 0.17 ^a	10.8 \pm 0.16 ^{ab}
Wing yield (% LW)	7.9 \pm 0.13 ^a	7.3 \pm 0.11 ^b	8.0 \pm 0.12 ^a	7.7 \pm 0.12 ^{ab}

^{a-c} Different superscripts within the same row represent differences among categories ($P < 0.05$).

¹ Number of birds per strain at Target Weight 1: Strain D: n = 32; Strain J: n = 47; Strain K: n = 47; Strain N: n = 47.

² Yields calculated relative to live weight obtained one day before processing.

³ Yields calculated as a ratio to the eviscerated carcass weight.

⁴ Number of birds per strain at Target Weight 2: Strain D: n = 44; Strain J: n = 48; Strain K: n = 47; Strain N: n = 48.

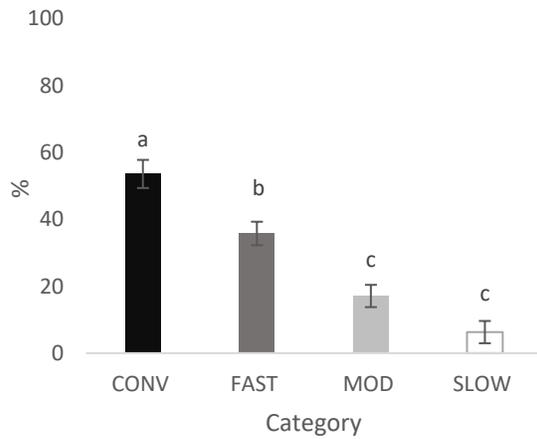
Table 4.6: Spearman correlation coefficients (r_s) between total incidence of myopathies and carcass parts and yield (as hatched broiler chickens) with the respective p-values in each category. Results were calculated on a pen basis.

Variable	Coef. of correlation and p-value	Conventional		Fast		Mod		Slow	
		% of WB ¹	% of WS ²	% of WB ¹	% of WS ²	% of WB ¹	% of WS ²	% of WB ¹	% of WS ²
Live weight	r_s	0.482	0.505	0.309	0.291	0.535	0.404	0.285	0.275
	p	0.003	0.002	0.006	0.010	<0.001	<0.001	0.008	0.001
Carcass yield	r_s	0.546	0.369	0.605	0.719	0.417	0.478	0.233	0.279
	p	0.001	0.028	<0.001	<0.001	<0.001	<0.001	0.030	0.009
Breast yield	r_s	0.450	0.431	0.589	0.596	0.462	0.236	0.338	0.265
	p	0.007	0.009	<0.001	<0.001	<0.001	0.012	0.001	0.013
Thigh yield	r_s	-0.055	-0.068	-0.303	-0.188	-0.094	0.012	-0.095	-0.077
	p	0.752	0.697	0.007	0.101	0.368	0.921	0.382	0.476
Drumstick yield	r_s	-0.042	-0.385	-0.282	-0.338	-0.025	0.055	-0.164	-0.106
	p	0.809	0.002	0.012	0.003	0.850	0.606	0.128	0.330
Wing yield	r_s	-0.265	-0.167	-0.511	-0.502	-0.377	-0.282	-0.223	-0.168
	p	0.123	0.337	<0.001	<0.001	0.003	0.007	0.038	0.119

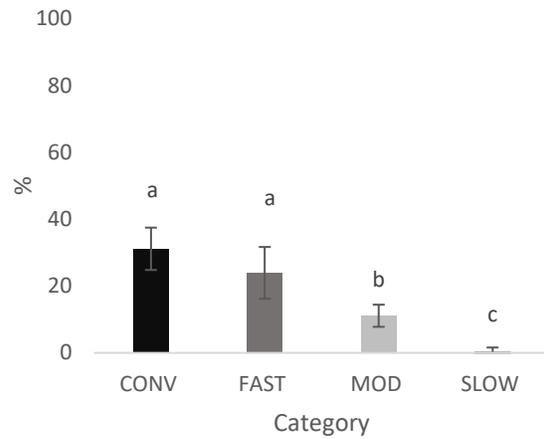
¹ Number of birds per category: CONV: n = 184; FAST: n = 332; MOD: n = 371; SLOW: n = 371.

² Number of birds per category: CONV: n = 155; FAST: n = 256; MOD: n = 330; SLOW: n = 287.

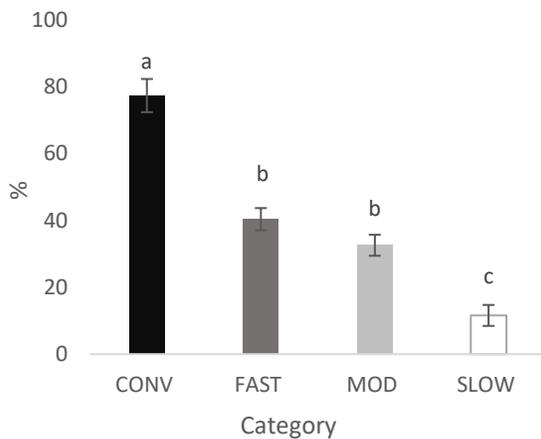
Incidence of wooden breast- Target weight 1¹



Incidence of white striping- Target weight 1³



Incidence of wooden breast- Target weight 2²



Incidence of white striping- Target weight 2⁴

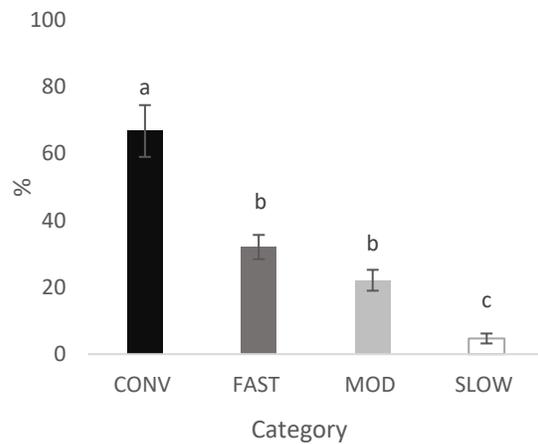


Figure 4.1: Effects of category on the total incidence of breast fillets presenting wooden breast^{1,2} and white striping^{3,4} at Target Weights 1 and 2 (LS-means \pm SEM). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

^{a-c} Different superscripts represent differences among categories ($P < 0.05$).

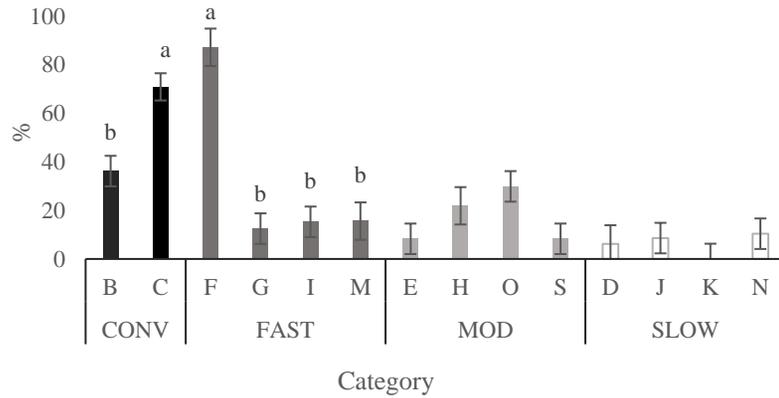
¹ Number of birds per category at Target Weight 1: CONV: $n = 104$; FAST: $n = 156$; MOD: $n = 175$; SLOW: $n = 173$.

² Number of birds per category at Target Weight 2 CONV: $n = 80$; FAST: $n = 176$; MOD: $n = 196$; SLOW: $n = 198$.

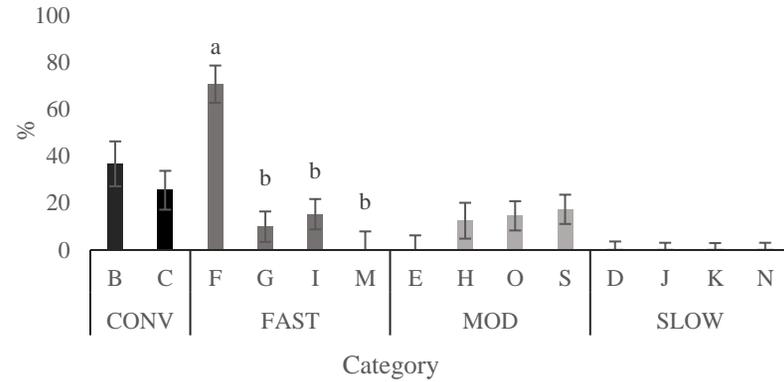
³ Number of birds per category at Target Weight 1: CONV: $n = 91$; FAST: $n = 124$; MOD: $n = 159$; SLOW: $n = 125$.

⁴ Number of birds per category at Target Weight 2: CONV: $n = 64$; FAST: $n = 132$; MOD: $n = 171$; SLOW: $n = 162$.

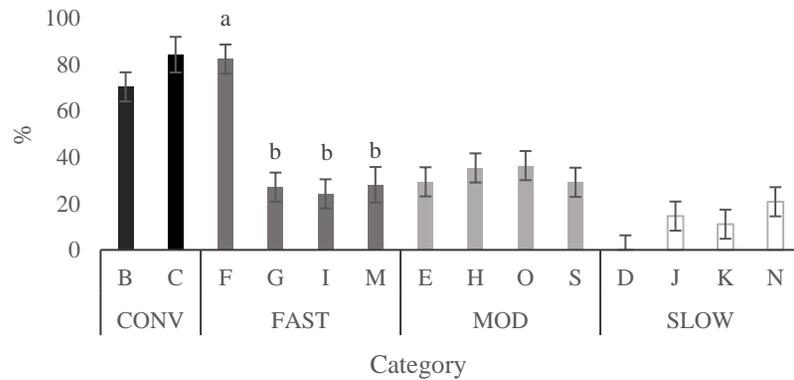
Incidence of wooden breast - Target weight 1¹



Incidence of white striping- Target weight 1³



Incidence of wooden breast- Target weight 2²



Incidence of white striping- Target weight 2⁴

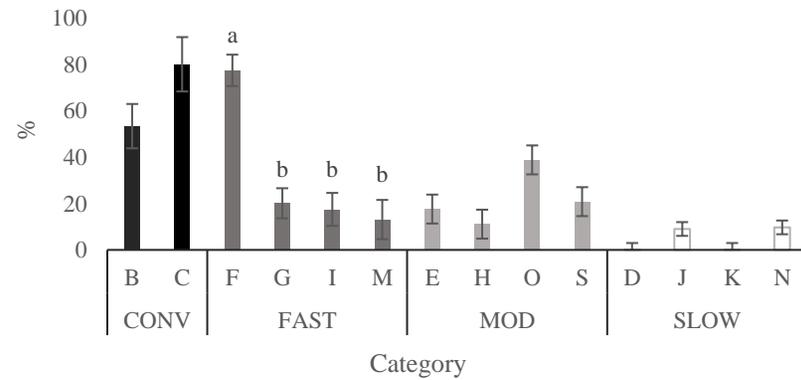


Figure 4.2: Effects of strains (within category) on the total incidence of breast fillets presenting wooden breast^{1,2} and white striping^{3,4} at Target Weights 1 and 2 (LS-means \pm SEM). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

^{a-b} Different superscripts within the same category represent differences among strains ($P < 0.05$).

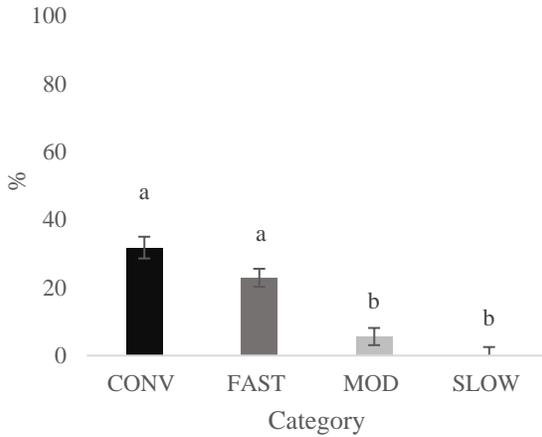
¹ Number of birds per strain at Target Weight 1: B: n = 52, C: n = 52, F: n = 30, G: n = 47, I: n = 47, M: n = 32, E: n = 48, H: n = 32, O: n = 47, S: n = 48; D: n = 32; J: n = 47, K: n = 47, N: n = 47.

² Number of birds per strain at Target Weight 2: B: n = 48, C: n = 32, F: n = 52, G: n = 48, I: n = 47, M: n = 29, E: n = 48, H: n = 52, O: n = 48, S: n = 48, D: n = 55, J: n = 48, K: n = 47, N: n = 48.

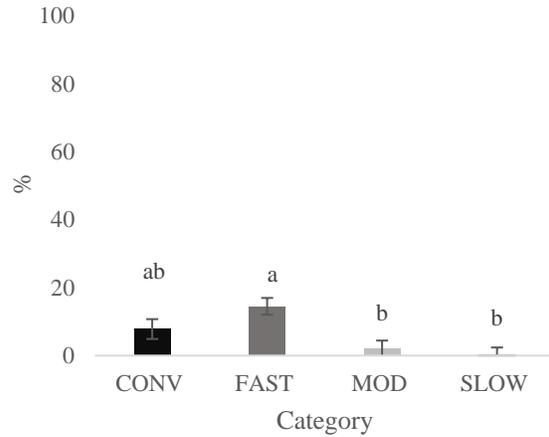
³ Number of birds per strain at Target Weight 1: B: n = 45, C: n = 46, F: n = 25, G: n = 39, I: n = 39, M: n = 21, E: n = 47, H: n = 24, O: n = 44, S: n = 44, D: n = 26, J: n = 28, K: n = 39, N: n = 32.

⁴ Number of birds per strain at Target Weight 2: B: n = 36, C: n = 28, F: n = 42, G: n = 41, I: n = 31, M: n = 18, E: n = 40, H: n = 48, O: n = 44, S: n = 39, D: n = 45, J: n = 37, K: n = 44, N: n = 36.

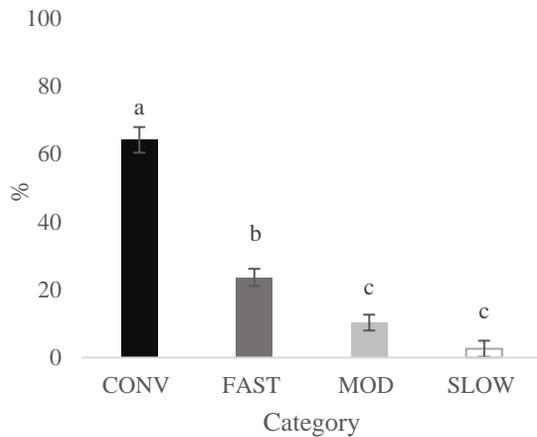
Moderate-severe wooden breast- Target weight 1¹



Moderate-severe white striping- Target weight 1³



Moderate-severe wooden breast- Target weight 2²



Moderate-severe white striping- Target weight 2⁴

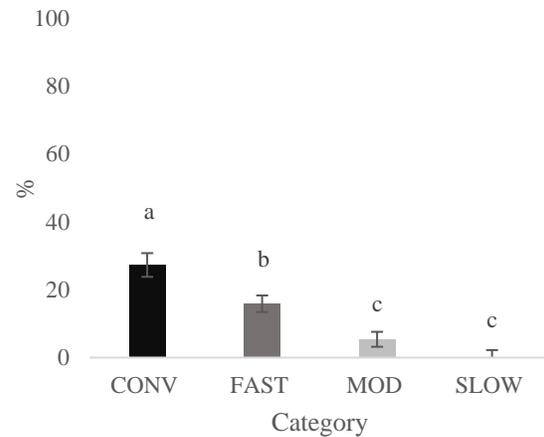


Figure 4.3: Effects of category on the total incidence of breast fillets presenting moderately severe (scores 2 or 3) wooden breast^{1,2} and white striping^{3,4} at Target Weights 1 and 2 (LS-means \pm SEM). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

^{a-c} Different superscripts represent differences among categories ($P < 0.05$).

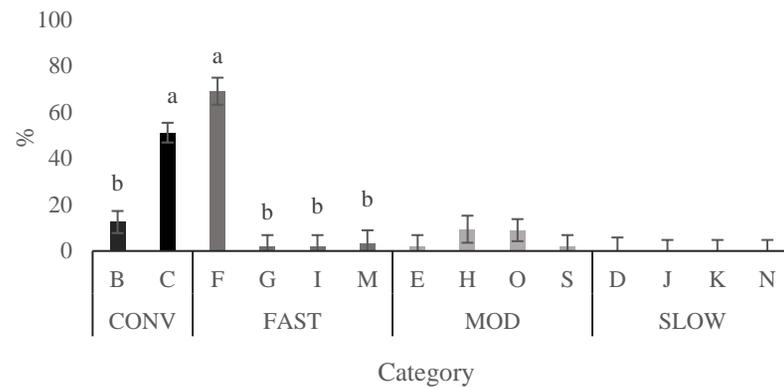
¹ Number of birds per category at Target Weight 1: CONV: $n = 104$; FAST: $n = 156$; MOD: $n = 175$; SLOW: $n = 173$.

² Number of birds per category at Target Weight 2 CONV: $n = 80$; FAST: $n = 176$; MOD: $n = 196$; SLOW: $n = 198$.

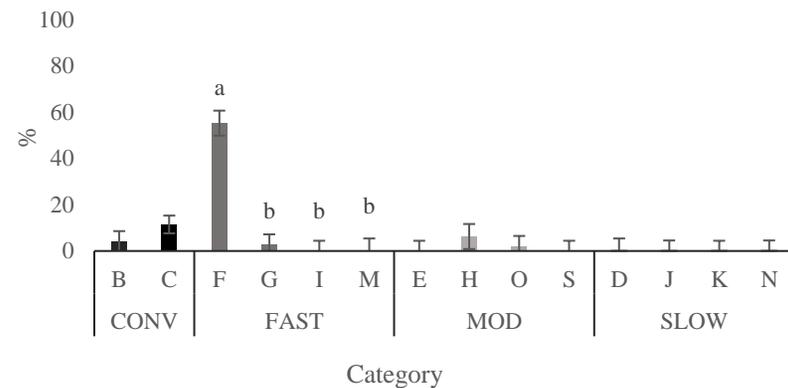
³ Number of birds per category at Target Weight 1: CONV: $n = 91$; FAST: $n = 124$; MOD: $n = 159$; SLOW: $n = 125$.

⁴ Number of birds per category at Target Weight 2: CONV: $n = 64$ FAST: $n = 132$; MOD: $n = 171$; SLOW: $n =$

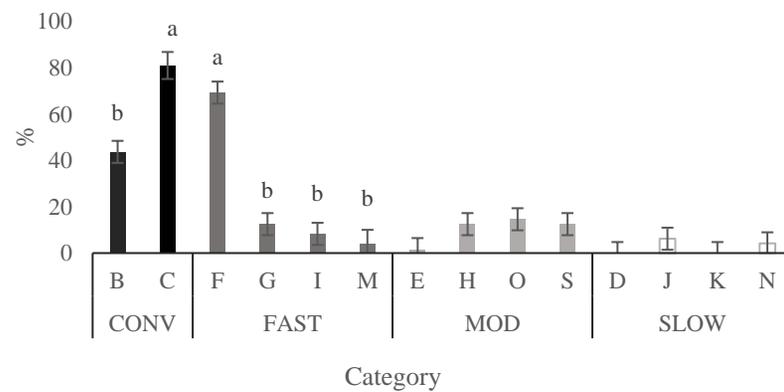
Moderate-severe wooden breast- Target weight 1¹



Moderate-severe white striping- Target weight 1³



Moderate-severe wooden breast- Target weight 2²



Moderate-severe white striping- Target weight 2⁴

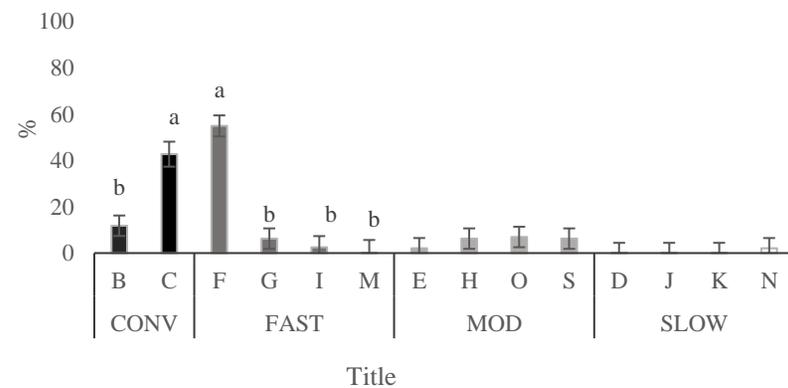


Figure 4.4: Effects of strains (within category) on the total incidence of breast fillets presenting moderately severe (scores 2 or 3) wooden breast^{1,2} and white striping^{3,4} at Target Weights 1 and 2 (LS-means \pm SEM). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

^{a-b} Different superscripts within the same category represent differences among strains ($P < 0.05$).

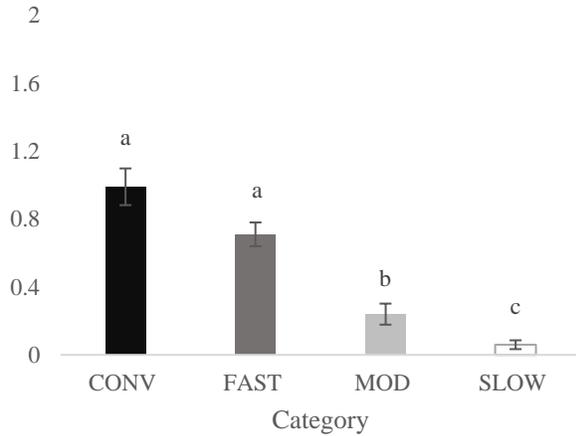
¹ Number of birds per strain at Target Weight 1: B: n = 52, C: n = 52, F: n = 30, G: n = 47, I: n = 47, M: n = 32, E: n = 48, H: n = 32, O: n = 47, S: n = 48, D: n = 32, J: n = 47, K: n = 47, N: n = 47.

² Number of birds per strain at Target Weight 2: B: n = 48, C: n = 32, F: n = 52, G: n = 48, I: n = 47, M: n = 29, E: n = 48, H: n = 52, O: n = 48, S: n = 48, D: n = 55, J: n = 48, K: n = 47, N: n = 48.

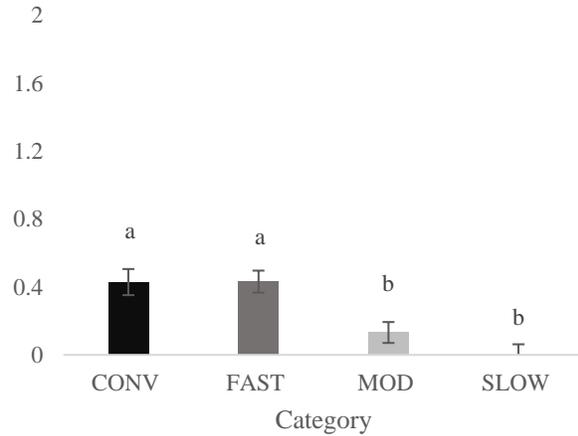
³ Number of birds per strain at Target Weight 1: B: n = 45, C: n = 46, F: n = 25, G: n = 39, I: n = 39, M: n = 21, E: n = 47, H: n = 24, O: n = 44, S: n = 44, D: n = 26, J: n = 28, K: n = 39, N: n = 32.

⁴ Number of birds per strain at Target Weight 2: B: n = 36, C: n = 28, F: n = 42, G: n = 41, I: n = 31, M: n = 18, E: n = 40, H: n = 48, O: n = 44, S: n = 39, D: n = 45, J: n = 37, K: n = 44, N: n = 36.

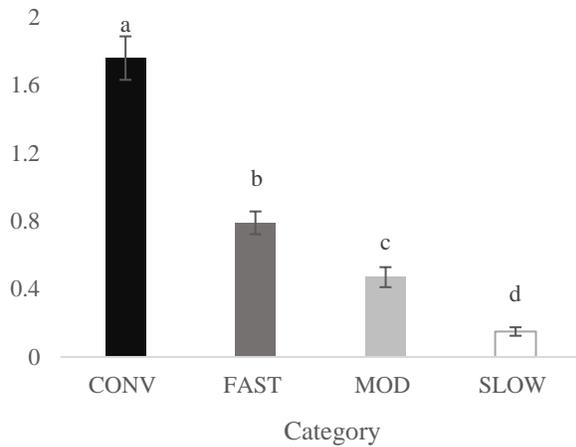
Average wooden breast score- Target weight 1¹



Average white striping score- Target weight 1³



Average wooden breast score- Target weight 2²



Average white striping score- Target weight⁴

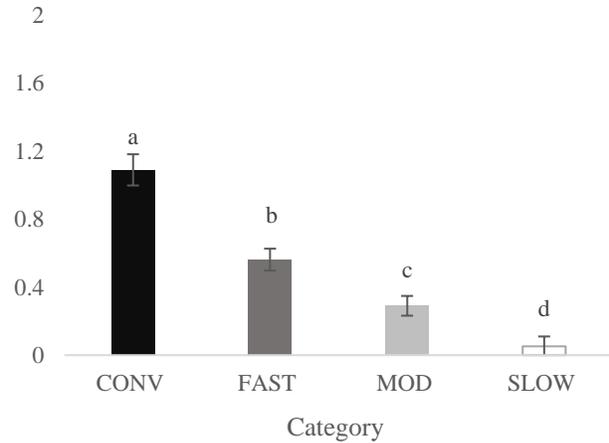


Figure 4.5: Effects of category on average of wooden breast^{1,2} and white striping^{3,4} scores at Target Weights 1 and 2 (LS-means \pm SEM). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

^{a-d} Different superscripts represent differences among categories ($P < 0.05$).

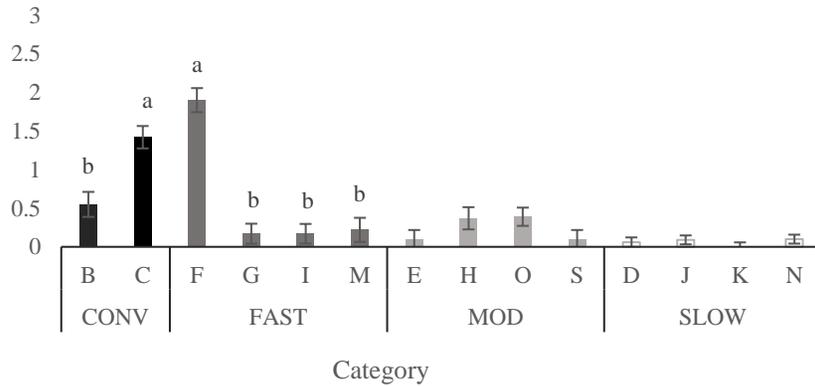
¹ Number of birds per category at Target Weight 1: CONV: $n = 104$; FAST: $n = 156$; MOD: $n = 175$; SLOW: $n = 173$.

² Number of birds per category at Target Weight 2 CONV: $n = 80$; FAST: $n = 176$; MOD: $n = 196$; SLOW: $n = 198$.

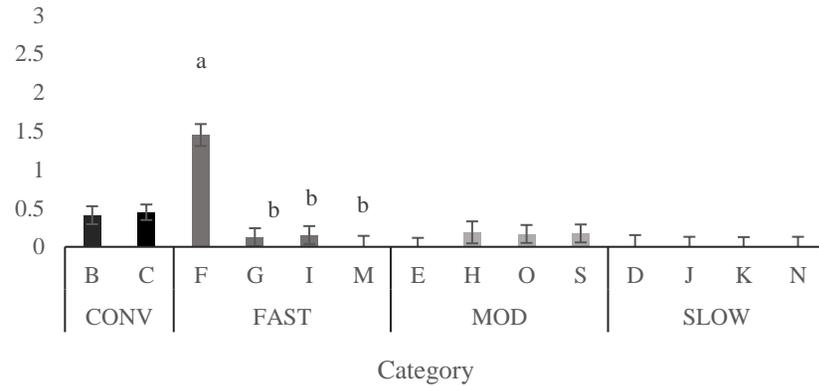
³ Number of birds per category at Target Weight 1: CONV: $n = 91$; FAST: $n = 124$; MOD: $n = 159$; SLOW: $n = 125$.

⁴ Number of birds per category at Target Weight 2: CONV: $n = 64$ FAST: $n = 132$; MOD: $n = 171$; SLOW: $n = 162$.

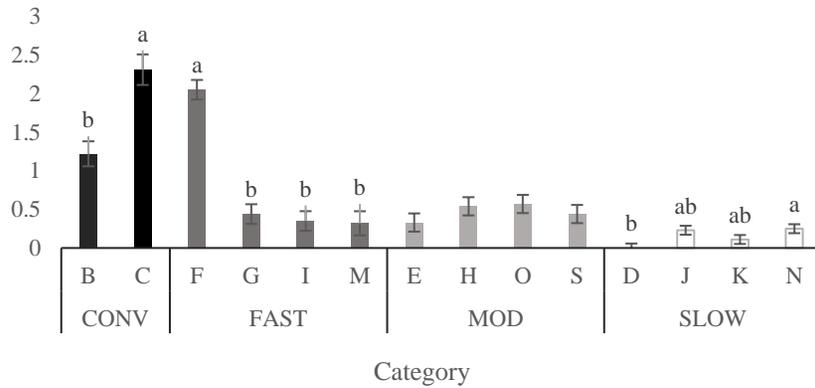
Average wooden breast score- Target weight 1¹



Average white striping breast score- Target weight 1³



Average wooden breast score- Target weight 2²



Average white striping score- Target weight 2⁴

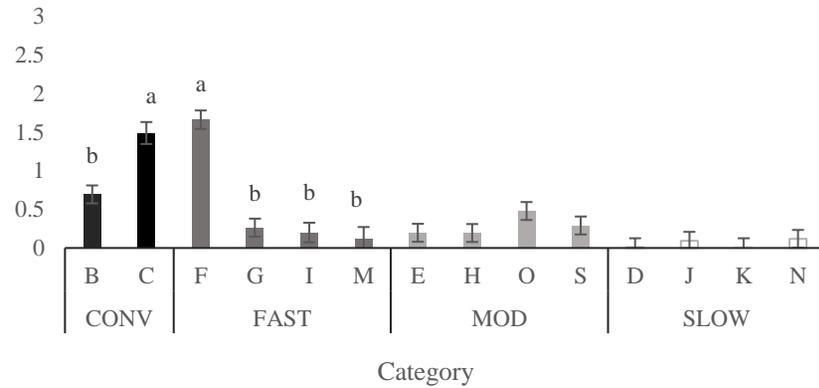


Figure 4.6: Effects of strains (within category) on average wooden breast^{1,2} and white striping^{3,4} scores at Target Weights 1 and 2 (LS-means \pm SEM). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

^{a-b} Different superscripts within the same category represent differences among strains ($P < 0.05$).

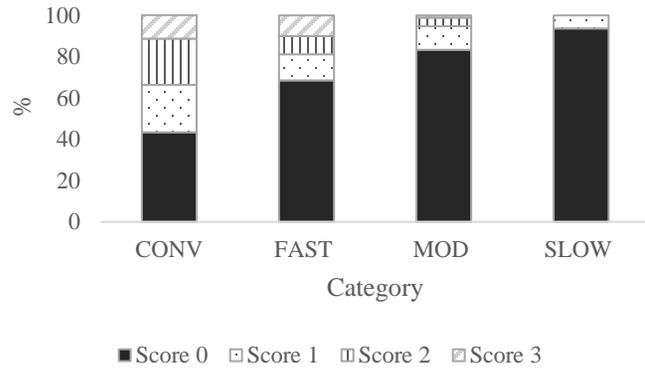
¹ Number of birds per strain at Target Weight 1: B: n = 52, C: n = 52, F: n = 30, G: n = 47, I: n = 47, M: n = 32, E: n = 48, H: n = 32, O: n = 47, S: n = 48, D: n = 32, J: n = 47, K: n = 47, N: n = 47.

² Number of birds per strain at Target Weight 2: B: n = 48, C: n = 32, F: n = 52, G: n = 48, I: n = 47, M: n = 29, E: n = 48, H: n = 52, O: n = 48, S: n = 48, D: n = 55, J: n = 48, K: n = 47, N: n = 48.

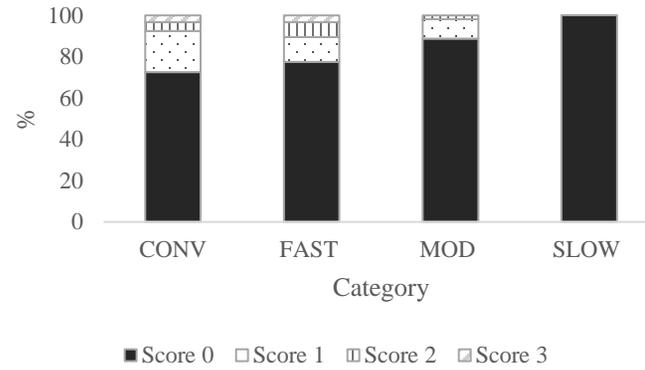
³ Number of birds per strain at Target Weight 1: B: n = 45, C: n = 46, F: n = 25, G: n = 39, I: n = 39, M: n = 21, E: n = 47, H: n = 24, O: n = 44, S: n = 44, D: n = 26, J: n = 28, K: n = 39, N: n = 32.

⁴ Number of birds per strain at Target Weight 2: B: n = 36, C: n = 28, F: n = 42, G: n = 41, I: n = 31, M: n = 18, E: n = 40, H: n = 48, O: n = 44, S: n = 39, D: n = 45, J: n = 37, K: n = 44, N: n = 36.

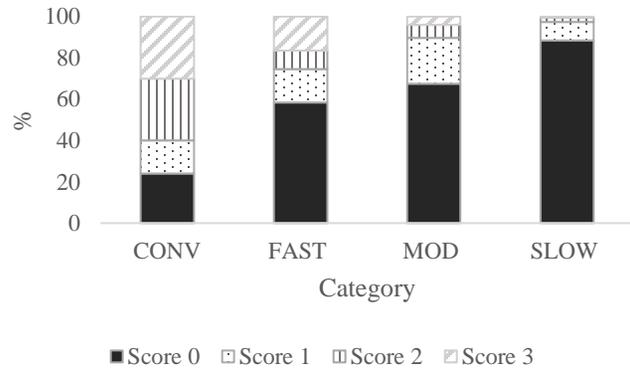
Wooden breast profile- Individual scores - Target weight 1¹



White striping profile- Individual scores- Target weight 1³



Wooden breast profile- Individual scores - Target weight 2²



White striping profile- Individual scores- Target weight 2⁴

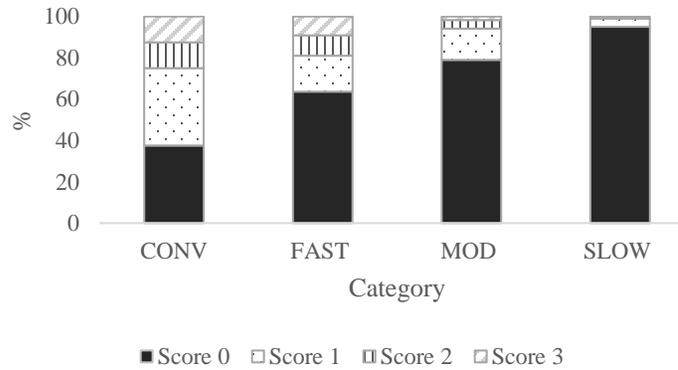


Figure 4.7: Wooden breast^{1,2} and white striping^{3,4} profile in different categories of broiler chickens at Target Weights 1 and 2. Only descriptive statistics are provided for this variable. At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

¹ Number of birds per category at Target Weight 1: CONV n = 104; FAST n = 156; MOD n = 175; SLOW n = 173.

² Number of birds per category at Target Weight 2: CONV n = 80; FAST n = 176; MOD n = 196; SLOW n = 198.

³ Number of birds per category at Target Weight 1: CONV n = 91; FAST n = 124; MOD n = 159; SLOW n = 125.

⁴ Number of birds per category at Target Weight 2: CONV n = 64; FAST n = 132; MOD n = 171; SLOW n = 162

Chapter 5: In Pursuit of a Better Broiler: Tibial Morphology, Breaking Strength, and Ash Content in Conventional and Slower-Growing Strains of Broiler Chickens²

5.1 Abstract

The rapid growth rate observed in fast-growing strains of broiler chickens has been linked to bone disorders that can cause lameness. Thus, the use of slow-growing (SG) strains has been suggested as an alternative for improved bone quality. To determine the effect of growth rate on bone traits, a population of birds from the study described in Chapter 4 were selected. From each pen, 4 birds (2 males and 2 females) were individually weighed and euthanized at 2 target weights (TWs) according to the time they reached 2.1 kg (TW 1: 34 d for CONV and 48 d for SG strains) and 3.2 kg (TW 2: 48 d for CONV and 62 d for SG strains). Tibia samples were dissected, and length and diameter recorded. Left tibiae were used for tibial breaking strength (TBS) at both TWs and tibial ash at TW 2. At TW 1, CONV birds' tibiae were narrowest and shortest ($P < 0.001$), yet had similar TBS compared to the other categories ($P > 0.69$). At TW 2, category ($P > 0.50$) had no effect on tibial diameter, yet CONV birds had the shortest tibiae ($P < 0.001$). The CONV birds had greater TBS:BW ratio than FAST and MOD birds at both TWs 1 and 2 ($P < 0.039$). The CONV birds had similar ash content as the other categories ($P > 0.220$). At 48 d of age, CONV birds had the greatest absolute TBS ($P < 0.003$), yet lower TBS:BW ratio than SLOW birds ($P < 0.001$). Tibiae from CONV birds were longer than MOD and SLOW birds, and thicker in diameter than the other categories, yet CONV birds had the lowest dimensions relative to BW ($P < 0.001$) at 48 d. These

² The abstract of this chapter has been presented to the Poultry Science Association Annual Meeting 2021. A version of this chapter will be submitted to Poultry Science.

results indicate a negative association between accelerated growth and tibial length. However, tibial mineral content and breaking strength were not negatively affected by fast growth, indicating that differences in functional abilities among categories may be due to differences in morphometric traits rather than differences in bone strength and mineralization. Future studies should include other indicators of bone quality at different ages to investigate possible differences in bone development among strains differing in growth rate.

Keywords: Chickens, slow-growth, bone health, genetic, growth rate, lameness

5.2 Introduction

Over the past 60 years, the growth rate of commercial breeds of broiler chickens has increased over 400%, resulting in more efficient birds that need less time to reach market weight (Zuidhof et al., 2014). It has been estimated that 85-90% of these improvements are attributed to genetic selection (Havenstein et al., 1994a, 2003a). However, this improved efficiency come at a cost, as fast and early growth has been linked to skeletal disorders, leg weakness, and impaired walking ability that can compromise the welfare of the birds (Julian, 1998; SCAHAW, 2000; Meluzzi and Sirri, 2009; Kierończyk et al., 2017).

The possible adverse effects of selection for growth and production traits on leg health and skeletal integrity have been mainly attributed to the structure of the bone (Lilburn, 1994; González-Cerón et al., 2015), suggesting that rapid muscle accretion is not fully accompanied by adequate bone development to support the heavy body weight (**BW**). The imbalance between accelerated growth and skeletal development may lead to a body mass and physical load that is too heavy to be properly supported by immature leg bones at a very early age (Yalcin et al., 2001; Bradshaw et al.,

2002; Caplen et al., 2014; González-Cerón et al., 2015; Kierończyk et al., 2017). This imbalance is likely to become greater every year, as the number of days for birds to reach a market weight of approximately 2 kg decreases while the age of skeletal maturity remains the same, at approximately 23 to 27 wk of age (Rath et al., 2000; Sherlock et al., 2010).

Research has shown that fast-growing (**FG**) broiler chickens have an accelerated and early increase in BW compared to a smaller increase in bone development (demonstrated by changes in length and diameter of the femur and tibia over a production cycle of 6 wk) (Applegate and Lilburn, 2002). In addition, FG strains of broiler chickens were reported to have a compromised ability to respond to mechanical load-bearing, suggesting a limited capacity of the skeletal system to adapt to the rapid changes in BW (Pitsillides et al., 1999; Angel, 2007). However, other studies have suggested that the dimensions of the tibiotarsus of FG strains of broiler chickens are appropriate to provide load support, but the bone itself is fragile, due to high porosity in the cortical bone and low mineral content (Corr et al., 2003b; Williams et al., 2004).

Poor bone quality has been associated with bone deformities, fragility, risk of fractures, and leg weakness (McDevitt et al., 2006; González-Cerón et al., 2015). Bone ash content is a well-validated method to assess bone mineralization (González-Cerón et al., 2015), whereas bone breaking strength is commonly measured to estimate fracture resistance and the force required to bend and break the bone (Kim et al., 2004). In poultry species, breaking strength has been found to be positively correlated with bone weight, ash content, and mineral quality (Shim et al., 2012b). These parameters, along with anatomical measurements (e.g., length, diameter, area, angulation) are commonly used to assess bone quality, development, and morphology (Shim et al., 2012b; Toscano et al., 2013; González-Cerón et al., 2015). However, there is some inconsistency in the

literature regarding the relationship between bone traits and walking ability in broiler chickens. While some researchers reported a link between some bone properties and walking ability (Toscano et al., 2013; Pedersen et al., 2020; Riber et al., 2021), other studies did not support such finding (Yalcin et al., 2001; Bizeray et al., 2002b; Venäläinen et al., 2006; Brickett et al., 2007; Talaty et al., 2010).

The possible negative impacts of selection for early and accelerated growth on bone health and walking ability may occur because the emphasis on productive traits results in less energy allocated to other metabolic processes (Tallentire et al., 2016). Furthermore, increased locomotor activity has been shown to improve walking ability and bone development (Reiter and Bessei, 1995; Pedersen et al., 2020). Therefore, the low activity levels consistently reported in FG birds (Bizeray et al., 2000; Bokkers and Koene, 2003; Dixon, 2020) may exacerbate the incidence of some leg disorders that cause lameness. However, in addition to the rapid increase in BW, other factors can induce skeletal problems in broiler chickens, including nutrition deficiencies, infectious diseases, mechanical trauma, and the interaction of these factors (Angel, 2007; Kierończyk et al., 2017). Furthermore, bone health and skeletal integrity have been included in breeding programs, resulting in a reduction of bone disorders that were commonly found in broiler chickens decades ago (Angel, 2007; Whitehead, 2007). Indeed, the percentage of birds with tibial dyschondroplasia (**TD**) and other growth plate deformities that can impact walking ability has decreased since the 1980s (Veltmann and Jensen, 1980, 1981; McKay et al., 2000). These improvements can be attributed to better nutrition, management practices, and development of genetic strategies incorporating leg health and robustness (McKay et al., 2000; Whitehead, 2007). However, despite these improvements, it has been estimated that 14-30% of broiler chickens raised worldwide have poor

gait score (Sanotra et al., 2003; Bassler et al., 2013; Kittelsen et al., 2017; Vasdal et al., 2018), which indicates that impaired walking ability is still an ongoing problem in broiler production.

Because there is a lack of fully effective strategies to improve leg health without influencing growth, there is an increasing interest in the use of slower-growing (**SG**) strains to decrease skeletal abnormalities and improve the walking ability, and welfare of broiler chickens (Bessei, 2006; Shim et al., 2012b; Dixon, 2020). Although comparisons between a few strains of FG and SG broilers have been performed (Bokkers and Koene, 2003; Dixon, 2020; Mancinelli et al., 2020), there is a scarcity of studies that investigate different strains of broiler chickens raised under similar conditions and tested at a similar BW. In addition, although the term FG commonly refers to conventional broiler chickens that are intensively selected for meat production and reach market weight at an early age (about 2.5 kg in 40 d; $ADG \geq 60$ g/d), the term "slow-growing" encompasses a wide range of growth rates, representing a heterogenous group of birds, commonly raised in alternative production systems (Doğan et al., 2019; Mancinelli et al., 2020). In this context, bone quality and morphologic traits were investigated in 14 strains of broiler chickens (2 FG and 12 SG), encompassing a wide range of growth rates. It was hypothesized that SG strains of broiler chickens would have better bone quality than FG strains, indicated by greater bone breaking strength and ash content.

5.3 Materials and Methods

5.3.1 Hatching and Husbandry

The procedures carried in this study were reviewed and approved by the University of Guelph's Animal Care Committee (AUP #3746) and were in accordance with the Canadian Council for Animal Care's guidelines (CCAC, 2009).

This study is part of a multidisciplinary project conducted to assess production performance, meat quality, behaviour, physiology, leg health, inactivity, and welfare of FG and SG strains selected for distinct growth rates, described in other associated papers. The complete details regarding incubation conditions, animal handling, husbandry, management, and housing are available elsewhere (Torrey et al., 2021). The overview of the methodology used in this chapter and other associated studies that encompass the multidisciplinary project is described in Chapter 3.

5.3.2 Tibial Morphology Parameters

A total of 4 wing-tagged focal birds (2 males and 2 females) from each pen were individually weighed and labeled for identification purposes the day before processing, at either TW 1 or TW 2. These birds were selected previously to determine birds' mobility as measured by latency-to-leave and group obstacle tests as described in Chapter 6. The group weight from each pen to be processed was obtained to determine the final BW and production performance of each strain (Torrey et al., 2021). Feed was removed from each pen the night before processing at 23:00. Birds had free access to water until loading. The next morning, the 4 focal birds were killed by cervical dislocation, left to cool at 4°C for a maximum of 6 h, and then kept in a freezer at -20°C until analyses. Prior to dissections, carcasses were transferred to a cooler room at 4°C and thawed for 48 to 72 h depending on body size to facilitate dissections and separation of tissues. Dissections were performed by 3 trained researchers to keep measurements consistent across strains.

Both left and right tibiae were dissected and completely defleshed to remove adherent soft tissues. The length of each tibia was measured from the lateral intercondylar tubercle to the inferior articular surface and the diameter was measured at the midpoint of the diaphysis using a digital caliper (Fisher Scientific carbon fiber composite digital calipers; Toronto, ON, Canada;

Resolution: 0.1 mm, Accuracy: ± 0.2 mm). The mean length and diameter from both tibiae were measured and used as a single value for each variable.

5.3.3 Tibial Breaking Strength

After morphometric measurements, the bone samples were placed in labeled plastic bags and stored in a freezer at -20°C . The left tibiae from the focal birds were thawed for 24 h in a cooler at 4°C prior to breaking. To measure tibial breaking strength (**TBS**), a 3-point bending Instron material tester with Bluehill Universal software (Model Material Testing, Norwood, MA) was used. Each bone sample was placed in the same orientation and held by cradle support with a span of 5 cm. A 5kN load cell at a speed of 20 mm/s was applied at the midpoint of the bone, with a fixed distance of 50 mm between upper and lower anvils. The maximum force required to break the bone was detected from the deformation curve and was used to determine the breaking strength in Newtons (**N**). The absolute values along with TBS expressed relative to the BW were determined.

5.3.4 Tibial Dyschondroplasia and Tibial Composition

Following breaking strength (see above), the left tibiae from the focal birds were stored in a freezer at -20°C until subject to the determination of TD and tibial composition. Due to time constraints and logistics, only bones from birds killed at TW 2 were used for these analyses. The tibia from each bird was thawed for 1 h at room temperature. The thawed weight was recorded using an analytical scale (Mettler AC 88 digital balance; Mississauga, ON, Canada; Accuracy: 0.0001 g). The proximal end of the left tibia was cut longitudinally to determine the presence of TD as described by Shim et al., 2012a (Figure 5.1) and all the pieces were kept for the determination of tibial composition.

Subsequently, each tibia was placed in hexane for 2 d for fat extraction (Kiarie et al., 2015) and later the defatted tibiae were transferred to pre-weighed crucibles and dried in an oven for 24 h at 105°C. Tibial dry matter weight (the remaining content after removal of fat and moisture) was obtained using an analytical scale (Mettler AC 88 digital balance). Next, crucibles and dry tibiae were transferred to a muffle furnace for 12 h at 600°C as described by Khanal et al. (2019). The samples were placed in a desiccator until they reached room temperature and the final weight was recorded to determine ash content relative to the dry weight of the tibia. The total organic matter content of the tibia was determined by subtracting ash content from the dry matter content. The organic matter weight was divided by the ash weight to estimate the ratio of organic to inorganic matter and both ash and organic matter were expressed as absolute values, while ash weight was also expressed relative to the tibial length as described by McDevitt et al. (2006). To provide a quantitative assessment of tibial weights considering the differences in BW, tibial dry weight and ash content were also expressed relative to the BW (Shim et al., 2012b; Guo et al., 2019).

5.3.5 Statistical Analyses

To facilitate analyses, strains were grouped into 4 categories based on their similar growth rates to TW 2 (48 d for FG and 62 d for SG strains, respectively) (Chapter 3). The strains were categorized as conventional (CONV; strains B and C; ADG = 66.0 to 68.7 g/d), fastest slow-growing (FAST; strains F, G, I, and M; $ADG_{0-62} = 53.5$ to 55.5 g/d), moderate slow-growing (MOD; strains E, H, O, and S; $ADG_{0-62} = 50.2$ to 51.2 g/d) and slowest slow-growing (SLOW; strains D, J, K, and N; $ADG_{0-62} = 43.6$ to 47.7 g/d). Comparisons between categories and within categories were assessed for each dependent variable. Comparisons between categories were conducted to assess

differences at different growth rates, whereas comparisons within categories were conducted to assess differences among strains at a similar growth rate.

Data were analyzed as an incomplete block design using Generalized linear mixed model (GLIMMIX) in SAS[®], version 9.4 (SAS Institute Inc., Cary, NC), with pen as the experimental unit. Different models were used to evaluate the effects of TW and age on a number of dependent variables. The *TW model* was used to assess tibial morphology (length and diameter) and TBS when the birds were evaluated at a similar BW. In these models, category, strain nested within category, sex, TW, and their interactions were included as main effects. The interactions in the TW models included category \times TW, category \times sex, category \times sex \times TW, strain (category) \times TW, strain (category) \times sex, strain (category) \times sex \times TW. These models allowed comparisons at the two TWs to investigate the effects of BW on tibial characteristics for both FG and SG strains. The *age model* was used to assess differences in tibial morphology traits when both FG and SG birds were 48 d (TW 1 and TW 2 for SG and FG birds, respectively). In these models, category, strain nested within category, and sex were included as main effects. The interactions between category \times sex, and strain (category) \times sex were kept in the model if significant. Because tibial dry matter, ash, and organic content were only evaluated in birds processed at TW 2, another model was used, named *TW 2 model*, which omitted the TW and its interactions. In these models, category, strain (category), and sex were included as main effects, with the inclusion of the interactions between category \times sex and strain (category) \times sex, which were kept in the model if significant. The random effects for all models included trial (i.e., production cycle) and block nested within the trial.

Differences between categories and among strains within each category were compared using contrast statements. For all models, the residuals were checked for normality using a quantile-quantile plot and Shapiro-Wilk test. Linearity, randomness, and homogeneity of residuals were assessed using scatterplots and boxplots of studentized residuals. Residual analysis was used to select the most appropriate distribution that met all of the model assumptions, with Gaussian distribution used by default for those variables that met all the assumptions. For BW, tibial dimensions traits (absolute and relative to the BW), and TBS (absolute and relative to the BW), a lognormal distribution was required to meet the model assumptions. Differences were considered significant at adjusted $P < 0.05$. Pairwise comparisons were corrected using Tukey adjustment to explore multiple comparisons and differences between categories, strains, and sex. Differences between sex or TW are not provided in data tables, though they are described in the Results section with their respective P-value, if significant.

5.4 Results

Differences in tibial parameters at both TWs are described as differences between categories of strains (Table 5.1) and between strains within the same category (Table 5.2 - 5.5). As expected, category affected most of the variables evaluated at both TWs. For variables measured at the same age (i.e., BW, tibial breaking strength, length, and diameter), only differences among categories are provided in Table 5.6, as birds from strains within the same category were evaluated at the same age. Significant interactions between category, TW, and sex are not presented here but are included in Appendix A.1. Differences between TWs and sexes are described in the text if significant.

5.4.1 Body Weight and Leg Traits at Target Weight 1 and 2

5.4.1.1 BW

The BW was affected by category, TW, strain, and sex ($P < 0.001$). However, category interacted with TW ($P < 0.001$), indicating that the effect of category on BW depended on the TW (Table 5.1). At TW 1, a significant difference was observed among the categories with CONV birds having the lowest BW, followed by SLOW, MOD, and FAST birds. However, at TW 2, SLOW birds had the lightest BW, while CONV birds were similar to FAST and MOD birds, with FAST being heavier than MOD birds. Despite the effect of strains on BW, strains within the same category had similar BW at both TWs ($P > 0.091$ for all combinations among strains within the same category at both TWs). As expected, males were heavier than females ($2,901 \pm 19.2$ g vs. $2,376 \pm 16.7$ g). The 3-way interactions of sex by category by TW ($P = 0.065$) or sex by strain nested within category by TW ($P = 0.641$) were not significant.

5.4.1.2 Tibial Breaking Strength

Overall, there was an increase in absolute TBS from TW 1 to TW 2 ($P < 0.001$; 286.9 ± 4.70 N vs. 334.9 ± 4.79 N). However, differences in TBS among categories varied according to the TW. At TW 1, category did not affect TBS, whereas at TW 2, CONV birds had greater TBS than MOD and SLOW birds (Table 5.1; $P < 0.022$). Within categories, TBS did not significantly differ among strains ($P > 0.05$ for all pairwise comparisons between strains within category at TW 1 and TW 2). Sex influenced TBS, with males being greater than females ($P < 0.001$; 353.8 ± 4.87 N vs. 271.7 ± 3.96 N), with no interaction between sex and category or sex and strain ($P > 0.083$).

5.4.1.3 Tibial Diameter

Tibial diameter differed among TWs, categories, strain, and sex ($P < 0.001$). Overall, tibiae became wider in diameter from TW 1 to TW 2 (8.06 ± 0.061 mm vs. 9.23 ± 0.061 mm). However, a TW by category interaction was found (Table 5.1, $P < 0.001$). At TW 1, CONV birds had the smallest tibial diameter while at TW 2, no difference in tibial diameter was observed among the categories. Despite the significant differences among strains ($P < 0.001$), strain within category did not influence tibial diameter ($P > 0.068$ for all combinations among strains within category at both TWs). Males had wider tibial diameter than females ($P < 0.001$; 9.31 ± 0.054 mm vs. 7.98 ± 0.048 mm), with no significant interaction between category or strain with sex and TW ($P > 0.204$).

5.4.1.4 Tibial Length

Tibial length was affected by TW, category, strain, and sex ($P < 0.001$). Similar to diameter, tibial length increased from TW 1 to TW 2 ($P < 0.001$; 108.7 ± 0.319 mm vs. 125.6 ± 0.321 mm). However, there was an interaction between category and TW (Table 5.1; $P < 0.001$). At TW 1, CONV birds had shorter tibia than other categories, while FAST had longer tibiae than MOD and SLOW birds. At TW 2, CONV birds still had the shortest tibial length, while FAST birds were similar to MOD and longer than SLOW birds. Within categories, CONV and FAST birds had different tibial lengths at TW 1 and TW 2, respectively. Among CONV strains, at TW 1, birds from strain B had longer tibia than strain C (Table 5.2), whereas among FAST strains, at TW 2, strain I had longer tibia than strain F (Table 5.3). Overall, males had longer tibia than females ($P < 0.001$; 120.4 ± 0.297 mm vs. 113.4 ± 0.291 mm). However, at TW 1, sex did not influence tibial length for CONV birds, whereas at TW 2, sex influenced tibial length in all categories, with males having longer tibiae than females (category \times TW \times sex interaction; $P = 0.009$, Appendix A.2).

5.4.1.5 Relative Tibial Breaking Strength

Overall, relative TBS decreased as the birds grew ($P < 0.001$; TW 1 = 131.8 ± 2.09 N/kg, TW 2 = 105.4 ± 1.46 N/kg). Category affected relative TBS per unit of BW at both TWs (Table 5.1; $P = 0.038$). At TW 1, CONV birds had the highest relative TBS, while FAST and MOD birds were similar and lower than SLOW birds. At TW 2, CONV and SLOW birds had similar relative TBS that was higher than FAST and MOD birds. At TW 1, no difference among strains within category was observed. At TW 2 among MOD strains, strain H had higher relative TBS than strain O (Table 5.4; $P = 0.013$) while among SLOW strains, strain D had higher relative TBS than J (Table 5.5; $P = 0.006$). Males had higher relative TBS than females ($P < 0.001$; 121.5 ± 1.61 N/kg, *vs.* 114.4 ± 1.61 N/kg), with no interaction between sex, category, and TW or sex, strain, and TW ($P > 0.170$).

5.4.1.6 Relative Tibial Diameter

Birds had greater relative tibial diameter at TW 1 than those at TW 2 ($P < 0.001$; 3.69 ± 0.029 mm/kg *vs.* 2.91 ± 0.019 mm/kg). However, there was a category by TW interaction that affected tibial diameter per unit of BW (Table 5.1; $P < 0.001$). At TW 1, CONV and SLOW birds had similar relative tibial diameter, which was greater than FAST and MOD birds. At TW 2, CONV, FAST, and MOD birds had lower relative tibial diameter than SLOW birds, while FAST was similar to CONV, yet lower than MOD birds. There was no effect of strain within category ($P > 0.110$). Overall, males had lower relative tibial diameter than females ($P < 0.001$; 3.19 ± 0.020 mm/kg *vs.* 3.36 ± 0.022 mm/kg). The interaction between sex, category, and TW was not significant ($P = 0.682$).

5.4.1.7 Relative Tibial Length

Overall, relative tibial length was greater in TW 1 compared to TW 2 ($P < 0.001$; 49.84 ± 0.352 mm/kg vs. 39.53 ± 0.241 mm/kg). However, TW interacted with category (Table 5.1; $P < 0.001$). At TW 1, SLOW birds had the greatest length relative to the BW, while CONV birds were greater than FAST and MOD birds. At TW 2, relative length decreased as the growth rate increased among categories (CONV < FAST < MOD < SLOW). Strain within category did not affect relative tibial length ($P > 0.207$). Males had lower relative tibial length than females ($P < 0.001$; 41.29 ± 0.229 mm/kg vs. 47.73 ± 0.276 mm/kg). No significant interaction was observed between sex, category, and TW ($P = 0.576$).

5.4.1.8 Ratio of Tibial Length to Diameter

Length: diameter was not affected by TW ($P = 0.172$). However, the ratio of length: diameter was affected by category, strain, and sex ($P < 0.002$). At TW 1, category did not affect length: diameter, whereas at TW 2, CONV birds had a lower ratio than the other categories (Table 5.1; $P < 0.001$). Within categories, FAST and SLOW strains differed in length: diameter at TW 1. Among the FAST strains, strain F had lower length: diameter than strains G ($P = 0.049$) and M ($P = 0.009$) (Table 5.3), while among SLOW strains, strain D had higher ratio than strain N ($P = 0.046$; Table 5.5). However, these differences were not found at TW 2. Overall, males had lower tibial length: diameter than females ($P < 0.001$; 12.92 ± 0.067 mm/kg vs. 14.18 ± 0.076 mm/kg). There was no interaction between sex, category, and TW ($P = 0.834$).

5.4.2 Tibial Content- Dry Matter, Organic, and Inorganic Content at TW 2

Tibial dry matter weight differed by category and sex (Table 5.1; $P < 0.001$). The CONV birds had the lightest tibial dry matter, which was similar to SLOW and lighter than both FAST and

MOD birds. Strain within categories did not differ in tibial dry matter weight. Overall, males had heavier tibial dry matter than females ($P < 0.001$; 12.35 ± 0.104 g vs. 9.07 ± 0.118 g). However, there was an interaction between sex and category ($P < 0.001$, Appendix A.1). Dry matter weight was not affected by category among females, whereas among males, FAST birds had heavier dry matter weight than the other categories, and MOD had heavier weight than CONV, yet similar to SLOW birds.

Both tibial ash weight and the weight of organic content followed a similar pattern observed for tibial dry matter, with CONV being similar to SLOW birds yet lighter than both FAST and MOD birds (Table 5.1). There was no effect of strain within category on tibial ash weight ($P = 0.106$). While males had heavier tibial ash than females ($P < 0.001$; 4.94 ± 0.046 g vs. 3.57 ± 0.053 g), an interaction between sex and category affected tibial ash weight ($P < 0.001$, Appendix A.1). Similar to the interaction observed for tibial dry matter weight, no difference in tibial ash weight was observed among categories for females, while for males there was an effect of category, with CONV birds being lower than the other categories. Males had heavier organic matter weight than females ($P < 0.001$; 7.40 ± 0.066 g vs. 5.49 ± 0.076 g), with an interaction between sex and category ($P = 0.009$) that followed a similar pattern to those observed for tibial dry matter and ash weight (Appendix A.1).

Category, strain, and sex affected tibial dry matter and ash weight relative to the BW (Table 5.1; $P < 0.001$). Among categories, CONV birds had the lowest and SLOW birds had the highest values, while FAST and MOD showed intermediate values. Within categories, only FAST strains differed in dry matter weight relative to BW (Table 5.3; $P < 0.001$), with strain F being lower than strain M ($P = 0.041$). For tibial ash weight relative to the BW, significant differences were detected

in FAST and MOD strains. Among FAST strains (Table 5.3), strain F had lower tibial ash weight per unit of BW than strains I ($P = 0.006$) and M ($P = 0.001$). Among MOD birds (Table 5.4), strain E had higher tibial ash weight relative to BW than strain O ($P = 0.041$), while strains H and S were similar and did not differ from strains E and O. Males had greater tibial dry matter relative to the BW than females ($P < 0.001$; 3.49 ± 0.026 g/kg *vs.* 3.18 ± 0.029 g/kg,) and there was no interaction between sex and category or sex and strain ($P > 0.200$). Overall, males had higher tibial ash weight per unit of BW than females ($P < 0.001$; 1.39 ± 0.010 g/kg *vs.* 1.25 ± 0.012 g/kg). However, sex interacted with category ($P = 0.033$; Appendix A.1); there was an effect of sex in all the categories except CONV, in which females and males had similar tibial ash weight relative to BW.

Tibial ash content did not differ among categories ($P = 0.354$), but it was affected by strain ($P < 0.001$) and sex ($P = 0.038$). Within categories, only FAST strains differed in tibial ash content (Table 5.3). Strain F had lower tibial ash content than strain I ($P < 0.001$), yet similar to the other FAST strains ($P > 0.554$). Males had greater tibial ash content than females ($40.08 \pm 0.209\%$ *vs.* $39.43 \pm 0.241\%$) and no interaction between sex and category or sex and strain was found ($P > 0.620$).

While category had no effect on organic matter content and organic: inorganic matter ($P > 0.354$), these variables were affected by strain and sex ($P < 0.037$). In the FAST category (Table 5.3), strain F had greater organic matter content ($P < 0.001$) and organic: inorganic ($P = 0.001$) than strain I, while strains G and M did not differ from strains F and I. Males had lower tibial organic content ($P = 0.038$; $59.91 \pm 0.209\%$ *vs.* $60.56 \pm 0.241\%$) and organic relative to inorganic matter ($P = 0.037$; 1.50 ± 0.014 *vs.* 1.55 ± 0.016) than females. There was no interaction between sex and

category or sex and strain for both tibial organic matter content and organic relative to inorganic matter ($P > 0.583$).

The amount of ash per unit of length of the tibial [ash: length (g/mm)] was influenced by category and sex ($P < 0.001$). Among categories, CONV birds did not differ from the remaining categories, while FAST was greater than MOD and SLOW birds (Table 5.1). Overall, males had higher tibial ash: length than females ($P < 0.001$; 0.379 ± 0.003 g/mm vs. 0.295 ± 0.004 g/mm). However, among females, category did not affect tibial ash: length, whereas among males FAST was greater than CONV and SLOW males, resulting in an interaction between category and sex ($P = 0.012$; Appendix A.1).

5.4.3 Tibial Dyschondroplasia

Due to the low number of birds affected by TD, statistical analyses were not performed. Therefore, only descriptive statistics are included for this trait. Based on macroscopic examination of the growth plate of the focal birds, the overall incidence of TD was 2.58%, with FAST and MOD birds accounting for 77.78 % of the TD observed in our study. While TD was not found in CONV birds, FAST, MOD, and SLOW birds exhibited incidences of TD at 1.09, 6.06 and 1.96%, respectively. The MOD birds had a TD incidence over twice as high as the overall incidence of TD with strains O and S exhibiting incidences of 8.70 and 17.39%, respectively. The other strains that had TD were: FAST, strain I (4.35%) and SLOW, strains J (4.00%) and K (4.17%). The other strains had no evidence of TD in any samples. All the birds affected showed mild TD lesions.

5.4.4 Body Weight and Leg Traits at a Similar Age

Differences in BW and leg traits obtained at the same age (48 d) among categories are provided in Table 5.6. Category ($P < 0.006$) and sex ($P < 0.034$) affected all variables evaluated at the same age. Because the interactions between sex and category were not significant for any trait ($P > 0.05$), only the main effects of category and sex are presented.

As expected, BW was greater as the growth rate was higher across among categories ($P < 0.001$; CONV > FAST > MOD > SLOW) and males were heavier than females ($P < 0.001$; $2,754 \pm 24.1$ g, vs. $2,267 \pm 20.5$ g). At 48 d of age, CONV birds had the highest TBS among the categories ($P < 0.002$), whereas FAST was similar to MOD birds yet greater than SLOW birds. Sex had an effect within all categories, with males exhibiting greater TBS than females ($P < 0.001$; 344.6 ± 7.06 N vs. 269.0 ± 5.81 N). When TBS was expressed per unit of BW, CONV was lower than SLOW ($P < 0.001$) birds, while FAST and MOD birds were similar and not different from CONV and SLOW birds. In all categories, males had higher TBS: BW than females ($P = 0.034$; 124.9 ± 2.43 N/kg vs. 118.4 ± 2.42 N/kg.).

Among categories, CONV birds had wider tibia than the other categories ($P < 0.001$), while FAST birds were greater than SLOW ($P = 0.036$) and similar to MOD birds ($P = 0.321$), which did not differ from SLOW birds ($P = 0.728$). Males had wider tibia than females ($P < 0.001$; 9.15 ± 0.073 mm vs. 7.96 ± 0.066 mm.).

Tibial length differed by category ($P < 0.001$), with CONV and FAST birds being similar and longer than MOD and SLOW birds ($P < 0.001$), which did not differ from each other. As expected, males had longer tibia than females ($P < 0.001$; 117.4 ± 0.379 mm vs. 111.3 ± 0.373 mm).

Tibial diameter and length per unit of BW (mm/kg) followed a similar pattern, with CONV birds being lower than the other categories ($P < 0.001$) and FAST and MOD birds being similar, yet lower than SLOW birds ($P < 0.001$). Males had lower relative tibial diameter ($P < 0.001$; 3.32 ± 0.026 mm/kg vs. 3.50 ± 0.029 mm/kg) and relative length ($P < 0.001$; 42.43 ± 0.308 mm/kg vs. 49.00 ± 0.370 mm/kg) than females, which was consistent in all categories.

The CONV birds had the lowest tibial length: diameter compared to the other categories ($P < 0.031$). Males had lower tibial length relative to diameter than females ($P < 0.001$; 12.80 ± 0.091 vs. 13.97 ± 0.103), which was observed in all categories.

5.5 Discussion

The link between selection for accelerated growth and susceptibility to leg disorders in broiler chickens has been well-documented (Julian, 1998; Shim et al., 2012b; Williams et al., 2004; Dixon, 2021). Since reducing the growth rate decreases leg disorders to some extent, it has been suggested that the use of SG strains may decrease leg abnormalities that cause both welfare and economic issues in the poultry industry (Julian, 1998; Bessei, 2006; Shim et al., 2012b). However, SG strains encompass a wide range of growth rates and they are often reared in alternative production systems that do not represent the conditions commonly found in commercial broiler production. Therefore, the aim of this study was to investigate the differences in tibial morphology, breaking strength, and composition (inorganic and organic content) as indicators of bone quality and bone measurements in 16 strains of broiler chickens, representing FG and SG strains with distinct growth rates and raised under similar conditions. Strains A and T were not analyzed due to the reduced sample size evaluated throughout the study. Therefore, only descriptive statistics are provided for these two strains (see Appendix A.1). The remaining 14 strains were separated

into 4 categories (CONV, FAST, MOD, SLOW) based on the similarity of growth rate to reach TW 2. Category affected most of the variables evaluated at TW 1 and TW 2, whereas only a few differences were found between strains within category, suggesting similar selection criteria for bone quality and morphology in strains selected for similar growth rates. While some of the differences were only observed when the birds were evaluated at a similar BW, other differences were also present when the birds were evaluated at a similar age, which indicates that both growth rate and age may contribute to the changes in the leg traits measured in our study.

5.5.1 Effect of Category

5.5.1.1 BW

Even though skeletal disorders are multifactorial conditions, growth rate and BW are among the factors considered to play a crucial role in the incidence of leg abnormalities (Julian, 1998; Bessei, 2006; Shim et al., 2012b). In addition, bone development and morphology are affected by age (Lilburn, 1994; Talaty et al., 2009). To account for the effect of growth rate, BW, and age, FG and SG strains of broiler chickens representing a wide range of growth rates were evaluated at a similar TW and age. However, because the ADG ranged from 43.6 to 68.7 g/d, differences in BW were observed among the categories. When possible, traits were adjusted relative to BW to account for the differences.

5.5.1.2 Tibial Breaking Strength

Bone breaking strength is a measure of the toughness and capacity of the bone to endure stress and resist fracture (Rath et al., 2000). This variable is affected by different properties, including shape, size, mineral and organic matrices, and collagen crosslinks (Turner, 2006; Foutz et al., 2007; Rath et al., 2000). Bone breaking strength has been shown to be correlated with cortical bone thickness,

a crucial indicator of bone development and quality (Dibner et al., 2007). Low bone strength increases the risk of fractures during rearing, catching, transport, unloading, and stunning, which can contribute to higher mortality, culling, and carcass condemnations (Onyango et al., 2003; Sun et al., 2018).

At TW 1, TBS was not affected by category while at TW 2, CONV birds were similar to FAST birds, yet greater than MOD and SLOW birds. When the values were expressed relative to the BW, at TW 1, CONV birds had higher TBS than the other categories, while at TW 2, CONV birds exhibited similar relative TBS compared to SLOW birds, yet greater than FAST and MOD birds. These findings are in line with the results of McDevitt et al. (2006), who reported similar absolute bone breaking strength between FG and SG birds evaluated at a similar BW, indicating that bones from FG birds were as strong as or stronger than those of SG birds at the same TW. The greater relative TBS values found in CONV compared to MOD and FAST birds may be a result of more balanced selection criteria practiced by breeding companies in the past 25 years for FG birds, which has incorporated not only growth performance traits but also skeletal integrity (Whitehead, 2007; Kapell et al., 2012). Despite the negative association between leg health (e.g., walking ability, bone deformities, leg disorders, and bone quality) and growth rate, a previous study demonstrated an increase in leg strength coupled with an increase in growth rate over time, suggesting the simultaneous selection of both traits as part of a more balanced breeding goal in FG strains (Neeteson-van Nieuwenhoven et al., 2013).

The greater relative TBS values observed in SLOW birds compared to MOD and FAST birds is likely due to the improved bone strength and quality associated with slower-growth as demonstrated in other studies (Williams et al., 2004; Shim et al., 2012b), since the SG birds

evaluated in our study encompassed a wide range of growth rates, with FAST and MOD birds classified as SG birds but showing ADG greater than those observed in SLOW birds. Another explanation for the greater relative TBS of SLOW birds is the differences in BW observed among the categories, rather than differences in TBS *per se*. Despite the original plan to process the birds at a similar BW, SLOW birds were not able to reach the same BW as the other categories at TW 2 due to their slower growth rate. Thus, the comparisons between SLOW birds and other categories at both TWs may not accurately represent the differences at the same BW. In fact, SLOW birds had the lowest absolute TBS at both TWs and at the same age compared to other categories.

While BW significantly increased in all strains from TW 1 to TW 2, the changes in TBS as the birds grew did not follow the same trend in some strains, showing a small change that did not match the slope of the increase in BW. It has been suggested that the rapid growth and increase in muscle accretion observed in FG birds are not accompanied by a sufficient increase in leg strength capable of supporting their heavy BW (Kierończyk et al., 2017). However, the small increase in TBS as the birds grew were also observed in SG birds. These findings suggest that expressing TBS values relative to the BW might not accurately represent the differences in TBS among the categories due to the considerable differences in BW observed. A study by Wilson et al. (1990) suggested that BW could not be used to predict bone strength. Some studies comparing FG and SG birds have shown greater bone-breaking strength (corrected for BW) in the latter group, which differs from the results found in our study (Williams et al., 2004; Shim et al., 2012b). The birds in our study were evaluated at a similar TW in order to account for possible effects of BW on bone strength. While CONV birds were lighter than the other categories at TW 1 due their earlier processing age, CONV birds were similar to FAST and MOD birds, yet heavier than SLOW at

TW 2. On the other hand, some studies comparing birds differing in growth rate evaluated bone breaking strength when FG was heavier than SG birds. Due to the possible limitations of correcting TBS to BW, it is unknown if such corrections can precisely capture differences in bone breaking strength as a function of the BW, especially when evaluating strains differing in growth rates and potentially bone development and growth as demonstrated in this study. When a FG (BW at 35 d = 2,201 g) and a SG (BW at d 49 = 1,767 g) strain were evaluated two weeks apart, Mussini (2012) reported greater bone strength (kg/mm^2) in FG birds, despite the greater tibial diameter observed in SG birds. The author suggested that selection for growth rate and rapid muscle deposition has led to changes in bone structure, with the increase in TBS being considered a side effect of selection.

Age of the birds is known to affect bone strength, as the BW and absolute bone mass increase as the birds grow, with the latter being proportional to bone strength (Frost, 1997). Rath et al. (2000) demonstrated changes in TBS and ash content over a production cycle of broiler chickens, measured as a percentage of change relative to day 1. The researchers reported a continuous increase in both variables that peaked between 3 to 5 wk of age. The changes in bone strength as the birds age may be a result of modifications in collagen crosslink content, making the bones tougher and less brittle (Rath et al., 2000). Because FG and SG birds were processed two weeks apart at both TWs, age differences may have influenced the differences in relative TBS among the categories observed in our study. In fact, when evaluated at the same age, CONV birds exhibited similar relative TBS to FAST and MOD, yet lower than SLOW strains. However, when BW was not considered, CONV birds had the greatest TBS at the same age. Similarly, McDevitt et al. (2006) reported greater absolute bone breaking strength in FG compared to SG birds at the same

age. This indicates that the incorporation of bone health into breeding programs has been successful at improving tibia breaking strength in FG birds, suggesting that issues related to lameness are most likely not related to tibia strength. However, because lameness is a multifactorial disorder that may be triggered by disturbances in different tissues and bones (Bradshaw et al., 2002), future studies should also investigate the bone status of other pelvic limb bones, such as femur and tarsometatarsus due to their contribution to walking ability (Paxton et al., 2014). In addition, other indicators of bone quality such as bone mineral density and bone stiffness (Rath et al., 2000), should be studied to provide a better understanding of the differences in bone health and development among strains differing in genetic potential for growth.

It is worth mentioning that the methodology used in our study to determine tibia breaking strength differed from previous studies regarding freezing procedures and loading rate. Due to logistics of the project, the samples were frozen and thawed before the determination of breaking strength. Previous studies have demonstrated that bone strength (Wilson et al., 1990a) and ash content (Park et al., 2003) are not affected by freezing and thawing. In addition, because all the bones samples were submitted to the same procedure before analyses were conducted, the results were likely not affected by the methods used in our study. Although the loading rate used in our study (20 mm/s) has been previously reported (Rath et al., 1999), more recent studies conducted in poultry species have adopted a much slower loading rate, commonly ranging from 5-50 mm/min (Shim et al., 2012b; Park et al., 2003; Whitehead et al., 2004; Candelotto et al., 2020). In fact, Crenshaw et al.(1981) reported that differences in procedures used to analyze breaking strength in swine led to variation in values found for bone breaking strength. The authors suggested that a loading speed of 5 mm/min should be used to evaluate bone breaking strength when a 3-point bending test is

used due to the effects of loading rate on bone mechanical properties. Similarly, a loading speed at a rate of 30-60 mm/min has been suggested for evaluating bone breaking strength in mice (Jepsen et al., 2015). It was also emphasized that a lower loading speed rate ranging from 3-6 mm/min may be more accurate to detect some differences in bone mechanical properties such as post yield displacement. We compared the TBS for broilers at two loading rates (20 mm/s and 50 mm/min) and found no difference (data in Appendix A.1), suggesting that the high loading speed used in our study did not significantly influence the results.

5.5.1.3 Tibial Dimensions

At TW 1, CONV birds had shorter and narrower tibiae than birds from the other categories. Because CONV birds were 2 wk younger than the SG strains at both TWs, the morphometric differences observed may be attributed to the difference in age, as tibial length and diameter increase as birds age (Lilburn, 1994; Talaty et al., 2009; Charuta et al., 2013). The differences in body conformation between FG and SG strains at the same BW were assessed by Weimer et al. (2020), who reported longer body and greater shank length in SG birds compared to FG birds, when these birds were 63 and 42 d, respectively, which agrees with the findings reported in our study. Interestingly, at TW 2, CONV birds still had shorter tibiae than the three categories of SG birds, while the differences in tibial diameter disappeared despite the age and BW differences between FG and SG strains. The differences among categories were still present when length and diameter were expressed relative to the BW, with SLOW birds exhibiting greater values at both TWs, while the differences among the other categories differed in each TW, suggesting age and/or BW-dependent changes.

A continuous increase in tibial length and diameter was reported by Talaty et al. (2009), who weekly measured changes in bone size traits from 2-7 wk of age to represent a lifecycle of commercial strains of broiler chickens. Specifically, tibial length showed a continuous linear increase from 2-7 weeks, while tibial diameter grew at a fast rate from weeks 2-4, with steady, yet slower increases between weeks 4-7. Although this study only evaluated FG strains of broiler chickens, these results suggest that bone length and diameter may have different rates of increase over time. Therefore, the differences in rate of increase in tibial length and diameter throughout the life cycle of the birds likely contributed to the similar values of tibial diameter among categories, despite the differences observed tibial length at TW 2. Because bone width plays a role in bone breaking strength (Williams et al., 2004), the increase in tibial diameter observed in CONV strains from TW 1 to TW 2 may indicate greater resistance to breaking, with less susceptibility to bone fracture. However, the increase in tibial diameter observed in FG strains may be due to the accelerated increase in BW, leading to a rapid load-induced expansion of the tibiotarsus by increasing the periosteal surface of the bone as suggested by Williams et al. (2004). Nonetheless, this rapid increase in tibial diameter is likely not accompanied by adequate osteonal infilling by osteoblast, resulting in a more porous cortical bone (Williams et al., 2004). Further research is needed to investigate differences in bone mineral density between FG and SG birds to determine if the enlargement in tibial diameter observed in CONV birds in this present study is accompanied by an increase in porosity of their cortical bone compared to SG birds, as described in Williams' study (2004). A recent study by Harash et al. (2020) showed that tibial width increased more rapidly in FG birds compared to SG birds. In addition, regression analyses revealed that SG birds had greater tibial length, weight, and width than FG birds at the same BW. However, the

researchers reported that although bone width was greater in FG birds at a similar age compared to SG birds, the cortical thickness of the bone did not differ between strains. Because the present study only measured total tibial diameter, it is not possible to determine if strains and categories differed in cortical thickness, which should be investigated in future studies.

Even though bone characteristics (e.g., mineral content, breaking strength, mineral density, and morphology) are commonly used to assess bone quality and development, the effects of these bone characteristics on leg health and walking ability are somewhat unclear in broiler chickens. For example, when evaluating the effects of diet, feed form, photoperiod, and sex on walking ability and bone quality, Brickett et al. (2007) found that females and birds provided with a longer period of darkness had greater bone mineral content, which reflected on their better walking ability compared to males and birds reared in a lighting program with short darkness period, respectively. However, in the same study tibial and femur dimensions (length and width) did not influence birds' gait score. These findings are congruent with the study conducted by Talaty et al. (2010), who reported similar bone mineral content and bone size traits in 4 crosses of FG birds, despite differences in walking ability. Nonetheless, other researchers have found a relationship between several tibial anatomic measurements and gait score (Toscano et al., 2013). Furthermore, differences in bone dimensions (Cruickshank and Sim, 1986; Guo et al., 2019), tibial weight (relative to BW), and bone mineral density were reported in birds exhibiting valgus-varus deformity compared to non-affected birds (Guo et al., 2019). Similarly, the incidence of TD was also associated with lower bone mineral content (Tablante et al., 2003).

It is important to emphasize that leg health is complex and multifactorial, encompassing bone deformities, contact dermatitis, leg abnormalities, gait score (as an indicator of walking ability),

as well as the aforementioned bone attributes (Bradshaw et al., 2002; Pedersen and Forkman, 2019). Therefore, solely focusing on differences in bone traits may not accurately translate into differences in walking ability and overall leg health. Still, the assessment of bone traits may indicate peculiarities and disturbances in bone development and health, which may be associated with some leg disorders that compromise leg health and contribute to lameness.

It is possible that the shorter tibiotarsus of CONV birds combined with their greater breast muscle yield compared to the remaining categories at both TWs (Chapter 4) may affected their locomotor ability (Chapter 6). Paxton et al. (2014) investigated the changes in pelvic limb muscles and bones relative to the BW in a FG strain of broiler chicken. Even though tibiotarsus length was found to be isometric to BW, other studies demonstrated that an emphasis on breast muscle yield and accelerated growth is related to reduced metabolic resources directed to leg muscles (Havenstein et al., 2003b; Fanatico et al., 2008; Arthur and Albers, 2009; Santos et al., 2021). This increase in breast muscle yield in FG strains has been shown to displace their center of mass cranially (Corr et al., 2003a; Paxton et al., 2014). Maintaining shorter legs may help birds control lateral motion of the center of mass and stabilize balance (Bauby and Kuo, 2000; Paxton et al., 2014). However, shorter limbs may be less efficient (Steudel-Numbers and Tilkens, 2004), which may lead to a decrease in walking and activity as a means to compensate for this greater energetic demand (Paxton et al., 2014). These changes may have direct or indirect welfare implications, as the reduction in locomotion and activity may cause and/or aggravate leg disorders, contact dermatitis, and lameness (Bessei, 2009). In agreement with these findings, differences between CONV and SLOW birds were observed in the latency-to-lie and group obstacle test, both objective tests to assess leg strength and walking ability (Chapter 6).

Although the changes in body conformation mentioned above may be associated with a decrease in activity, differences in activity can also affect bone traits. The concept that exercise and locomotor activity positively impact bone health has been widely documented in several species, including poultry (Reiter and Bessei, 1998; Kohrt et al., 2004; Pedersen et al., 2020; Pufall et al., 2021). In fact, stress and strain resulted from muscle forces and external loads are known to modulate bone remodeling, improving its mechanical function, with walking being the most common activity involved in mechanical loading of the appendicular skeleton (Shipov et al., 2010; Ruiz-Feria et al., 2014). Fast-growing birds are often reported to exhibit lower levels of activity, reduced walking behaviour, and prolonged time spent sitting compared to SG birds (Bizeray et al., 2000; Bokkers and Koene, 2003; Meluzzi and Sirri, 2009; Dixon, 2020).

Recent work by Pulcini et al. (2021) found remarkable differences in tibial shape in broiler strains raised in organic systems and differing in growth rate and walking behaviour, with a more pronounced curvature of the antero-posterior axis of the tibia being correlated with more static behaviour (e.g., resting, roosting). A companion study evaluating the behaviour, inactivity, and enrichment use of the same strains presented in this paper, revealed that CONV birds spent more time sitting, and less time standing and walking than the other SG categories at d 26 (Dawson et al., 2021). Overall, a similar pattern was also observed at d 42, with increased growth rate being associated with shorter time spent standing and walking (Dawson et al., 2021). While an overall increase in inactivity was observed as the birds grew, reaching 78-80% for all strains, these levels of inactivity were reached at a later age in birds with slower growth rates. In addition, at TW 2, an increase in growth rate was associated with a lower proportion of birds using all the enrichments provided as well as accessing the elevated platforms (Dawson et al., 2021).

Even though there is some inconsistency in the literature regarding the benefits of environmental enrichment, several studies reported an improvement in leg health, (indicated by differences in gait score, bone measurements, and incidence of leg deformities, HB, and FPD) likely as a result of an increase in activity (reviewed in Pedersen and Forkman, 2019). As an example, Yildiz et al. (2009) reported an increase in tibial length and weight in birds provided with intermittent lighting compared to birds reared under a continuous lighting program. This increase in bone measurements was attributed to a possible increase in the activity in the intermittent lighting group. This is in line with Pedersen et al.(2020), who reported thicker tibia distal diameter and lower incidence of arthritis and tenosynovitis when a 7 m distance between resources was added, which was probably due to an increase in walking behaviour. Collectively, these findings suggest that other than age, differences in behaviour and activity between FG and SG birds may have affected the differences in bone dimensions observed in this study.

5.5.1.4 Bone Dry Matter, Mineral, and Organic Content

Tibial dry matter, ash, and organic content were only measured at TW 2, when FG and SG birds were 48 and 62 d, respectively. The determination of ash content is used as an indicator of bone mineralization. In our study, the tibiae were selected due to their essential role for BW support and because disturbances in the tibia growth place can be associated with TD and lameness (Julian, 1998). The absolute weights of tibial traits provide a quantitative assessment of bone development, as bone mass increases during growth (Iwaniec and Turner, 2016). On the other hand, the relative values provide an “index” and quantitative indicator of bone growth in comparison to the increase in BW; this value has been shown to be altered in some bone disorders such as valgus-varus deformity (Guo et al., 2019).

Bones from the CONV birds had lighter dry matter, ash, and organic weight in comparison to FAST and MOD birds, despite the similarities in BW among these categories at TW 2. This difference in absolute weight of tibial components is likely attributed to the shorter tibial length of CONV birds at TW 2. As previously mentioned, CONV birds were 2 wk younger than the SG strains at the same TW, which may have contributed to shorter tibial length in the former, as tibia dimensions increase as birds grow (Yalcin et al., 2001; Talaty et al., 2009). Even though BW did not significantly differ among CONV, MOD, and FAST birds at TW 2, CONV birds still had the lowest dry matter and ash weight relative to the BW, which may indicate that bone growth and development may not keep pace with the sharp increase in body mass, resulting in excessive physical load for the bones to support, increasing the susceptibility of bone disorders (Rath et al., 2000). These findings are corroborated by McDevitt et al. (2006), who reported that at a similar BW, FG birds had shorter and lighter tibia than SG birds. However, in the same study FG birds exhibited greater ash and lower organic content (relative to the dry matter) than SG birds. In our study, despite the differences in some absolute dry matter, ash, and organic matter, the percentage of ash, organic matter, and the ratio of organic to inorganic matter did not differ among categories. In addition, CONV birds had similar ash per unit of length of the tibia compared to the other categories. Similar results were reported by Talaty et al. (2009), who did not find differences in bone mineral content (determined through dual-energy x-ray) among different commercial strains of broiler chickens at different ages. However, all the strains evaluated had similar BW and growth rate, which differs from our study. Talaty et al. (2009) also reported a lack of increase in bone mineral density (mineral per area of bone) after 4 wk of age, which was attributed to the reduction in locomotor activity as the birds age. Similarly, Rath et al. (2000) reported an increase in tibial

ash content of a commercial strain of broiler chickens that peaked between 3 to 5 wk of age and remained constant until the end of the production cycle. In our study, bone ash content was determined as an estimation of mineral content at the final TW, when the birds reached the heaviest BW (TW 2). Therefore, it was not possible to investigate the changes in bone mineralization over time. However, because the peak of bone mineral content was reported to occur before the birds were slaughtered, all the categories likely had reached the maximum mineral content at the age of the evaluation (7 and 9 wk of age for FG and SG strains, respectively). Thus, the different ages in which the tibial ash was assessed was likely not a major factor in the lack of difference in tibial ash and organic matter content among the categories, suggesting similarities in bone mineralization among strains, despite the large differences in growth rates.

Differences in bone morphology and mineralization between two commercial strains of broiler chickens were reported by Yalcin et al. (2001) during the first 16 d of age, whereas at later ages these differences disappeared. Similarly, Shim et al. (2012b) reported no difference in ash content between FG and SG strains evaluated at 6 wk of age. Because ash content was only evaluated at one time point in our study, it was not possible to determine if differences among the categories occurred at earlier stages of growth. Further investigation into bone mineral content and density at different time points, especially at younger ages in FG and SG strains is warranted to determine if bone development and susceptibility to leg disorders differ among strains at earlier stages of growth, due to the rapid increase in BW and bone growth in this period.

5.5.1.5 Tibial Dyschondroplasia

In our study, the incidence of TD was mainly found in MOD and FAST birds. This suggests that genetic selection for leg health variables has not been as intense in SG birds as in FG birds,

likely due to their reduced growth performance that is associated with lower incidence of leg disorders. However, because birds showing signs of lameness were promptly euthanized in our study, the incidence of TD may be higher than the values reported, although the overall mortality and cull rates were low (Torrey et al., 2021). Although the presence of TD has been documented to negatively affect bird's walking ability based on gait score assessment (Sanotra et al., 2001a), this finding has not been supported by other researchers (Fernandes et al., 2012). Because the lesions observed in our study were mild (presence of irregular cartilage in less than one third of the growth plate) (Edwards and Veltmann, 1983; Sanotra et al., 2001a), the walking ability of birds exhibiting TD lesions was likely not affected by the condition. .

5.5.2 Live Weight and Leg Traits at a Similar Age

At d 48, FG and other SG birds were expected to reach 3.2 and 2.1 kg, respectively. However, due to the differences in growth rates, categories differed in BW, with greater values observed as the growth rate was higher (CONV > FAST > MOD > SLOW). These results were expected as birds were grouped into categories based on their growth rate (Torrey et al., 2021). Overall, CONV birds exhibited longer and wider tibiae, but when the values were expressed as a ratio to the BW, CONV birds were lower than the other categories, suggesting that the rapid increase in BW is not accompanied by an equally fast increase in bone size, which could lead to excessive weight for the immature bones to support (Rath, 2000). As mentioned above, the potential effect of differences in behaviour and activity among the categories on bone traits should not be ruled out. Alternatively, BW and tibial size traits may have different rates and patterns of increase over a production cycle. To investigate bone growth as a function of age and BW during a commercial growing period (from hatch to slaughter), Applegate and Lilburn (2002) measured the growth of long bones (femur

and tibia) of a FG strain of broiler chicken at 1-wk intervals from 0-42 d old. The researchers reported a 3.7-fold increase in femur and tibial length, and a 3.8 and 5-fold increase in diameter (at the midpoint of the bone) of femur and tibia, respectively, whereas a 40-fold increase in BW was observed in the same period. These findings agree with the results presented in this study, providing evidence that bone dimensions show a slower increase relative to the rapid and sharp increase in BW over the growing period. However, because Applegate and Lilburn did not study a SG strain, it is unknown how bone growth changes over time for SG birds and more importantly, if these changes are associated with differences in walking ability.

Despite the differences in tibial morphology, CONV birds had similar TBS relative to the BW compared to FAST and MOD birds, yet lower than SLOW birds. The differences between CONV and SLOW birds are supported by other studies that found greater bone strength in SG birds in comparison to FG birds, suggesting that growth rate may have deleterious effects on bone strength and quality (Julian, 1998; Shim et al., 2012b). However, the similarities in TBS between CONV, FAST, and MOD birds suggest that these effects may only be evident when birds greatly differ in growth rate and BW. In addition, CONV birds exhibited the greatest absolute values of TBS suggesting that large differences in BW may have caused the differences in TBS relative to the BW between CONV and SLOW birds. The assessment of bone breaking strength at earlier ages would be relevant to investigate differences among the categories due to the rapid skeletal growth in this period and the smaller disparity in BW between FG and SG strains.

5.5.3 Effect of Sex and Strain

Differences between males and females were observed for most of the variables evaluated, with males generally having greater values than females at both TWs and the same age. These sex

differences in bone morphological and compositional traits are in accordance with findings reported in several previous studies (Yalcin et al., 2001; Venäläinen et al., 2006; Talaty et al., 2009), indicating that hormonal differences may influence BW and anatomical differences (Yalcin et al., 2001). While most of the variables differed among categories, only a few differences were observed among strains within the same category, especially among FAST birds. The differences among strains selected for a similar growth rate may be due to distinct selection criteria of each breeding company (Yalcin et al., 2001).

5.6 Conclusion

Based on the results of this study, differences in growth rate were associated with differences in most of the bone traits examined at a similar TW and at a similar age. Morphometric traits differed by category, with CONV birds having shorter absolute tibial length at both TWs and the shortest tibial length relative to the BW at TW 2 and at 48 d of age. However, both absolute TBS and TBS relative to BW of CONV were similar or greater than SG birds at both TW. Tibial ash content did not differ among categories at TW 2, suggesting similar bone mineralization among categories. These results suggest that differences in functional abilities of CONV compared to SG birds at a similar BW may be due to morphometric differences rather than differences in bone strength and bone mineralization. Other bone quality indicators (e. g. bone stiffness, bone mineral density, chemical composition, and cortical thickness) would be useful to provide a better understanding of bone development at different stages of growth over a life cycle of FG and SG birds especially at earlier stages that are characterized by a rapid increase in body mass and bone development.

Table 5.1: Effect of category on body weight (BW), tibial breaking strength (TBS), tibial morphology, and tibial ash and organic content (LS-means \pm SEM) at Target Weights 1 and 2. At Target weight 1, birds of conventional and slower-growing strains were 34 and 48 d of age, respectively. At Target Weight 2, birds of conventional strains and the remaining categories were 48 and 62 d, respectively.

Variable	Category			
	CONV	FAST	MOD	SLOW
Target Weight 1¹				
BW (g)	1,857 \pm 40.9 ^d	2,519 \pm 38.6 ^a	2,359 \pm 34.2 ^b	2,015 \pm 29.5 ^c
TBS (N) ²	290.1 \pm 12.55	300.3 \pm 8.87	284.2 \pm 7.95	274.1 \pm 7.60
TBS:BW (N/kg) ³	156.0 \pm 6.49 ^a	119.0 \pm 3.42 ^c	120.7 \pm 3.26 ^c	136.0 \pm 3.63 ^b
Diameter (mm)	7.21 \pm 0.144 ^c	8.59 \pm 0.116 ^a	8.33 \pm 0.108 ^{ab}	8.18 \pm 0.105 ^b
Length (mm)	95.6 \pm 0.75 ^c	116.1 \pm 0.61 ^a	112.5 \pm 0.56 ^b	111.8 \pm 0.55 ^b
Diameter:BW (mm/kg)	3.87 \pm 0.081 ^a	3.40 \pm 0.047 ^b	3.54 \pm 0.047 ^b	4.04 \pm 0.053 ^a
Length:BW (mm/kg)	51.48 \pm 0.954 ^b	46.10 \pm 0.583 ^c	47.70 \pm 0.572 ^c	55.50 \pm 0.657 ^a
Length:Diameter	13.28 \pm 0.226	13.50 \pm 0.154	13.50 \pm 0.147	13.66 \pm 0.148
Target Weight 2⁴				
BW (g)	3,264 \pm 60.0 ^{ab}	3,437 \pm 48.5 ^a	3,185 \pm 42.3 ^b	2,844 \pm 37.6 ^c
TBS (N)	362.8 \pm 12.63 ^a	339.4 \pm 9.30 ^{ab}	318.6 \pm 8.13 ^b	320.6 \pm 8.08 ^b
TBS:BW (N/kg)	111.2 \pm 3.42 ^a	98.7 \pm 2.60 ^b	100.0 \pm 2.46 ^b	112.7 \pm 2.74 ^a
Diameter (mm)	9.05 \pm 0.143	9.31 \pm 0.012	9.29 \pm 0.109	9.26 \pm 0.108
Length (mm)	116.9 \pm 0.73 ^c	129.9 \pm 0.64 ^a	128.3 \pm 0.59 ^{ab}	127.6 \pm 0.58 ^b
Diameter:BW (mm/kg)	2.78 \pm 0.046 ^{bc}	2.71 \pm 0.036 ^c	2.90 \pm 0.035 ^b	3.25 \pm 0.039 ^a
Length:BW (mm/kg)	35.80 \pm 0.528 ^d	37.80 \pm 0.444 ^c	40.28 \pm 0.439 ^b	44.90 \pm 0.489 ^a
Length:Diameter	12.90 \pm 0.173 ^b	13.95 \pm 0.149 ^a	13.81 \pm 0.137 ^a	13.77 \pm 0.136 ^a
Dry matter wt (g) ⁵	9.98 \pm 0.207 ^c	11.62 \pm 0.154 ^a	10.85 \pm 0.161 ^b	10.39 \pm 0.163 ^{bc}
Ash wt (g)	3.95 \pm 0.091 ^c	4.61 \pm 0.074 ^a	4.30 \pm 0.073 ^b	4.18 \pm 0.068 ^{bc}
Organic matter wt (g)	6.04 \pm 0.125 ^c	7.01 \pm 0.910 ^a	6.54 \pm 0.102 ^b	6.21 \pm 0.105 ^{bc}
Dry matter wt:BW (%)	3.05 \pm 0.046 ^c	3.38 \pm 0.030 ^b	3.40 \pm 0.043 ^b	3.65 \pm 0.042 ^a
Ash wt:BW (%)	1.21 \pm 0.015 ^c	1.33 \pm 0.014 ^b	1.35 \pm 0.018 ^b	1.47 \pm 0.017 ^a
Ash content (%)	39.42 \pm 0.363	39.62 \pm 0.278	39.63 \pm 0.326	40.35 \pm 0.323
Organic matter (%)	60.58 \pm 0.363	60.38 \pm 0.278	60.37 \pm 0.326	59.65 \pm 0.323
Organic:Inorganic	1.53 \pm 0.024	1.52 \pm 0.017	1.52 \pm 0.023	1.49 \pm 0.0193
Ash:Length (g/mm)	0.34 \pm 0.006 ^{ab}	0.35 \pm 0.005 ^a	0.33 \pm 0.005 ^b	0.33 \pm 0.005 ^b

¹ Number of birds per category at Target Weight 1: CONV: n = 34; FAST: n = 78; MOD: n = 85; SLOW: n = 86.

² Absolute tibial breaking strength (TBS). Maximum TBS expressed in newtons (N).

³ Relative tibial breaking strength (TBS). Maximum TBS was obtained in newtons (N) and adjusted for the BW.

⁴ Number of birds per category at Target Weight 2: CONV: n = 54; FAST: n = 95; MOD: n = 101; SLOW: n = 103.

⁵ Tibial dry matter, and organic and inorganic content were only obtained at Target Weight 2.

^{a-d} Different superscripts within the same row represent significant differences between categories (P < 0.05).

Table 5.2: Differences in body weight (BW), tibial breaking strength (TBS), tibial morphology, and tibial ash and organic content (LS-means \pm SEM) among CONV strains at Target Weights 1 and 2. At Target Weights 1 and 2, CONV strains were 34 and 48 days, respectively.

Variable	Strain	
	B	C
Target Weight 1¹		
BW (g)	1,953 \pm 56.5	1,766 \pm 58.8
TBS (N) ²	326.5 \pm 18.67	257.6 \pm 16.73
TBS:BW (N/kg) ³	167.2 \pm 9.25	146.0 \pm 9.04
Diameter (mm)	7.47 \pm 0.196	6.95 \pm 0.211
Length (mm)	98.2 \pm 1.01 ^a	93.1 \pm 1.09 ^b
Diameter:BW (mm/kg)	3.80 \pm 0.104	3.94 \pm 0.123
Length:BW (mm/kg)	50.00 \pm 1.224	52.70 \pm 1.473
Length:Diameter	13.14 \pm 0.294	13.42 \pm 0.345
Target Weight 2⁴		
BW (g)	3,298 \pm 89.7	3,231 \pm 79.9
TBS (N)	376.6 \pm 19.81	349.5 \pm 15.95
TBS:BW (N/kg)	114.2 \pm 5.81	108.1 \pm 4.81
Diameter (mm)	9.29 \pm 0.223	8.82 \pm 0.183
Length (mm)	118.6 \pm 1.09	115.4 \pm 0.99
Diameter:BW (mm/kg)	2.81 \pm 0.069	2.74 \pm 0.059
Length:BW (mm/kg)	35.95 \pm 0.798	35.70 \pm 0.693
Length:Diameter	12.75 \pm 0.257	13.08 \pm 0.232
Dry matter wt (g) ⁵	10.12 \pm 0.305	9.84 \pm 0.285
Ash wt (g)	4.06 \pm 0.134	3.81 \pm 0.125
Organic matter wt (g)	6.06 \pm 0.183	6.02 \pm 0.174
Dry matter wt:BW (%)	3.05 \pm 0.063	3.04 \pm 0.057
Ash wt:BW (%)	1.22 \pm 0.022	1.17 \pm 0.021
Ash content (%)	40.24 \pm 0.540	38.60 \pm 0.489
Organic matter (%)	59.76 \pm 0.540	61.40 \pm 0.489
Organic:Inorganic	1.49 \pm 0.035	1.59 \pm 0.032
Ash:Length (g/mm)	0.34 \pm 0.009	0.34 \pm 0.009

¹ Number of birds per strain at Target Weight 1: B: n = 19; C: n = 15.

² Absolute tibial breaking strength (TBS). Maximum TBS expressed in newtons (N).

³ Relative tibial breaking strength (TBS). Maximum TBS was obtained in newtons (N) and adjusted for the BW.

⁴ Number of birds per strain at Target Weight 2: B: n = 23; C: n = 31.

⁵ Tibial dry matter, and organic and inorganic content were only obtained at Target Weight 2.

^{a-b} Different superscripts within the same row represent significant differences between categories (P < 0.05).

Table 5.3: Differences in body weight (BW), tibial breaking strength (TBS), tibial morphology, and tibial ash and organic content (LS-means \pm SEM) among FAST strains at Target Weights 1 and 2. At Target Weights 1 and 2, FAST strains were 48 and 62 days, respectively.

Variable	Strain			
	F	G	I	M
Target Weight 1¹				
BW (g)	2,617 \pm 87.2	2,470 \pm 6.8	2,428 \pm 66.0	2,567 \pm 86.9
TBS (N) ²	303.5 \pm 19.72	319.5 \pm 17.10	286.5 \pm 15.37	292.8 \pm 18.54
TBS:BW (N/kg) ³	116.0 \pm 7.29	129.0 \pm 6.65	118.0 \pm 6.17	114.1 \pm 7.02
Diameter (mm)	9.24 \pm 0.271	8.46 \pm 0.207	8.58 \pm 0.206	8.12 \pm 0.238
Length (mm)	113.8 \pm 1.28	116.2 \pm 1.09	115.7 \pm 1.06	118.7 \pm 1.40
Diameter:BW (mm/kg)	3.53 \pm 0.106	3.41 \pm 0.086	3.54 \pm 0.087	3.16 \pm 0.095
Length:BW (mm/kg)	43.48 \pm 1.182	47.04 \pm 1.065	47.66 \pm 1.059	46.00 \pm 1.324
Length:Diameter	12.31 \pm 0.304 ^b	13.72 \pm 0.284 ^a	13.48 \pm 0.271 ^{ab}	14.62 \pm 0.371 ^a
Target Weight 2⁴				
BW (g)	3,497 \pm 84.2	3,419 \pm 92.9	3,467 \pm 95.3	3,366 \pm 11.2
TBS (N)	327.4 \pm 15.34	321.5 \pm 16.62	317.2 \pm 17.77	397.6 \pm 25.17
TBS:BW (N/kg)	93.6 \pm 4.16	94.0 \pm 4.67	91.5 \pm 4.94	118.1 \pm 7.22
Diameter (mm)	9.61 \pm 0.203	9.13 \pm 0.227	9.48 \pm 0.232	9.03 \pm 0.269
Length (mm)	126.6 \pm 1.07 ^b	129.4 \pm 1.25 ^{ab}	133.3 \pm 1.24 ^a	130.6 \pm 1.50 ^{ab}
Diameter:BW (mm/kg)	2.75 \pm 0.059	2.70 \pm 0.071	2.72 \pm 0.069	2.68 \pm 0.083
Length:BW (mm/kg)	36.20 \pm 0.706	37.80 \pm 0.897	38.42 \pm 0.863	38.80 \pm 1.072
Length:Diameter	13.15 \pm 0.238	14.15 \pm 0.298	14.05 \pm 0.291	14.47 \pm 0.364
Dry matter wt (g) ⁵	11.30 \pm 0.266	11.50 \pm 0.298	11.56 \pm 0.298	12.15 \pm 0.360
Ash wt (g)	4.29 \pm 0.129	4.61 \pm 0.143	4.79 \pm 0.144	4.78 \pm 0.173
Organic matter wt (g)	7.01 \pm 0.158	6.89 \pm 0.176	6.77 \pm 0.176	7.37 \pm 0.212
Dry matter wt:BW (%)	3.23 \pm 0.061 ^b	3.36 \pm 0.070 ^{ab}	3.30 \pm 0.07 ^{ab}	3.60 \pm 0.084 ^a
Ash wt:BW (%)	1.22 \pm 0.025 ^b	1.34 \pm 0.028 ^{ab}	1.38 \pm 0.028 ^a	1.41 \pm 0.033 ^a
Ash content (%)	38.15 \pm 0.479 ^b	39.81 \pm 0.541 ^{ab}	41.39 \pm 0.541 ^a	39.14 \pm 0.650 ^{ab}
Organic matter (%)	61.85 \pm 0.497 ^a	60.18 \pm 0.541 ^{ab}	58.61 \pm 0.541 ^b	60.86 \pm 0.650 ^{ab}
Organic:Inorganic	1.63 \pm 0.031 ^a	1.52 \pm 0.035 ^{ab}	1.42 \pm 0.035 ^b	1.57 \pm 0.0415 ^{ab}
Ash:Length (g/mm)	0.34 \pm 0.009	0.34 \pm 0.009	0.36 \pm 0.009	0.36 \pm 0.011

¹ Number of birds per strain at Target Weight 1: F: n = 16; G: n = 22; I: n = 24; M: n = 16.

² Absolute tibial breaking strength (TBS). Maximum TBS expressed in newtons (N).

³ Relative tibial breaking strength (TBS). Maximum TBS was obtained in newtons (N) and adjusted for the BW.

⁴ Number of birds per strain at Target Weight 2: F: n = 32; G: n = 24; I: n = 23; M: n = 16.

⁵ Tibial dry matter, and organic and inorganic content were only obtained at Target Weight 2.

^{a-b} Different superscripts within the same row represent significant differences between categories (P < 0.05).

Table 5.4: Differences in body weight (BW), tibial breaking strength (TBS), tibial morphology, and tibial ash and organic content (LS-means \pm SEM) among MOD strains at Target Weights 1 and 2. At Target Weights 1 and 2, MOD strains were 48 and 62 days, respectively.

Variable	Strain			
	E	H	O	S
Target Weight 1¹				
BW (g)	2,551 \pm 70.1	2,226 \pm 74.1	2,434 \pm 66.2	2,241 \pm 61.6
TBS (N) ²	308.3 \pm 16.21	249.8 \pm 16.24	277.7 \pm 14.60	304.9 \pm 16.03
TBS:BW (N/kg) ³	120.8 \pm 6.14	112.2 \pm 7.07	114.1 \pm 5.79	136.2 \pm 6.91
Diameter (mm)	8.64 \pm 0.209	7.75 \pm 0.235	8.61 \pm 0.206	8.33 \pm 0.202
Length (mm)	114.9 \pm 1.07	109.6 \pm 1.29	113.5 \pm 1.04	111.9 \pm 1.04
Diameter:BW (mm/kg)	3.38 \pm 0.08	3.50 \pm 0.111	3.54 \pm 0.087	3.72 \pm 0.093
Length:BW (mm/kg)	45.02 \pm 1.012	49.30 \pm 1.401	46.63 \pm 1.035	49.93 \pm 1.122
Length:Diameter	13.29 \pm 0.271	14.12 \pm 0.363	13.17 \pm 0.265	13.42 \pm 0.274
Target Weight 2⁴				
BW (g)	3,198 \pm 87.9	3,101 \pm 74.1	3,368 \pm 91.6	3,080 \pm 83.7
TBS (N)	339.3 \pm 17.84	338.8 \pm 15.25	282.4 \pm 15.11	317.6 \pm 16.69
TBS:BW (N/kg)	106.0 \pm 5.38 ^{ab}	109.1 \pm 4.71 ^a	83.90 \pm 4.345 ^b	103.1 \pm 5.24 ^{ab}
Diameter (mm)	9.55 \pm 0.236	8.99 \pm 0.189	9.48 \pm 0.227	9.15 \pm 0.221
Length (mm)	131.2 \pm 1.26	126.2 \pm 1.04	129.8 \pm 1.19	126.2 \pm 1.17
Diameter:BW (mm/kg)	2.97 \pm 0.076	2.88 \pm 0.062	2.81 \pm 0.069	2.97 \pm 0.074
Length:BW (mm/kg)	41.00 \pm 0.940	40.70 \pm 0.792	38.51 \pm 0.855	40.98 \pm 0.921
Length:Diameter	13.74 \pm 0.288	14.05 \pm 0.249	13.70 \pm 0.276	13.78 \pm 0.282
Dry matter wt (g) ⁵	11.40 \pm 0.329	10.77 \pm 0.297	10.93 \pm 0.329	10.29 \pm 0.329
Ash wt (g)	4.63 \pm 0.149	4.25 \pm 0.134	4.25 \pm 0.149	4.08 \pm 0.149
Organic matter wt (g)	6.77 \pm 0.209	6.53 \pm 0.189	6.68 \pm 0.209	6.20 \pm 0.209
Dry matter wt:BW (%)	3.56 \pm 0.088	3.47 \pm 0.078	3.24 \pm 0.088	3.34 \pm 0.088
Ash wt:BW (%)	1.44 \pm 0.037 ^a	1.37 \pm 0.034 ^{ab}	1.25 \pm 0.037 ^b	1.32 \pm 0.036 ^{ab}
Ash content (%)	40.55 \pm 0.669	39.40 \pm 0.592	38.86 \pm 0.669	39.70 \pm 0.669
Organic matter (%)	59.44 \pm 0.670	60.59 \pm 0.592	61.13 \pm 0.670	60.30 \pm 0.670
Organic:Inorganic	1.47 \pm 0.047	1.55 \pm 0.042	1.62 \pm 0.047	1.53 \pm 0.047
Ash:Length (g/mm)	0.35 \pm 0.010	0.33 \pm 0.009	0.32 \pm 0.009	0.32 \pm 0.009

¹ Number of birds per strain at Target Weight 1: E: n = 23; H: n = 15; O: n = 24; S: n = 23.

² Absolute tibial breaking strength (TBS). Maximum TBS expressed in newtons (N).

³ Relative tibial breaking strength (TBS). Maximum TBS was obtained in newtons (N) and adjusted for the BW.

⁴ Number of birds per strain at Target Weight 2: E: n = 23; H: n = 32; O: n = 23; S: n = 23.

⁵ Tibial dry matter, and organic and inorganic content were only obtained at Target Weight 2.

^{a-b} Different superscripts within the same row represent significant differences between categories ($P < 0.05$).

Table 5.5: Differences in body weight (BW), tibial breaking strength (TBS), tibial morphology, and tibial ash and organic content (LS-means \pm SEM) among SLOW strains at Target Weights 1 and 2. At Target Weights 1 and 2, SLOW strains were 48 and 62 days, respectively.

Variable	Strain			
	D	J	K	N
Target Weight 1¹				
BW (g)	1,993 \pm 66.3	2,132 \pm 57.9	1,967 \pm 53.5	1,970 \pm 53.6
TBS (N) ²	273.9 \pm 17.34	265.5 \pm 13.96	269.6 \pm 14.18	287.8 \pm 15.13
TBS:BW (N/kg) ³	137.5 \pm 8.41	124.4 \pm 6.32	137.1 \pm 6.88	146.0 \pm 7.40
Diameter (mm)	7.58 \pm 0.226	8.59 \pm 0.206	8.13 \pm 0.197	8.46 \pm 0.205
Length (mm)	110.9 \pm 1.27	114.3 \pm 1.05	110.9 \pm 1.03	110.9 \pm 1.03
Diameter:BW (mm/kg)	3.80 \pm 0.116	4.03 \pm 0.099	4.10 \pm -0.102	4.29 \pm 0.107
Length:BW (mm/kg)	55.61 \pm 1.526	53.61 \pm 1.191	56.40 \pm 1.254	56.19 \pm 1.263
Length:Diameter	14.63 \pm 0.368 ^a	13.30 \pm 0.268 ^{ab}	13.65 \pm 0.278 ^{ab}	13.10 \pm 0.267 ^b
Target Weight 2⁴				
BW (g)	2,838 \pm 68.1	3,022 \pm 81.5	2,764 \pm 75.1	2,760 \pm 75.9
TBS (N)	377.6 \pm 16.99	308.8 \pm 15.73	300.6 \pm 15.54	301.7 \pm 16.19
TBS:BW (N/kg)	133.0 \pm 5.77 ^a	102.2 \pm 5.04 ^b	108.8 \pm 5.43 ^{ab}	109.3 \pm 5.66 ^{ab}
Diameter (mm)	8.89 \pm 0.186	9.57 \pm 0.230	9.04 \pm 0.217	9.57 \pm 0.236
Length (mm)	127.3 \pm 1.07	129.9 \pm 1.19	126.4 \pm 1.16	127.0 \pm 1.21
Diameter:BW (mm/kg)	3.12 \pm 0.067	3.17 \pm 0.078	3.27 \pm 0.081	3.46 \pm 0.088
Length:BW (mm/kg)	44.88 \pm 0.883	43.00 \pm 0.958	45.72 \pm 1.015	46.04 \pm 1.052
Length:Diameter	14.30 \pm 0.255	13.58 \pm 0.275	13.98 \pm 0.282	13.28 \pm 0.276
Dry matter wt (g) ⁵	10.96 \pm 0.299	10.54 \pm 0.328	10.33 \pm 0.332	9.73 \pm 0.344
Ash wt (g)	4.26 \pm 0.126	4.32 \pm 0.139	4.08 \pm 0.141	4.07 \pm 0.145
Organic matter wt (g)	6.69 \pm 0.194 ^a	6.22 \pm 0.211 ^{ab}	6.24 \pm 0.215 ^{ab}	5.66 \pm 0.223 ^b
Dry matter wt:BW (%)	3.86 \pm 0.076	3.48 \pm 0.085	3.70 \pm 0.086	3.50 \pm 0.090
Ash wt:BW (%)	1.50 \pm 0.031	1.42 \pm 0.034	1.47 \pm 0.034	1.47 \pm 0.036
Ash content (%)	39.21 \pm 0.584	40.89 \pm 0.647	39.60 \pm 0.659	41.70 \pm 0.687
Organic matter (%)	60.79 \pm 0.584	59.10 \pm 0.647	60.40 \pm 0.659	58.30 \pm 0.687
Organic:Inorganic	1.56 \pm 0.035	1.46 \pm 0.038	1.54 \pm 0.039	1.41 \pm 0.0413
Ash:Length (g/mm)	0.33 \pm 0.009	0.33 \pm 0.009	0.32 \pm 0.009	0.32 \pm 0.010

¹ Number of birds per strain at Target Weight 1: D: n = 16; J: n = 24; K: n = 23; N: n = 23.

² Absolute tibial breaking strength (TBS). Maximum TBS expressed in newtons (N).

³ Relative tibial breaking strength (TBS). Maximum TBS was obtained in newtons (N) and adjusted for the BW.

⁴ Number of birds per strain at Target Weight 2: D: n = 32; J: n = 25; K: n = 24; N: n = 22.

⁵ Tibial dry matter, and organic and inorganic content were only obtained at Target Weight 2.

^{a-b} Different superscripts within the same row represent significant differences between categories ($P < 0.05$).

Table 5.6: Differences in body weight (BW), tibial breaking strength (TBS) and tibial morphology (LS-means \pm SEM) among categories at the same age (48 d).

Variable	Category ¹			
	CONV	FAST	MOD	SLOW
BW (g)	3,257 \pm 56.4 ^a	2,518 \pm 32.8 ^b	2,359 \pm 32.8 ^c	2,015 \pm 27.9 ^d
TBS (N) ²	363.7 \pm 13.60 ^a	305.9 \pm 9.54 ^b	285.0 \pm 8.51 ^{bc}	271.0 \pm 8.02 ^c
TBS:BW (N/kg) ³	111.8 \pm 4.03 ^b	121.0 \pm 3.66 ^{ab}	121.8 \pm 3.48 ^{ab}	134.1 \pm 3.83 ^a
Diameter (mm)	9.07 \pm 0.139 ^a	8.59 \pm 0.111 ^b	8.33 \pm 0.104 ^{bc}	8.18 \pm 0.101 ^c
Length (mm)	117.1 \pm 0.71 ^a	116.1 \pm 0.60 ^a	112.4 \pm 0.55 ^b	111.8 \pm 0.55 ^b
Diameter:BW (mm/kg)	2.78 \pm 0.039 ^c	3.39 \pm 0.040 ^b	3.53 \pm 0.040 ^b	4.03 \pm 0.046 ^a
Length:BW (mm/kg)	35.95 \pm 0.496 ^c	46.00 \pm 0.548 ^b	47.70 \pm 0.537 ^b	55.40 \pm 0.617 ^a
Length:Diameter	12.88 \pm 0.167 ^b	13.50 \pm 0.150 ^a	13.50 \pm 0.142 ^a	13.66 \pm 0.143 ^a

¹ Number of birds per category at 48 d of age: CONV: n = 54; FAST: n = 78; MOD: n = 85; SLOW: n = 86.

² Absolute tibial breaking strength (TBS). Maximum TBS expressed in newtons (N).

³ Relative tibial breaking strength (TBS). Maximum TBS was obtained in newtons (N) and adjusted for the BW.

^{a-d} Different superscripts within the same row represent significant differences between categories (P < 0.05).



A

B

Figure 5.1: Pictures illustrating absence (A) and presence (B) of tibial dyschondroplasia (TD) in fast-growing and slower-growing strains of broiler chickens at 48 and 62 d of age, respectively (target weight 2). The presence of TD is characterized by the abnormal accumulation of cartilage (lesion) in the growth plate (GP).

Chapter 6: In Pursuit of a Better Broiler: Walking Ability and Incidence of Contact Dermatitis in Conventional and Slower-Growing Strains of Broiler Chickens³

6.1 Abstract

There is an increasing interest in the use of slower-growing (SG) birds as an attempt to improve broilers' walking ability. In this study, the mobility, incidence and severity of contact dermatitis, and litter moisture content were assessed from a population of birds described in Chapter 4. A total of 4 to 6 birds per pen (equal number of males and females) were tested the latency-to-lie (LTL) and group obstacle tests within one week prior to the birds reaching 2 target weights (TWs) of 2.1 kg (TW 1: 34 d for CONV and 48 d for SG strains) and 3.2 kg (TW 2: 48 d for CONV and 62 d for SG strains). The individual bird BW was measured prior to each test. The incidence of contact dermatitis was evaluated a day prior to each TW. Litter moisture content was determined biweekly from d 14 to d 56. At both TWs, the BW of CONV birds was similar to SLOW birds, yet lighter than FAST and MOD in the LTL test. At TW1, CONV remained standing in the LTL test longer than FAST birds. At TW 2, CONV, MOD, and FAST birds had similar LTL. CONV birds were lighter than FAST in the group obstacle test, yet CONV and FAST birds exhibited a similar number of obstacle crossings at both TWs. At TW 1, CONV birds had greater incidence of FPD than FAST and MOD, while at TW 2, CONV birds had greater incidence than the other categories. The incidence of HB was greater in CONV and MOD compared to SLOW birds at TW 1, while at TW 2, the incidence of HB was greater in CONV and FAST birds than MOD and SLOW birds. Litter moisture content was high in all categories from d 28 onwards. Our results indicate that both BW

³ A version of this chapter will be submitted to Poultry Science.

and growth rate influence leg strength and walking ability, whereas the incidence of contact dermatitis was influenced by litter moisture content and to a lesser extent, growth rate.

Keywords: Lameness, leg health, growth rate, genotypes, slower-growth, mobility tests

6.2 Introduction

The growing global demand for animal products has contributed to the intensification and growth of the poultry industry (Petracci et al., 2015). Due to improvements in nutrition, health, management strategies, veterinary care, and genetic selection, conventional strains of broiler chickens, commonly referred to as fast-growing (**FG**) strains, have better feed conversion, higher growth rate (> 60 g/d), and reach market weight at an earlier age than ever before (about 2.5 kg in 40 d) (Havenstein et al., 1994, 2003a, b; Doğan et al., 2019; Mancinelli et al., 2020). However, this heavy body weight reached over a short time frame has been linked to the development of bone abnormalities and lameness (Julian, 1998; Bradshaw et al., 2002; Kierończyk et al., 2017).

Lameness is a broad term used to describe impaired walking ability and several debilitating conditions, resulting from multifactorial origins (Sørensen et al., 1999; Bradshaw et al., 2002; Kierończyk et al., 2017). Lameness commonly shows reduced walking ability, difficulty standing, prolonged and frequent squatting while walking, and reduced ability to perform natural behaviours (Julian, 1998; McGeown et al., 1999; Weeks et al., 2000; Danbury et al., 2000; Dawkins et al., 2009; Caplen et al., 2013; Vasdal et al., 2018; Norring et al., 2019; Rayner et al., 2020). Lameness can be caused by injuries, trauma, infectious, and non-infectious factors, which can affect bones, muscle, skin, or the nervous system (Bradshaw et al., 2002; Kierończyk et al., 2017). Previous studies have demonstrated that lameness can be painful; lame birds self-selected feeds with

analgesics (Danbury et al., 2000) and analgesic treatment improved lame birds' walking ability (McGeown et al., 1999; Caplen et al., 2013). In addition, severely lame birds have difficulty reaching the feeders and drinkers, leading to malnutrition and mortality (Bradshaw et al., 2002; Kierończyk, 2017). In the USA alone, economic losses caused by lameness and skeletal disorders have been estimated to cost producers more than \$150 million USD per year (Kierończyk, 2017). Therefore, lameness represents both a welfare and economic issue in broiler production.

The most common disorders that affect broiler chickens' walking ability include bacterial chondronecrosis with osteomyelitis, tibial dyschondroplasia (**TD**), valgus-varus deformity, and contact dermatitis (Bradshaw et al., 2002). These abnormalities mainly affect the bones and joints of broiler chickens (Edwards and Veltmann, 1983; Julian, 1984; Bradshaw et al., 2002; Shim et al., 2012a), causing problems with skeletal or structural development. Unlike other conditions causing lameness, contact dermatitis is associated with inflammation and lesions of the skin rather than disturbances in the bone structure (Bessei, 2006). In broiler chickens, contact dermatitis is commonly found on the feet or hocks, being referred to as footpad dermatitis (**FPD**) and hock burns (**HB**), respectively (Hartcher and Lum, 2020).

Severe contact dermatitis can be associated with pain and increased propensity to secondary bacterial infection, which may aggravate leg disorders (Bessei, 2006; Hartcher and Lum, 2020). Even though the low locomotor activity and prolonged time spent sitting are not a welfare problem *per se*, they can cause or aggravate the risk of contact dermatitis, especially if the birds are raised in poor environmental conditions, with wet litter and high ammonia levels (Robins and Phillips, 2011). In addition, increased locomotor activity is associated with improved bone development

and quality, potentially decreasing the propensity for leg disorders (Reiter and Bessei, 1998; Bizeray et al., 2000; Hartcher and Lum, 2020).

The combination of selection for accelerated growth and low locomotor activity are considered risk factors for the development of skeletal disorders and skin lesions that may impair birds' walking ability (Bradshaw et al., 2002). Because of the large influence of lameness on both welfare and economics, breeding companies began incorporating skeletal health traits into breeding programs to mitigate the occurrence of bone abnormalities and lameness in FG strains over the past 25 years (Angel, 2007; Whitehead, 2007; Kapell et al., 2012). However, recent studies have estimated moderate to severe gait impairment in about 14 - 30% of FG broiler chickens (Bassler et al., 2013; Kittelsen et al., 2017; Vasdal et al., 2018), with 3.3% of the birds being almost unable to walk based on gait scores (Knowles et al., 2008), suggesting that lameness is still an ongoing issue.

The Bristol six-point gait scoring system developed by Kestin et al. (1992) is the most widely used methodology to investigate walking ability and lameness in broiler chickens (Caplen et al., 2012). Scores equal to or greater than 3 are assumed to be painful, indicating that the welfare of birds may be compromised (McGeown et al., 1999; Weeks et al., 2000; Knowles et al., 2008; Caplen et al., 2013; Hartcher and Lum, 2020). Despite the widespread use of the Bristol gait scoring scheme to assess lameness in commercial broiler flocks at farm and research settings, this method provides a subjective estimation of birds' walking ability and requires observers to classify the different degrees of gait problems. This can be difficult when comparing strains with vastly different phenotypes, as differences in motivation to walk (Bizeray et al., 2002a; Bokkers and Koene, 2004), body conformation (Corr et al., 2003a), and temperament (Bizeray et al., 2000; Castellini et al.,

2002; Bokkers and Koene, 2004; Bessei, 2006; Dixon, 2020) may influence birds' locomotion and/or gait. Therefore, other more objective tests have been studied to assess lameness and leg health in broiler chickens.

Two validated behavioural tests for lameness and mobility include the latency-to-lie (**LTL**) test, developed by Weeks et al. (2002), and the group obstacle test, developed by Caplen et al. (2014). The LTL assesses the length of time the birds will stand to avoid lying in shallow water, which is considered to be a novel experience and an aversive stimulus. The group obstacle test measures the frequency with which birds will cross an obstacle placed in their home pen to obtain access to water and feed, critical resources that are located on opposite sides of the obstacle. Both tests are correlated with the traditional Bristol gait scoring system, with lame birds (high gait score) lying down earlier in water (Weeks et al., 2002; Caplen et al., 2014) and showing fewer obstacle crossings (Caplen et al., 2014) compared to sound birds (low gait score), in the LTL and group obstacle test, respectively. However, these tests were mainly conducted using FG chickens (Berg and Sanotra, 2003; Caplen et al., 2014) or they compared a limited number of SG strains (Singh et al., 2021). Therefore, there is scarce information on the possible behavioural differences between FG and SG birds using objective tests that assess walking ability and lameness in broiler chickens.

Due to the welfare implications associated with fast growth rate, especially those causing lameness and impaired locomotion, there is an increasing interest in the use of SG strains in commercial broiler production. Previous comparisons between strains diverging in growth rate demonstrated better walking ability and a lower incidence of contact dermatitis in slower-growing (**SG**) strains compared to FG chickens (Castellini et al., 2016; Dixon, 2020). However, there is a scarcity of studies comparing strains differing in growth rates under the same confined conditions. In addition,

considering the continuous and dynamic changes in genetic selection, previous studies comparing SG and FG strains may not accurately reflect the genetics of modern broiler chickens (Rayner et al., 2020). Although the term “slow growing” commonly refers to birds with reduced growth rate and feed efficiency compared to FG birds, it encompasses a heterogeneous group of birds that represents various rates of growth (Doğan et al., 2019; Mancinelli et al., 2020).

Therefore, the aim of this study was to investigate the differences in mobility and contact dermatitis between 2 FG and 12 SG strains of broiler chickens raised under the same conditions and processed at similar market weights. The LTL and group obstacle tests were used to investigate leg strength and walking ability of both FG and SG birds. We hypothesized that SG birds would have better leg health, as indicated by longer time spent standing in water, more obstacle crossings, and lower incidence and severity of contact dermatitis compared to FG chickens.

6.3 Materials and Methods

6.3.1 Hatching and Husbandry

The procedures carried in this study were approved by the University of Guelph’s Animal Care Committee (AUP #3746) and were in accordance with the Canadian Council for Animal Care’s guidelines (CCAC, 2009).

This study is part of a multidisciplinary project that investigated production performance, meat quality, behaviour, physiology, bone traits, health, inactivity, and welfare of FG and SG strains selected for distinct growth rates, described in other associated papers. The complete details regarding the overall methodology of this multidisciplinary study (e.g., incubation conditions, animal handling, husbandry, management, and housing) are available elsewhere (Torrey et al.,

2021). The overview of the methodology used in this chapter and other associated studies that encompass the multidisciplinary project is described in Chapter 3.

6.3.2 Latency-to-Lie Test

Four focal birds (2 males and 2 females) were tested 2 to 7 days prior to processing at each TW to assess leg strength. These were the same birds used to determine bone traits, described in Chapter 5. The LTL test followed a similar methodology described by Berg and Sanotra (2003) and Caplen et al. (2014). The apparatus was a clear plexiglass tank (Figure 6.1; $98 \times 48 \times 103$ cm; length \times width \times height) with a non-slip flooring and a plastic mesh partition that divided the test tank into 2 separate sections for simultaneously testing a pair of birds (Figure 6.1). A wood cover prevented the birds from flying out of the tank. Prior to testing, the container was filled with warm water (30 to 32°C) at a depth of 4 cm. Water temperature was measured prior to testing additional birds to maintain a similar temperature for all the birds tested. For each test, two focal birds (one male and one female) from the same pen were removed from their home pen, weighed, and simultaneously placed in the tank by two researchers, with the test starting when both birds were placed standing in the water. Because the birds were next to each other, removing one bird could affect the behaviour of the adjacent bird. Therefore, both birds were kept in the tank and continuously recorded using a digital video camera (Sony Digital High-Definition Video Camera; HDR-CX405 and DCR-SR68 models; Sony, Japan) throughout the 10-minute test. The time spent standing before lying down for the first time and the frequency of lying events per bird were recorded. Lying down was defined as a bird with its breast touching the water for at least 5 seconds, in which the last second of this 5-second period was considered the latency-to-lie.

As described by Caplen et al. (2014), some birds briefly sat but immediately stood up, suggesting they were initially unaware of presence of the water when placed in the tank. In the present study, these “dips” represented short lying periods (< 5 seconds) and were only considered to represent the bird’s ability to stand if three or more dips were observed within 20 s, with the third dip being considered as the latency-to-lie. The total number of dips were estimated but were not included in this chapter, as the interpretation of such behaviour would be challenging and may not reflect birds’ inability to stand, since birds quickly stood up after lying down in water. Thus, latency-to-lie was determined if birds laid down in water for at least 5 continuous seconds or if a bird laid down in water 3 consecutive times, with each dip lasting less than 5 seconds and the third dip being considered the latency-to-lie. These short 3-consecutive dips within 20 seconds were considered as one “lying event” per bird while each time the bird laid down in water for at least 5 continuous seconds was counted as one lying event per bird. Therefore, the total frequency of lying events per bird was determined as the sum of these two lying events, although only about 10% of birds tested performed consecutive dips in water (i.e. at least 3 dips of less than 5 seconds each within 20 seconds). If a bird did not lie down in water throughout the test, the latency-to-lie was considered to be 600 seconds, which corresponds to the maximum time and cut-off point of the test. The latency-to-lie and frequency of lying events were determined for each bird tested. The percentage of birds lying in water was determined on a pen basis for each sex, which included the 4 birds tested (2 males and 2 females), with the inclusion of sex as a main effect in the model for all the variables tested (refer to Section 6.3.6 for details about statistical models used).

6.3.3 Group Obstacle Test

The group obstacle test was conducted 3 to 7 days prior to processing and followed the methodology described by Caplen et al. (2014). For this test, a wooden barrier (160 × 9 × 10 cm; length × width × height; painted white) was placed alongside one wall in each pen 24 h before the test to habituate the birds to the presence of the new object, without preventing access to the feeder and drinker. Prior to the test, 6 birds per pen (3 males and 3 females) were individually marked and weighed. Four of these birds (2 males and 2 females) were the same birds used for the LTL test and bone quality assessment (Chapter 5), whereas the other 2 birds were part of the focal birds selected at hatch in order to represent a larger proportion of the pen. The LTL and group obstacle tests were conducted 4 to 7 days apart to allow the birds to recover, preventing a possible interaction between the tests. After the birds were weighed, the feeder was removed from each pen for a 1 h period to increase the birds' motivation to obtain access to feed at the beginning of the test. Birds had free access to water during this period. After completion of the 1-hr feed withdrawal (from the removal of feed of the last pen tested), one researcher used a board to corral the birds to the back of the pen, which contained the drinker line. The obstacle was placed horizontally across the pen, creating a barrier between the feeder and drinker (Figure 6.2). Therefore, the obstacle required the birds to step up and over it to reach the feed or water, which were located on opposite ends of the pen. The feeder was returned to the pen after placement of the obstacle. After the obstacle and feeder were placed into the pens, birds were continuously recorded for 5 h using a digital video camera (Sony Digital High Definition Video Camera) held by a monopod (Digiart MP-3606 Professional Video Monopod 70", Zhejiang, China) positioned in front of each pen and angled towards the obstacle to allow visualization of both sides (feeder and drinker). The

experimenter left the room after all the pens were set up (i.e., placement of feeders, obstacle, and video camera). The latency to first cross the obstacle and the total number of crossings (a combination of step-up and step-down towards the feeder or drinker side) per focal bird were tallied. Similar to variables evaluated in the LTL test, sex of the birds was included as a main effect in all the statistical models (refer to Section 3.3.6 for details about statistical models used).

6.3.4 Litter Moisture Content

On d 14, 28, 42, and 56, litter samples were collected from each pen to obtain a biweekly estimate of the litter moisture content. After each processing, litter was collected from the pens with birds remaining. Therefore, on d 14 and 28, litter was collected from all the pens, whereas on d 42 and 56, fewer pens were analyzed due to the different processing ages of FG and SG strains, as previously described. Because FG birds were processed at 34 d and 48 d, there are no data for this category at 56 d.

A square metal box ($10 \times 10 \times 10$ cm) was used to collect litter from five pre-determined locations in each pen, accounting for the left and right (front and back of the pen) and middle area, avoiding areas under the drinkers. The samples from the different locations were pooled and placed into sealed plastic bags identified with the pen number. Next, the samples were transferred to a dry bucket and thoroughly mixed until homogenized and a representative sample that ranged from 100 to 120 g was obtained and placed into pre-weighed aluminum containers. The total initial weight (litter + aluminum container) was recorded, the samples were dehydrated for 24 h at 65°C , then the final weight was recorded (Arrazola et al., 2019). Litter moisture content was estimated by dividing the water lost (difference between initial weight and final weight) by the initial weight.

6.3.5 Footpad Dermatitis and Hock Burns

One day prior to processing (33, 47, or 61 d), 22 birds per pen (11 males and 11 females) were assessed to determine the prevalence and severity of FPD and HB using a 5-point scale from 0 to 4, as described by the Welfare Quality Protocol[®] with 0 representing no lesions and 4 representing severe lesions. These birds included the 12 focal birds and 10 randomly selected birds (5 males and 5 females) to represent a large percentage (50%) of the pen. Because a low incidence of HB was observed in pilot studies and the first three trials, the assessment of HB was only conducted in trials 4 to 8. Therefore, strains D, H, and M were not represented in the HB evaluation, because these strains were only tested in the first three trials.

Both right and left feet and hocks were evaluated, and the highest score of the two was recorded. For data analyses, scores were categorized as 0- no lesions (score 0), 1-mild lesions (scores 1 and 2 combined) and 2-severe lesions (scores 3 and 4 combined). This classification was adapted from the National Chicken Council (2017), in which lesions covering less than 50% of the footpad (scores 1 and 2 in our scoring system) are classified as “pass” or acceptable, while lesions covering more than 50% of the footpad (scores 3 and 4 in our scoring system) are classified as “fail” or unacceptable. A similar classification was used for HB lesions, where scores 0 to 5 were categorized as to 0- no lesions, 1-mild lesions (red/light brown and superficial lesions) and 2-severe lesions (black and/or deep ulcers). The total incidence of birds exhibiting any FPD and HB and the incidence of birds exhibiting severe lesions (scores 3 and 4 combined) of FPD and HB were determined on a pen basis and reported as a percentage of total birds evaluated.

6.3.6 Statistical Analyses

To facilitate analysis, strains were categorized into four groups based on their realized growth rate to TW 2 (48 d for FG and 62 d for SG strains, respectively, Table 3.1, previously published by Torrey et al., 2021). The 14 strains were categorized as conventional (CONV; strains B and C; $ADG_{0-48} = 66.0$ to 68.7 g/d), fastest slow-growing (FAST; strains F, G, I, and M; $ADG_{0-62} = 53.5$ to 55.5 g/d), moderate slow-growing (MOD; strains E, H, O, and S; $ADG_{0-62} = 50.2$ to 51.2 g/d) and slowest slow-growing (SLOW; strains D, J, K, and N; $ADG_{0-62} = 43.6$ to 47.7 g/d). Comparisons among and within categories were conducted for all variables analyzed to assess differences among strains differing in growth rates and to compare strains with similar growth rates, respectively. Data were analyzed as an incomplete block design, using generalized linear mixed models (GLIMMIX) in SAS[®], version 9.4 (SAS Institute Inc., Cary, NC), with pen considered as the experimental unit. The random effects for all models included trial (i.e., each of the 8 production cycles) and block nested within trial. For all of the variables analyzed, except litter moisture, two models were used to assess the differences at a similar BW (*TW model*) and similar age (*Age model*). The main effects of the TW model included category, strain nested within category, TW, and sex. The interactions between category \times TW, category \times sex, category \times sex \times TW, strain (category) \times TW, strain (category) \times sex, and strain (category) \times sex \times TW were tested and included in the model if significant. This model allowed the determination of the effect of BW on leg strength (LTL test), number of obstacle crossings (group obstacle test), and contact dermatitis. The age model allowed the evaluation of all the strains at approximately 48 d, which corresponded to TW 1 and TW 2 for SG and FG strains, respectively. In the age model category,

strain (category), and sex were included as main effects. The interactions between category \times sex or strain (category) \times sex were tested and kept in the model if significant.

Because litter moisture was assessed at different ages, the day of collection (14, 28 and 42 d) was used as a repeated measure in the *Litter moisture by age model*. The interactions between category \times age and strain (category) \times age were included as main effects and kept in the model if significant. This model included an ARH (1) structure, selected based on fit statistics with the lowest Akaike information criterion value. Because CONV birds were processed at d 34 and 48 at TW 1 and TW 2, respectively, the litter moisture was determined at d 56 only in the remaining SG birds. Therefore, an additional model (*Litter moisture at d 56*), including category and strain (category) was used to investigate the litter moisture content at d 56 for SG stains.

Contrast statements were used to identify differences between categories and between strains within categories. For multiple comparisons, P-values were adjusted using Tukey adjustment. Residuals were checked for normality using quantile-quantile plots and the Shapiro-Wilk test. Linearity, randomness, and homogeneity of residuals were assessed using scatterplots and boxplots of studentized residuals. Residual analyses were used to select the most appropriate model that met all the model assumptions. The Gaussian distribution was used by default if all of the model assumptions were met. For the total incidence of FPD (TW model), the number of lying events in the LTL test (TW model), and latency to cross the obstacle (TW and age models), binary, Poisson, and lognormal distributions were used, respectively, to meet the model assumptions. The main effects of sex and TW are not included in data tables and figures, but are described with respective P-values in the results section if significant.

Due to the potential impacts of contact dermatitis on the variables evaluated in the LTL and group obstacle tests (Caplen et al., 2014), Spearman's rank correlation coefficients were used to investigate the relationships between the severity of FPD and HB with the LTL and group obstacle tests for each category. In addition, the correlation between the LTL and group obstacle tests were also investigated to determine possible relationships between the tests for each category. Correlation coefficients were classified as weak ($r_s < |0.35|$), moderate ($r_s |0.35| \leq r_s < |0.67|$), or strong ($r_s \geq |0.68|$) (Bohrer et al., 2018). For all tests performed, statistical significance was considered at $P < 0.05$.

6.4 Results

Data are presented as differences among categories and among strains within categories at both TWs. At similar ages, only differences among categories are presented, as strains within the same category were processed at a similar age at both TWs. As expected, category and TW affected most of the variables evaluated, indicating the effects of both growth rate and BW on leg strength, leg health, and mobility.

Due to the large number of strains tested and lack of significant differences among strains within category for most of the variables evaluated, only differences among categories at both TWs are provided. Differences among categories for the LTL and group obstacle tests are shown in Table 6.1. At the same age, differences obtained in these tests are given in Table 6.2. The effect of category at each TW on the incidence and severity of skin lesions are provided in Figures 6.3- 6.6, while differences among categories at a similar age are given in Figures 6.7-6.9. The effects of category on litter moisture content measured biweekly throughout the trails are given in Figure 6.10.

Differences among strains within category are included in Appendix A.2 and described in the results section if trends or significant differences were found. Significant interactions between category, sex and TW are also provided in Appendix A.2. Differences between sex and TWs are described in the text if significant.

6.4.1 Differences among Categories, Strains, and Sexes at TW 1 and TW 2

6.4.1.1 Latency-to-Lie Test

6.4.1.1.1 Body Weight at LTL test

As expected, BW was heavier in birds evaluated at TW 2 compared to TW 1 ($P < 0.001$; TW 1 = $2,015 \pm 24.5$ g, TW 2 = $3,030 \pm 25.7$ g), which was consistent in all categories. Body weights differed by category at both TWs, with CONV and SLOW birds being lighter than FAST and MOD birds ($P < 0.001$). The FAST and MOD birds had differences in BW among strains (Appendix A.2). Among FAST birds, strain M was lighter ($P < 0.010$) than the other FAST strains at TW 1, while at TW 2 no difference in BW was observed. Among MOD birds, strain E was heavier ($P < 0.004$) than strain H at TW 1 and TW 2. Overall, males were heavier than females ($P < 0.001$; $2,777 \pm 21.5$ g vs. $2,267 \pm 22.7$ g). However, sex interacted with category ($P = 0.001$, Appendix A.2). At TW 1, for both females and males, CONV was lighter than MOD and FAST birds. However, at TW 2, CONV females were lighter than FAST females, yet similar to MOD females, whereas CONV and SLOW males were similar yet both categories were lighter than FAST and MOD males.

6.4.1.1.2 Latency-to-lie

Time spent standing in water before lying down (i.e., LTL) was longer at TW 1 ($P = 0.042$; 450.8 ± 11.96 s) than TW 2 (413.2 ± 12.78 s). However, TW interacted with category, indicating that

differences in LTL between categories depended on TW (Table 6.1; $P = 0.016$). At TW 1, CONV and SLOW birds remained standing longer than FAST birds. At TW 2, CONV and FAST birds had a shorter time standing in the water than SLOW birds, while MOD birds did not differ from the other categories at either TW. There was no difference in LTL among strains within category at either TW (all pairwise comparisons among strains within category $P > 0.171$ for both TWs). Overall, standing time in shallow water was shorter ($P < 0.001$) in males (373.9 ± 11.15 s) than in females (489.9 ± 12.83 s). However, there was a category \times TW \times sex interaction ($P = 0.001$; Appendix A.2). At TW 1, LTL was affected by sex in FAST and MOD categories, whereas at TW 2, all the categories were affected by sex, with males standing for less time in water than females. At both TW 1 and TW 2, no difference in LTL was observed between females across the different categories. At TW 1, CONV and SLOW males had greater values than FAST and MOD males, whereas at TW 2, SLOW males had greater values than males from the remaining categories.

6.4.1.1.3 Percentage of Birds that Laid Down in Water

Category interacted with TW to influence the percentage of birds lying down in water (Table 6.1; $P = 0.029$). At TW 1, similar percentages of FAST and MOD birds laid down in water and more FAST birds laid down than CONV and SLOW birds. At TW 2, there was a tendency for more CONV ($P = 0.067$) and FAST ($P = 0.083$) birds to lie down than SLOW birds. Despite the large numeric differences, the percentages of birds lying down in the water did not significantly differ among strains within the same category (all pairwise contrasts between strains within category $P > 0.581$ at both TWs). The percentage of birds lying down in the water was greater ($P < 0.001$) in males ($58.5 \pm 2.71\%$) compared to females ($33.53 \pm 2.95\%$). However, an interaction between sex, category, and TW affected the percentage of birds lying in water (Appendix A.2). There was no

difference in the percentage of birds lying in water among females, while among males there was an effect of category at both TWs. At TW 1, there were lower percentages of CONV and SLOW males lying down than FAST and MOD males, whereas at TW 2, a lower percentage of SLOW males laid down compared to males in the other categories.

6.4.1.1.4 Frequency of Lying Down Events per Bird

The interaction between category and TW affected the number of times the birds laid down in water (Table 6.1; $P = 0.047$). At TW 1, CONV and SLOW birds laid down in the water less often than FAST birds, while at TW 2, CONV and FAST birds laid down more times than SLOW. At both TWs, MOD birds did not differ from the remaining categories. At both TWs, there was no difference in the frequency of lying events among strains within category (all pairwise $P > 0.615$ for comparisons among strains within category at TW 1 and TW 2). Sex affected the number of times lying down in water, with males ($P < 0.001$; 1.07 ± 0.081 times per bird) lying down more often than females (0.56 ± 0.056 times per bird).

6.4.1.2 Obstacle test

6.4.1.2.1 Body Weight at Obstacle Test

As expected, birds tested at TW 1 ($P < 0.001$; $1,820 \pm 20.9$ g) were lighter than those tested at TW 2 ($2,719 \pm 21.0$ g). At TW 1, CONV and SLOW were lighter than FAST birds, while MOD birds were similar to CONV and FAST, yet heavier than SLOW birds ($P < 0.001$; Table 6.1). At TW 2, FAST birds had the heaviest BW, while CONV and MOD birds were similar and greater than SLOW birds. Within categories, MOD strains differed in BW at TW 1 (Appendix A.2), with strain E being heavier ($P = 0.008$) than strain S, while strains H and O did not differ from the remaining

MOD strains. Heavier ($P < 0.001$) BW in males ($2,469 \pm 16.9$ g) compared to females ($2,070 \pm 17.6$ g) was consistent in all categories and strains.

6.4.1.2.2 Frequency of Obstacle Crossings per Bird

Overall, birds evaluated at TW 1 crossed the obstacle more often ($P < 0.001$; 8.9 ± 0.32 per bird) compared to those tested at TW 2 (6.9 ± 0.32 per bird). Nevertheless, differences among categories depended on TW (Table 6.1; $P < 0.001$). At TW 1, SLOW birds made the greatest number of crossings, while no difference was observed among the other categories. At TW 2, CONV and FAST birds made fewer crossings than SLOW, whereas MOD birds did not differ from the other categories. There was no effect of strain within categories on the total number of obstacle crossings (all pairwise comparisons among strains within category $P > 0.602$ at both TWs). Sex affected the total number of obstacle crosses ($P = 0.028$), with males (8.4 ± 0.27 per bird) crossing more than females (7.4 ± 0.29 per bird), which was consistent among all the categories.

6.4.1.2.3 Latency to Cross Obstacle for the First Time

At TW 2 birds tended ($P = 0.093$) to have longer latency to cross the obstacle (5.9 ± 0.09 s) than at TW 1 (5.6 ± 0.09 s). Although CONV birds tended to have greater latency to cross the obstacle than SLOW birds ($P = 0.079$) at TW 2, category (Table 6.1) and strain within category ($P > 0.881$) did not influence the latency to cross the obstacle (Appendix A.2). Males had shorter latency to cross the obstacle than females ($P = 0.018$; 278.7 ± 24.19 s vs. 362.8 ± 33.75 s), with no interaction between sex and category ($P = 0.167$) or strain ($P = 0.270$).

6.4.1.3 Contact Dermatitis

The total incidence of FPD was affected by category (Figure 6.3; $P < 0.001$) at both TWs. At TW 1, CONV birds had a greater incidence of FPD than FAST and MOD birds, while SLOW birds did not differ from the other categories. At TW 2 CONV birds had the greatest incidence of FPD, with no difference observed among the SG categories. Despite the large numeric differences across strains within categories, the total incidence of FPD did not differ ($P > 0.05$ for all pairwise comparisons among strains within category at both TWs, Appendix A.2). Sex affected the incidence of FPD ($P < 0.001$), with fewer males ($45.4 \pm 3.04\%$) exhibiting FPD than females ($59.6 \pm 2.95\%$).

At TW 1, there was no difference in the incidence of severe FPD among categories. However, at TW 2, CONV had a greater incidence of severe FPD than FAST birds and tended to have greater incidence than MOD ($P = 0.060$) and SLOW birds ($P = 0.094$) (Figure 6.4). Within categories, there were no differences among strains at both TWs ($P > 0.227$ for all pairwise comparisons between strains within category at TW 1 and TW 2, Appendix A. 2). Sex ($P = 0.367$) and TW ($P = 0.243$) did not affect the incidence of severe FPD.

Total incidence of HB was affected by category ($P < 0.001$). However, category interacted with TW ($P = 0.011$) as shown in Figure 6.5. At TW 1, CONV and MOD categories exhibited a similar percentage of birds affected by HB, which was greater than SLOW, while FAST did not significantly differ from the other categories. At TW 2, CONV and FAST birds had greater incidence of HB than MOD and SLOW birds, which had similar values. Within categories, CONV strains had a different incidence of HB at TW 1, with strain B being greater than strain C ($P = 0.005$, Appendix A.2). At TW 2, no difference among strains within category were observed ($P >$

0.789 for all pairwise comparisons between strains within category at TW 2; Appendix A.2). The incidence of HB was affected by sex ($P < 0.001$), with more males ($25.5 \pm 2.87\%$) having HB than females ($13.0 \pm 0.017\%$).

Birds processed at TW 1 had lower incidence of severe HB than those processed at TW 2 ($P = 0.031$; $0.61 \pm 0.739\%$ vs. $2.82 \pm 0.739\%$). There was no effect of category ($P > 0.129$ for all pairwise comparison among categories at TW 1 and TW 2; Figure 6.6) or strain within category ($P > 0.970$ for all pairwise comparison among strains within category at TW 1 and TW 2; Appendix A.2) in the incidence of severe HB lesions at both TWs. However, sex influenced the incidence of severe HB ($P = 0.001$), with more males having severe HB than females ($2.57 \pm 0.571\%$ vs. $0.86 \pm 0.570\%$).

6.4.2 Effect of Category at a Similar Age

6.4.2.1 Latency-to-Lie and Group Obstacle Tests

Comparisons among categories at similar ages, which corresponds to 41 d and 46 d of age for the group obstacle and latency-to-lie tests, respectively, are described in Table 6.2. Category affected all the variables evaluated. The BW of birds differed among categories in both the LTL and group obstacle tests ($P < 0.001$), with CONV birds being heavier than the other categories, while FAST and MOD birds were similar and heavier than SLOW birds.

Category affected LTL ($P < 0.003$), with CONV and FAST birds having a shorter latency than SLOW birds. Similarly, a higher percentage of CONV and FAST birds laid down in water than SLOW birds ($P = 0.0012$). The frequency of lying events was greater ($P = 0.016$) in CONV than SLOW birds, while FAST and MOD birds did not differ from CONV and SLOW birds.

The CONV birds had the lowest total frequency of obstacle crossings, followed by FAST and MOD, which did not differ from each other and were lower than SLOW birds. The latency to first cross the obstacle was greater in CONV birds compared to FAST and SLOW birds ($P < 0.045$), while MOD birds did not significantly differ from the other categories.

Differences in the LTL test between males and females at a similar age were similar to the differences previously described in the comparisons among categories and strains at TW 1 and TW 2. Males were heavier ($P < 0.001$; $2,534 \pm 27.13$ g vs. $2,119 \pm 28.2$ g), had shorter LTL ($P = 0.001$; 375.2 ± 16.49 s vs. 447.0 ± 18.40 s), had a higher percentage of birds lying down in the water ($P = 0.005$; $58.7 \pm 3.98\%$ vs. $43.7 \pm 4.27\%$ s), and laid down in the water more times ($P = 0.027$; 1.29 ± 0.133 times vs. 0.89 ± 0.148 times) than females. However, when compared at a similar age, sex did not affect the frequency of obstacle crossings ($P = 0.167$; males = 8.5 ± 0.39 times vs. females = 7.9 ± 0.42 times). Males tended to take less time to first cross the obstacle than females at a similar age ($P = 0.071$; 293.6 ± 33.89 s vs. 386.9 ± 48.51 s). There was no interaction between sex and category or sex and strain for any variable evaluated ($P > 0.05$).

6.4.2.2 Contact Dermatitis

Category affected both FPD and HB when compared at a similar age. The CONV birds had a greater total incidence (Figure 6.7; $P = 0.004$) and severity (Figure 6.8; $P = 0.026$) of FPD compared to FAST birds, while MOD and SLOW birds were similar and did not differ from CONV and FAST birds. For HB, CONV birds had a greater total incidence (Figure 6.9; $P = 0.001$) compared to SLOW, whereas FAST and MOD birds did not differ from CONV and SLOW birds. Due to the low incidence of severe HB lesions in all categories (CONV: 4.16 %, FAST: 1.27%, MOD: 1.14%, SLOW: 0.01%), statistical analyses were not performed.

When compared at the same age, differences in contact dermatitis between the sexes followed a similar trend to the differences at the two TWs, with males exhibiting a lower total incidence of FPD than females ($P < 0.001$; $47.4 \pm 3.11\%$ vs. $58.3 \pm 3.21\%$), whereas sex did not affect the incidence of severe FPD ($P = 0.662$; males: $12.9 \pm 1.69\%$, females: $12.1 \pm 1.79\%$). For HB lesions, males had a greater ($P < 0.001$) total incidence than females ($32.1 \pm 3.05\%$ vs. $15.3 \pm 3.08\%$).

6.4.2.3 Litter Moisture Content

As shown in Figure 6.10, litter moisture content differed among categories at d 14 and 28, with CONV having higher moisture than the SG categories ($P < 0.003$). However, at d 42, there was no difference among categories in litter moisture content. No difference among SG categories was observed at d 56 (Figure 6.10). Litter moisture differed within category for FAST and SLOW birds at d 42, but not at other ages. Among FAST birds (Appendix A.2), strain M had lower litter moisture than strains G and I ($P < 0.022$) while strain F did not differ from the other FAST strains. Among SLOW strains (Appendix A.2), strain J had greater litter moisture content than strain D and K ($P < 0.001$), whereas strain N was similar to the remaining SLOW strains. Litter moisture did not differ among strains within category for CONV and MOD birds at any age evaluated.

6.4.3 Correlation among Contact Dermatitis, Latency-to-lie and Group Obstacle test

For all the categories, there was no correlation between FPD scores and time spent standing in the LTL test or FPD scores and the number of times lying down in the water ($P > 0.1226$ for all categories, data not shown). For the obstacle test, there was a negative correlation between FPD scores and latency to first cross the obstacle for CONV birds only ($P = 0.025$; $r_s = -0.264$) suggesting a decrease in latency to cross as the FPD scores increased. In addition, a positive correlation was found between FPD scores and BW for CONV birds ($P = 0.048$; $r_s = 0.233$). The

correlations between HB scores and the mobility tests were not consistent among the categories. While for CONV and SLOW birds there were no correlations between HB scores and any of the variables measured (data not shown), that was not the case for FAST and MOD birds. For FAST birds, there was a negative correlation between HB scores and time standing in water ($P = 0.010$; $r_s = -0.264$) and a positive correlation between HB scores and the number of times the birds laid down ($P = 0.004$; $r_s = 0.292$), which was also observed for MOD birds ($P = 0.004$; $r_s = 0.2929$).

A strong negative correlation was found between time standing in the water and the number of times the birds laid down in the water for all the categories in the LLT test ($P < 0.001$; $r_s > 0.85$ for all categories, data not shown), whereas in the group obstacle test there was a moderate negative correlation between latency to the first cross and total obstacle crossings ($P < 0.001$; $0.3863 < r_s < 0.6138$ for all categories, data not shown). No correlation between the variables measured in the LTL and group obstacle tests were observed for all the categories.

6.5 Discussion

Numerous studies have suggested negative effects of growth rate and body mass on leg health and walking ability of different poultry species (Julian, 1998; Kestin et al., 2001; Shim et al., 2012a; Dixon, 2020; Rayner et al., 2020; Singh et al., 2021). However, in some studies walking ability was assessed when birds differing in growth rates had different BW (Kestin et al., 2001; Shim et al., 2012a, b), making it difficult to disentangle the effects of body mass versus genotypes selected for increased growth rate on locomotion. Therefore, the primary goal of this study was to investigate the differences in mobility, leg strength, and contact dermatitis among 14 strains of broiler chickens differing in growth rate when raised under similar rearing conditions and when those traits were compared at similar BWs and at a similar age. Many of the variables evaluated

in this study were affected by growth rate category and/or BW at a similar TW and age, indicating their effects on traits associated with locomotion. However, the differences in FPD and HB lesions may be more associated with environmental conditions than growth rate, suggesting the importance of maintaining good litter quality to prevent the occurrence of contact dermatitis in both FG and SG birds.

The LTL and group obstacle tests used in the present study are considered mobility tests that have been shown to be correlated with gait score. However, gait and mobility have different definitions. Gait is defined as “a manner of walking, or pattern of steps at a particular speed”, while mobility is defined as “the ability to move or be moved freely and easily” (Oxford Dictionary, 2020). As such, the tests performed in this study were used to indirectly assess birds’ leg strength and walking ability mainly by determining the time spent standing in water before lying down in the LLT test and total obstacle crossings in the group obstacle test, respectively. Therefore, differences among categories in these mobility tests were used to estimate possible differences in gait score and locomotion.

6.5.1 Latency-to-Lie Test

The gait score system is widely used to assess walking ability and lameness in broiler chickens (Kestin et al., 1992; Garner et al., 2002; RSPCA, 2017). However, some of the scores require the observers to compare a bird’s walking ability to ‘normal gait’. Because our study encompassed strains that differed in BW (Torrey et al., 2021), muscle size (Chapter 4), bone length (Chapter 5), and behaviour (Dawson et al., 2021), differences among the strains in gait may not necessarily reflect their walking ability. Therefore, other validated tests of mobility and leg strength were used in this study.

The LTL test is an alternative and validated method to assess lameness in broiler chickens. The time spent standing before the bird first lies down in shallow water has been associated with gait score, with birds that exhibit higher (poorer) gait scores lying down in the water sooner compared to birds with lower gait scores, likely due to pain or difficulty of lame birds to support their BW (Weeks et al, 2002; Caplen et al., 2014).

6.5.1.1 Effect of Category at Each TW

At TW 1, CONV and SLOW birds remained standing in the water for longer and had the lowest percentage of birds lying down and frequency of times lying down in water. These categories were also the lightest birds at TW 1, suggesting that differences in BW contributed more to performance in the LTL than did genetic potential for growth rate. The lower BW of CONV birds despite their greater growth performance was mainly due to the processing age that occurred earlier than the expected time for CONV strains to reach 2.1 kg (refer to Chapter 4). On the other hand, the lower BW of SLOW birds is attributed to their reduced growth rate ($ADG < 50$ g/d) compared to the other categories (Torrey et al., 2021). The longer LTL down in water at TW 1 in CONV and SLOW birds indicates a better ability to support their BW (i.e., leg strength) compared to FAST birds. These results differ from those recently reported by Dixon (2020), who found a greater proportion of lower gait scores in SG birds, suggesting better walking ability in this group compared to 3 FG strains when birds were evaluated at 2.2 and 2.5 kg. Kestin et al. (2001) reported better walking ability (as measured by gait scores) in SG strains compared to FG strains when the birds were fed a similar diet. However, the same authors found that the effect of genotype disappeared when differences in BW were considered; this suggests that the variation in gait score in that study was mainly attributed to the differences in BW, similar to results reported here

Although BW was likely the determining factor for the differences observed at TW 1, this was not the case at TW 2. At the heavier TW, CONV and SLOW birds were lighter than MOD and FAST birds, yet CONV and FAST birds spent less time standing and had a greater number of times lying down compared to SLOW birds. While the differences between FAST and SLOW birds may be attributed to the heavier BW of the former, SLOW and CONV birds had a similar BW at TW 2. Therefore, the differences in the LTL test between CONV and SLOW birds at TW 2 appear related to genetic potential for growth rates. This suggests that as the birds grow, the differences in leg strength between SG and FG genotypes may become more evident even when the birds are evaluated at a similar BW, indicating a negative effect of accelerated growth rate and potentially an effect of body conformation on leg strength. These results agree with other studies, in which SG birds showed better walking ability (as measured by gait score) than FG birds even when the evaluations were performed at a similar BW (Corr et al., 2003b; Dixon, 2020; Rayner et al., 2020) or when differences in BW were taken into account (Kestin et al., 1999). A recent study by Singh et al. (2021) found shorter standing time during the LTL test in FG compared to SG birds when both strains were evaluated between 2.0 - 2.2 kg. The researchers suggested that this difference found in the LTL test could be attributed to the larger breast muscle of FG birds that may increase sternal mass and load, which consequently increases the metabolic costs associated with prolonged standing (Tickle et al., 2018), leading to a decrease in time standing for FG birds.

Although the LTL has been found to be associated with gait score (Weeks et al., 2002; Caplen et al., 2014) and validated by use of analgesics (Hothersall et al., 2016), the test relies on the notion that birds find sitting in water aversive. Aversion to water has also been used in behavioural tests with broiler breeders (Dixon et al., 2014). However, chickens' aversion to sitting in water *per se*

has never been directly validated. In a modified LTL test, in which no water was used and birds' spontaneous LLT was measured in their home pens, Bailie et al. (2013) and Norring et al. (2019) reported much shorter LTL (< 25 seconds) than those observed by Weeks et al. (2002), Caplen et al. (2014), and Berg and Sanotra (2003), in which birds' LTL were as long as 600 to 900 seconds when water was used to increase birds' motivation to stand. These differences in LTL obtained between the modified (no addition of water) and traditional LTL test (addition of water) suggest that water is likely considered an aversive stimulus to birds. Nevertheless, because these results were obtained from different studies, birds individual' aversion to water was not assessed. Furthermore, these studies only tested FG birds. As such, differences in aversion to water in the LLT test among strains differing in growth rate is unknown and should be further investigated. Just as there may be genetic differences in fearfulness (e.g., differences in tonic immobility between a SG and a FG strain: Lindholm et al.(2017); differences in ranging behaviour and tonic immobility among genotypes with distinct growth rates: Castellini et al. (2016)), there could also be genetic variation in aversion to sitting in water.

6.5.1.2 Effect of TW

The relationships between BW and leg health have been well documented (Weeks et al., 2000; Sørensen et al., 2000; Kestin et al., 2001; Nääs et al., 2010; Wilhelmsson et al., 2019; Dixon, 2020). From TW 1 to TW 2, CONV strains showed a significant increase in percentage of birds lying down in water, shorter time standing, and tended to exhibit a greater number of times lying down in water. No significant changes or trends were observed for the SG categories as the birds grew from TW 1 to TW 2, despite the significant increase in BW in all categories. These results suggest that the detrimental effects of increasing age and BW were more evident in CONV birds,

indicating potential negative effects of selection for growth on leg strength. However, because FG and SG birds were evaluated at different ages at TW 1 and TW 2, age-related changes in behaviour and aversion to water should not be discarded. Furthermore, CONV birds had an increase in BW from TW 1 to TW 2 that was 14.85 to 17.70% greater than the remaining categories. Therefore, the sharp increase in BW observed in CONV birds from TW 1 to TW 2 may have exacerbated the differences obtained in the LTL as the birds grew.

6.5.1.3 Differences among Categories at a Similar Age

To accommodate other behavioural tests conducted in this study the LTL test was performed at approximately 46 d, which corresponds to couple days prior to TW 1 and TW 2 for SG and CONV strains, respectively. As expected, CONV birds were heavier than the remaining categories. Despite the differences in BW, the time spent standing before lying down, percentage of birds lying down, and the number of lying events in the LTL test was similar in CONV, FAST, and MOD birds, while CONV birds significantly differed from SLOW birds, indicating better leg strength in the latter. It is important to mention that SLOW birds were over 1 kg lighter than CONV birds at the same age, which represents a 55.13% difference. The differences between CONV and SLOW in the LTL test agree with many studies that suggest negative effects of increased BW on leg health (Kestin et al., 2001; Shim et al., 2012b; Dixon, 2020). However, our results suggest that at the same age, the effects of growth rate and increased BW on leg strength, indicated by the birds' ability to stand in the LTL test, were only apparent when there were large differences in BW, since CONV birds performed similarly to FAST and MOD birds.

6.5.1.4 Effect of Sex

Males had worse indicators of leg strength than females as demonstrated by their shorter time standing in the LTL test, greater percentage of birds lying down in the water, and more times lying in the water. These results are corroborated by other studies that indicate poorer walking ability and welfare in males (Sanotra et al., 2001; Dixon, 2020), most likely because males have a faster growth rate and greater BW than females (Dixon, 2020).

6.5.1.5 Effect of Strain

During the LTL test, both CONV strains (B and C) had an increasing percentage of birds lying down and number of times lying down in the water as well as a decreasing time standing from TW 1 to TW 2. However, this pattern was not observed for some SG strains. As an example, strains I and M (FAST), E, H, and S (MOD), and D and K (SLOW) showed a slight increase in time standing in water from TW 1 to TW 2. These results suggest that the increase in BW may not have obvious negative effects on leg strength assessed via LTL test for some strains. Alternatively, as previously discussed, this may indicate differences in aversion to water among strains. Despite the large numeric differences among strains within category, the LTL, percentage of birds lying in water, and frequency of lying events did not differ statistically, probably due to the small sample size and large variation among strains, leading to low statistical power ($1-\beta < 0.65$).

6.5.2 Group Obstacle Test

6.5.2.1 Effect of Category at each TW

The group obstacle test assesses birds' mobility by determining their ability to cross an obstacle to obtain access to food or water. For lame birds, both standing and moving may be associated with discomfort and or pain (Weeks et al., 2000; Hothersall et al., 2016). Although no significant

difference in latency to cross the obstacle was observed among the categories at both TWs, SLOW birds had the lowest BW and greater number of obstacle crossings, suggesting that the differences in BW influenced the ability to cross the obstacle among the categories, but not the initial motivation to cross. In fact, differences in total obstacle crossings were only observed between SLOW and the other categories, despite differences in BW and growth rate among categories.

Because SLOW birds were lighter than the remaining categories and also had the slowest growth rate, it is unknown to which extent the differences in BW and genetic potential for growth caused the differences in total obstacle crossings between SLOW and other categories. While the number of total obstacle crossings at TW 2 appeared to be linearly related to growth rate, CONV, FAST, and MOD did not differ from each other, while SLOW was greater than CONV and FAST birds. Interestingly, despite the lighter BW of CONV birds compared to FAST birds at both TWs, CONV and FAST birds did not differ in total number of obstacle crossings. These results partially complement the observations obtained in the LTL test, supporting the hypothesis that selection for accelerated and early muscle accretion may have negative effects on mobility in broiler chickens.

The lower number of crossings observed in FG birds may be due to their body conformation (including large breast muscles and short legs; see Chapters 4 and 5, respectively) requiring an altered gait to increase their stability while walking (Paxton et al., 2014). Corr et al. (2003b) suggested that the gait alterations observed in FG birds are inefficient, rapidly tiring the birds and leading to low activity levels. The abnormal gait seen in FG birds may be a result of pain (Corr et al., 1998; McGeown, 1999; Caplen et al., 2013), biomechanical issues related to body conformation, or both (Corr et al., 2003b). The substantial increase in breast muscle observed in FG birds has moved the centre of gravity cranially (Corr et al., 2003a, b; Paxton et al., 2014). In

fact, it has been demonstrated that the centre of gravity of FG birds shifts from caudodorsal to craniodorsal between 28 and 42 d, likely as a result of the substantial breast muscle growth in this period, resulting in an increase in muscle forces required to balance it. However, in Giant Junglefowl, a wild progenitor population of modern chickens, the centre of gravity was only reported to move caudodorsally across ontogeny (Paxton et al., 2010, 2014). This change in body conformation observed in FG birds, leading to alterations in forces involved while walking (Corr et al., 1998) and the rapid muscle accretion on the immature skeleton are known to affect locomotion (Corr et al., 2003b). Despite the differences found in the LTL and group obstacle tests between CONV and SLOW birds, CONV birds had similar or greater tibial breaking strength and ash content compared to SG birds (Chapter 5). Therefore, it seems likely that the differences observed in both mobility tests were not influenced by differences in bone traits among categories.

There are other plausible reasons why categories may have differed in the number of obstacle crossings. It is possible that differences in the obstacle test may reflect differences in feeding strategies. Strains selected for differences in feed intake may have differences in feeding behaviour related to feeder visits and meal sizes. One study comparing four genetic lines differing in the intensity of selection for growth reported no change in hunger and satiety mechanisms that control feeding behaviour (Howie et al., 2009). Nevertheless, the organization of feeding behaviour differed among lines, with slightly fewer but larger and longer meals in the FG lines compared to the SG lines (Howie et al., 2009). Conversely, Barbato et al. (1980) reported that at all ages investigated, chicks from a heavy line had more meals, but a similar amount of feed consumed per meal compared to chicks from light lines.

As part of this larger research project, a study was conducted to investigate behavioural differences among the strains, including differences in feeding and drinking behaviour (Dawson et al., 2021). While CONV birds spent more time eating, no difference in feeding or drinking bouts was found between the CONV and SG birds, suggesting that the categories and strains evaluated in our study organized their feeding similarly. Therefore, it is unlikely that differences in feeding strategies and behaviour influenced total obstacle crossings. Weeks et al. (2000) reported significant changes in feeding behaviour in lame birds, with a decrease in visits to the feeder but increased duration of feeding per bout compared to sound birds, resulting in a similar total time spent feeding per day, despite the differences in feeding strategies. These behaviour changes caused by lameness reinforce that lame birds have more difficulty standing and walking to access resources. Nevertheless, it is unclear if the differences in total obstacle crossings between SLOW and other categories in our study occurred in response to lameness. Further work should be conducted to investigate feeding behaviour with and without the obstacle when birds differing in growth rate reach a similar BW to determine whether the total number of crossings are affected by differences in walking ability rather than by differences in feeding strategies, age, or BW.

Decreases in locomotor activity (Weeks et al., 2000; Bokkers and Koene, 2003) and walking ability (Nääs et al., 2010; Norring et al., 2019) occur as broilers age. However, even though SG birds were 2 wk older than CONV birds at both TWs, this difference in age among categories did not seem to play a role in total obstacle crossings. Interestingly, Dawson et al. (2021) also revealed that at a similar age, strains with faster growth rates were more inactive compared to those with slower growth rates. While inactivity reached 78 - 80% for all strains, this level was reached earlier by strains with faster growth rates. Fast growth rate was also associated with more time spent

sitting, and less time spent standing and walking when compared at similar age. Furthermore, at TW 2, faster growing birds had a lower proportion of birds using all the enrichments and accessing the platforms (Dawson et al., 2021). The positive effects of locomotor activity and to a lesser extent environmental enrichment on leg health and walking ability, have been demonstrated in several studies (Shipov et al., 2010; Ruiz-Feria et al., 2014; Pedersen et al., 2020; Pulcini et al., 2021). Therefore, it is possible that such differences in behaviour reported by Dawson et al. (2021) may have influenced birds' walking ability as indicated by differences in total obstacle crossings among categories.

6.5.2.2 Effect of TW

From TW 1 to TW 2, there was a lower number of total obstacle crossings, which was consistent for all the categories. These results agree with Kestin et al. (2001), who compared birds of various genotypes differing in growth rate and found a consistent decrease in walking ability with increasing age and BW.

6.5.2.3 Differences among Categories at a Similar Age

The group obstacle was performed at approximately 41 d for all categories, which corresponds to one week prior to TW 1 and TW 2 for SG and CONV strains, respectively. CONV birds were heavier than the remaining categories at a similar age. The increase in BW among categories was accompanied by a significant decrease in total obstacle crossings, with CONV birds crossing fewer times than the remaining categories and FAST and MOD birds being similar but crossing fewer times than SLOW birds. The BW followed an opposite trend, with the greatest and lowest values observed in CONV and SLOW birds, respectively, while MOD and FAST birds were intermediate, which confirms the deleterious effects of the increase in BW on mobility.

The CONV birds had a similar latency to cross the obstacle compared to MOD birds, but greater than FAST and SLOW birds. Because CONV birds were heavier and had the greatest feed intake at d 48 (Torrey et al., 2021), it is expected that CONV birds would be equally or more motivated to eat than other birds, suggesting that a greater latency to cross the obstacle was associated with lack of ability to cross rather than lack of motivation to obtain access to the feeder. However, because MOD birds showed a latency to cross that did not differ from the other categories despite the differences in BW and total obstacle crossings, differences in motivation could also contribute to the latency to cross the obstacle. In a study conducted to determine motivation and physical ability to walk for a food reward in FG and SG strains, Bokkers and Koene (2004) revealed that, following a similar feed deprivation period (likely leading to a similar motivation to access the food), SG birds had a shorter latency to start to walk and walked faster down a runway compared to FG birds. The authors concluded that while motivation is the determining factor for walking in lighter birds, physical ability plays a major role in the walking ability of heavy birds.

6.5.2.4 Effect of Sex

Although the increase in BW across categories was associated with fewer obstacle crossings, males were heavier yet crossed the obstacle more often than females, likely due to the higher feed intake of males (Marks, 1985; Benyi et al., 2015) and possibly differences in feeding strategies. In fact, when analyzing behavioural differences among categories and sexes Dawson et al. (2021) observed that sex affected time spent feeding at d 56, with males spending more time feeding than females. Because differences in obstacle crossing were only performed by TW and not by different ages in this present study, it is unknown if there are differences between sexes across ages. However, no interaction between sex and target weight was observed,

indicating the greater number of total obstacle crossings in males compared to females was consistent as the birds grew from TW 1 to TW 2.

6.5.2.5 Effect of Strain

Strains within category did not differ in total obstacle crossing or latency to cross the object, suggesting similar feeding strategies in strains selected for similar growth rates. All strains showed a numeric decrease in total crossings in the obstacle test from TW 1 to TW 2, except strain E (MOD), which had a slight increase. Others have reported a decrease in locomotor activity and walking ability as birds grow, which occurs in both FG and SG strains despite the differences in selection emphasis for growth (Kestin et al., 2001; Bokkers et al., 2003).

6.5.3 Contact Dermatitis and Litter Moisture

The relationship between walking ability (measured by alterations in gait score or mobility tests) and contact dermatitis has been investigated in previous studies (Martland, 1984; Caplen et al., 2014; Granquist et al., 2019). While the lack of activity and prolonged time sitting on poor quality litter can cause or aggravate skin lesions (Hester, 1994; Weeks et al., 2000), the incidence of contact dermatitis can also affect birds' mobility. For example, when determining the factors that explain the variation in the group obstacle and LTL tests, Caplen et al. (2014) reported that lameness, body mass, and HB were significant explanatory variables for the latency to first cross the obstacle, while lameness and FPD were predictors that accounted for the majority of the variance observed in the LTL test, although sex, body mass, and HB also had a significant relationship with time spent standing in the LTL test. In addition, the likelihood of lying in the LTL test was increased by lameness and FPD, indicating the effects of both variables on the traits

measured in Caplen's study (2014). Thus, the presence and severity of contact dermatitis may affect birds' leg health and influence the variables measured in the LTL and group obstacle tests.

6.5.3.1 Effect of Category at each TW

At TW 1, CONV birds had a similar prevalence of FPD compared to SLOW birds but greater than FAST and MOD birds, whereas at TW 2 CONV birds had a greater prevalence than the remaining categories. Litter quality is the main contributing factor for the occurrence and severity of FPD, with sudden deterioration of litter condition and prolonged contact with poor quality litter increasing the susceptibility of skin lesions (Allain et al., 2009; Shepherd and Fairchild, 2010)). To support their rapid growth, FG strains had higher feed intake than SG strains (Torrey et al, 2021), which would result in a higher amount of excreta. This most likely accelerated the increase in litter moisture in CONV birds' pens. At 14 and 28 d, CONV had higher litter moisture than SG pens. However, a similar litter moisture content was observed at 42 d, which was within one week before CONV birds reached TW 2. Although FPD was only assessed prior to processing, the onset of FPD has been found to occur as early as 2 wk of age in FG strains of broiler chickens, with an increase in the incidence observed as the birds grow (Kjaer et al., 2006).

The similar incidence of FPD between CONV and SLOW birds observed at TW 1 was unexpected, as a lower incidence of FPD has been reported in SG strains in other studies (Kjaer et al., 2006; Allain et al., 2009; Dixon, 2020). However, CONV had a greater incidence of FPD compared to FAST and MOD birds at TW 1. Litter moisture content measured the week prior to TW 1 for CONV (28 d) and SG categories (42 d) was similar, suggesting that the higher incidence of FPD in CONV birds compared to FAST and MOD birds cannot be solely attributed to differences in absolute values of litter moisture on a pen basis. The SLOW birds had the lowest ADG and average

daily feed intake amongst the categories (Torrey et al., 2021). Therefore, drier litter and lower incidences of FPD were expected. However, SLOW birds were observed perching on the waterline, which may have contributed to higher moisture content close to the drinking area. Because samples from different locations of the pens were pooled, the differences in moisture content in distinct areas of the pen between categories were not determined.

Litter quality may differ according to pen location, as demonstrated by Wilhelmsson et al. (2019), who found poorer litter quality in the feeding area in pens with FG birds compared to SG birds. Considering the heterogeneity of litter quality in the pen and the differences in behaviour and resource use among the different strains (Dawson et al., 2021), some birds may have spent a considerable amount of time close to the drinking area to obtain access to water and use the platform, both located in the back area of the pen. Thus, if this area had high moisture content likely because of the perching behaviour on the waterline, it may have favoured the incidence of FPD in SLOW birds.

The higher incidence of FPD in CONV birds at TW 2 is in line with earlier studies (Kjaer et al., 2006; Allain et al., 2009; Dixon, 2020). Even though BW has not been shown to affect FPD (Kjaer et al., 2006), the higher susceptibility of FG strains to develop FPD has some possible explanations. Kjaer et al. (2006) suggested that differences in FPD among lines may be partially attributed to genetic variation in biotin deposition, due to the role of this vitamin in skin integrity. In fact, skin strength has been shown to be genetically controlled (Kafri et al., 1984). In addition, the heavy BW combined with the lower activity of FG birds and prolonged time sitting may increase the pressure on the footpads, hocks, and breast, increasing the occurrence of contact dermatitis (Mayne, 2005).

The high incidence of FPD found in CONV birds in our study should be interpreted cautiously due to the high litter moisture content, observed throughout the trials. A precise definition of “wet litter” states that when litter moisture is greater than 25% (250 g/kg), its quality properties (e.g., cushioning, insulating, and water holding capacity) are compromised (Collett, 2012). In our study, litter moisture exceeded 30% from 28 d onwards for all categories. Litter moisture is a combination of water from spillage, condensation, and excretion (Collet, 2012). Although the amount of excreta produced at each TW likely differed among categories due to differences in feed intake, the consistent high litter moisture observed from 28 d is an indicator of suboptimal environmental conditions. While the solid walls between the pens in the current study prevented visual contact of adjacent pens, this design may have hindered air circulation at the pen level. As demonstrated in several studies, house design and ventilation system are important for maintaining good litter quality due to its influences on temperature, humidity, and airflow (Dawkins et al., 2004; Collett, 2012; Farhadi et al., 2016; Dunlop et al., 2016). It is possible that the standardized housing conditions in our study may have affected FG birds to a greater extent compared to SG birds due to their fast growth and greater amount of excreta produced at an earlier age compared to the other categories (Collett, 2012).

Despite the differences in the total incidence of FPD at TW 1, no difference in the severity of FPD was observed among categories. However, as previously mentioned, CONV birds had the lightest BW at TW 1. Therefore, differences observed at TW 1 may not accurately represent the influences of selection for growth on litter conditions and FPD. At TW 2, CONV had a greater incidence of severe FPD than FAST but was similar to MOD and SLOW. The reason for this difference is unclear. The litter moisture content the week before TW 2 (42 and 56 d for CONV and SG,

respectively) did not differ. However, as mentioned above, differences in litter quality in distinct locations of the pen may have influenced the results.

The incidence of HB was greater in CONV and MOD compared to SLOW birds at TW 1. However, CONV and MOD did not differ from FAST birds. This result was not expected as the increase in BW is commonly associated with a greater incidence of HB (Kjaer et al., 2006), which occurs as a result of the prolonged time sitting and decrease in the locomotor activity commonly observed as the BW increases (Bokkers and Koene, 2003). The overall lower BW of CONV birds at TW 1 (as determined on a pen basis prior to processing, Torrey et al., 2021) may explain the lack of difference between CONV, FAST and MOD at the time of the evaluation at TW 1. However, an effect of BW on the incidence of HB was observed at TW 2, when CONV and FAST were heavier at processing (Torrey et al., 2021) and also had greater incidence of HB compared to MOD and SLOW birds.

Overall, the incidence of severe HB was low at both TWs with no significant difference observed among the categories, a finding that differs from other studies (Kjaer et al., 2006; Dixon, 2020; Rayner et al., 2020). A possible explanation for this divergence in severity of HB could be the scoring systems used. While the present study classified the HB into a 3-point scale (no lesion, mild, and severe), other studies recorded 4 (Dixon, 2020) to 9 (Allain et al., 2009) different categories. Alternatively, the conditions in which the birds were reared may have contributed to the differences among studies. In Dixon's study (2020), wood shavings were added as needed to maintain dry and friable litter. However, in our study, bedding material was only replaced at the beginning of each trial. While this practice was adopted to simulate commercial broiler houses where litter material is commonly only replaced at the beginning of a production cycle, the

suboptimal conditions of the room in our study may have precluded finding any differences in severity of HB among categories, as HB is influenced by litter moisture (Mayne, 2005).

6.5.3.2 Effect of TW

Target weight did not affect the incidence or severity of FPD and HB, contrary to other studies that demonstrated that these variables tend to increase as the birds grow (Kaukonen et al., 2016; Wilhelmsson et al., 2019; Dixon, 2020). The difference in results may be attributed to the conditions in which the birds were reared and experimental design. In each trial, half of the pens of each strain were processed at TW 1. Because CONV birds were processed at an earlier age (34 d) than the other categories, only pens of CONV birds were processed at 34 d. At 48 d, TW 1 and TW 2 for SG and CONV birds respectively, the remaining pens containing CONV birds and half of the pens containing SG birds were processed. Thus, from TW 1 to TW 2, there was likely an improvement in environmental conditions in the room, due to the lower number of birds present, despite the increase in BW of the remaining birds. Because the majority of pens were no longer in the room from TW 1 to TW 2 for SG strains, it is possible that the changes in environmental conditions were more evident for these birds, as demonstrated by the slight decrease in litter moisture in all SG categories from d 42 to d 56. Drying out litter or transferring birds from wet to dry litter improves contact dermatitis (Greene et al., 1985; Martland, 1985). Therefore, the changes in litter conditions due to the reduced number of pens and birds may have contributed to the lack of differences in contact dermatitis from TW 1 to TW 2.

6.5.3.3 Differences among Categories at a Similar Age

At d 48, CONV birds only differed from FAST in incidence and severity of FPD, despite similar litter moisture content among categories at 42 d, suggesting that factors other than absolute litter

moisture may have influenced FPD at a similar age. At a similar age, CONV birds had a greater incidence of HB than SLOW birds, while no significant differences were found between CONV, FAST, and MOD birds. The differences between CONV and SLOW birds are congruent with other studies in which a greater incidence and severity of HB was observed in FG compared to SG birds (Bokkers and Koene, 2003; Dixon, 2020; Rayner et al., 2020). However, similar to the results obtained in the LTL test at a similar age, the differences in HB observed in the current study suggest that the negative effects of BW on HB may be more obvious when comparing strains greatly differing in growth rate and BW.

6.5.3.4 Effect of Sex

The incidence of HB, which relates to both BW and time spent sitting, was greater in males than females. However, females had a greater incidence of FPD than males. Although the relationship between FPD and sex is unclear, the greater incidence of FPD in females has been attributed to the differences in skin integrity between females and males. Females' skin contains higher fat and less protein and collagen content than males, suggesting that females' skin is more susceptible to tear than male skin, since the protein matrix is less dense and more prone to damage (Kamyab, 2001; Mayne, 2005).

6.5.3.5 Effect of Strain

Few significant or consistent differences were found among strains within categories, which may be due to the large variation or the lack of statistical power, which was lower than 0.700 for most of the variables evaluated, preventing the detection of differences among strains. The two CONV strains differed in HB incidence at TW 1 but not TW 2. Among FAST strains, strain M had lower litter moisture content at d 42 and slightly lower incidence of severe FPD than the other FAST

strains. The SLOW strains differed in litter moisture content on d 42, which reflected the incidence of FPD at both target weights, though this was not statistically significant. These results indicate differences in litter quality among strains selected for a similar growth rate, which may have influenced the incidence and severity of contact dermatitis.

6.5.4 Correlations Analyses

6.5.4.1 Mobility Tests and Contact Dermatitis

Overall weak correlations were observed between the severity of contact dermatitis and the outcome variables obtained in the LTL and group obstacle test. The negative correlation between FPD scores and the latency to cross the obstacle in CONV birds was likely due to the higher motivation of heavier birds to cross the obstacle and obtain access to the feeder as the FPD scores were associated with heavier BW in CONV birds. The increase in HB scores positively correlated with the percentage of birds lying down in water for FAST and MOD birds, suggesting that the increase in HB scores may have influenced the ability of MOD and FAST birds to remain standing in the water. However, because these correlations were weak ($r_s < |0.35|$; Bohrer et al., 2018) and inconsistent among categories, contact dermatitis was most likely not the determining factor in differences observed among categories in the mobility tests. These findings agree with Berg and Sanotra (2003) and Ruiz-Feria et al. (2014), who found no correlation between the incidence FPD and LTL test.

6.5.4.2 Latency-to-lie and Group Obstacle Tests

Although both LTL and group obstacle tests have been associated with gait scores, they are assessing different aspects of mobility. Even though there were moderate to strong correlations in the variables measured in each test, no correlation between the LTL and group obstacle tests was

found for any category. While LTL test assesses birds' leg strength and ability to stand to avoid lying down in shallow water, the group obstacle test measures birds' ability to cross the obstacle (i.e., step on and off of the obstacle) and walk to obtain access to resources (food or water) placed on opposite sides of the pen. Therefore, both tests could be used as alternatives to the gait score assessment as suggested by Caplen et al. (2014), but they might indicate different factors associated with walking ability. Because the group obstacle test is associated with a more complex task and requires the birds to cross the obstacle and walk to reach drinker or feeder, this test may better represent differences in walking ability. However, the differences in feeding motivation and feeding strategies should be considered when comparing strains differing in growth rate.

6.6 Conclusion

The effect of both growth rate and BW were observed in most of the variables investigated in this study. While at TW 1, the greater latency to lie down was associated with lower BW, total obstacle crossings per bird were affected by both BW and growth rate. However, at TW 2, growth rate, BW, and likely body conformation (i.e., larger breasts and shorter legs) affected the LTL and total obstacle crossings, with faster-growing birds showing indicators of poorer leg strength and mobility compared to slower-growing birds. Most of the variables investigated differed among categories at a similar age and between sexes. Overall, these differences were associated with differences in BW. Although some differences in contact dermatitis suggest the effects of selection for growth, most of the results indicate the relevance of good litter quality and moisture control as effective strategies to decrease the incidence of contact dermatitis in both FG and SG birds. Nevertheless, the incidence and severity of contact dermatitis most likely did not play a major role in the differences observed in the LTL and group obstacle tests, suggesting

the differences in indicators of leg strength and mobility obtained in the study were mainly affected by differences in BW, body conformation, and selection emphasis for growth.

Table 6.1: Effect of category on body weight (BW), latency-to-lie (LTL), and group obstacle test (LS means \pm SEM) in the week prior to Target Weights 1 and 2. At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 d of age, respectively.

Variable	Category			
	CONV	FAST	MOD	SLOW
Target Weight 1				
BW (g)- LTL test ¹	1,731 \pm 55.9 ^b	2,281 \pm 45.6 ^a	2,140 \pm 44.1 ^a	1,908 \pm 46.5 ^b
LTL (s) ²	499.9 \pm 31.19 ^a	390.9 \pm 23.01 ^b	410.8 \pm 21.38 ^{ab}	489.7 \pm 22.34 ^a
% of birds lying per pen	26.3 \pm 7.38 ^c	57.8 \pm 5.44 ^a	50.3 \pm 5.20 ^{ab}	35.1 \pm 5.32 ^{bc}
Lying events per bird	0.57 \pm 0.132 ^b	1.08 \pm 0.170 ^a	0.78 \pm 0.122 ^{ab}	0.54 \pm 0.100 ^b
Target Weight 2				
BW (g)- obstacle test ³	1,792 \pm 50.3 ^b	2,008 \pm 41.7 ^a	1,896 \pm 38.1 ^{ab}	1,585 \pm 34.9 ^c
No. of obstacle crossings ⁴	8.03 \pm 0.702 ^b	7.76 \pm 0.558 ^b	8.67 \pm 0.638 ^b	11.13 \pm 0.639 ^a
Latency to cross (s)	250.8 \pm 60.31	254.4 \pm 49.22	375.1 \pm 67.24	257.8 \pm 43.25
Target Weight 2				
BW (g)- LTL test	2,828 \pm 61.3 ^b	3,332 \pm 48.5 ^a	3,179 \pm 45.27 ^a	2,781 \pm 46.8 ^b
LTL (s) ⁵	350.1 \pm 32.25 ^b	378.2 \pm 24.18 ^b	422.4 \pm 22.96 ^{ab}	479.5 \pm 22.88 ^a
% of birds lying per pen	59.2 \pm 8.04	55.9 \pm 5.92	47.7 \pm 5.51	35.8 \pm 5.59
Lying events per bird	1.30 \pm 0.265 ^a	1.19 \pm 0.193 ^a	0.69 \pm 0.119 ^{ab}	0.49 \pm 0.096 ^b
BW (g)- obstacle test	2,640 \pm 50.3 ^b	3,019 \pm 42.4 ^a	2,796 \pm 38.1 ^b	2,423 \pm 35.8 ^c
No. of obstacle crossings ⁶	5.22 \pm 0.698 ^b	6.40 \pm 0.567 ^b	7.18 \pm 0.639 ^{ab}	9.15 \pm 0.644 ^a
Latency to cross (s)	546.3 \pm 130.4	342.5 \pm 67.41	333.5 \pm 59.76	271.4 \pm 46.11

¹ BW from focal birds tested in the latency-to-lie test. Birds were weighed on the same day the test was conducted.

² Latency- to-lie per focal bird. Number of birds per category tested in the latency-to-lie test at Target Weight 1: CONV: n = 69, FAST: n = 106, MOD: n = 123, SLOW: n = 115.

³ BW from focal birds tested in the group obstacle test. Birds were weighed on the same day the test was conducted.

⁴ Number of obstacle crossings per focal bird. Number of birds per category tested in the group obstacle test at Target Weight 1: CON: n = 71, FAST: n = 130, MOD: n = 144, SLOW: n = 144.

⁵ Latency-to-lie per focal bird. Number of birds per category tested in the latency-to-lie test at Target Weight 2: CONV: n = 54, FAST: n = 95, MOD: n = 103, SLOW: n = 103.

⁶ Number of obstacle crossings per focal bird. Number of birds per category tested in the group obstacle test at Target Weight 2: CONV: n = 73, FAST: n = 126, MOD: n = 144, SLOW: n = 138.

^{a-c} Different superscripts within the same row represent differences among categories (P < 0.05).

Table 6.2: Effect of category on body weight (BW) latency-to-lie (LTL), and group obstacle test (LS-means \pm SEM) obtained at a similar age. Birds were tested at approximately 41 d and 46 d of age for the group obstacle test and latency-to-lie, respectively. Data corresponds to the week prior to Target Weight 1 and Target Weight 2 for slower-growing and CONV categories, respectively.

Variable	Category			
	CONV	FAST	MOD	SLOW
BW (g)- LTL test ¹	2,859 \pm 66.0 ^a	2,292 \pm 44.3 ^b	2,148 \pm 43.3 ^b	1,908 \pm 44.5 ^c
LTL (s) ²	350.7 \pm 34.84 ^b	390.7 \pm 24.47 ^b	412.02 \pm 23.20 ^{ab}	490.1 \pm 24.35 ^a
% of birds lying per pen	60.0 \pm 8.39 ^a	58.9 \pm 5.66 ^a	51.0 \pm 5.39 ^{ab}	34.8 \pm 5.50 ^b
Lying events per bird	1.63 \pm 0.281 ^a	1.22 \pm 0.197 ^{ab}	0.89 \pm 0.187 ^{ab}	0.61 \pm 0.196 ^b
BW (g)- obstacle test ³	2,670 \pm 56.3 ^a	2,003 \pm 40.7 ^b	1,905 \pm 37.2 ^b	1,590 \pm 36.1 ^c
No. of obstacle crossings ⁴	5.21 \pm 0.731 ^c	7.79 \pm 0.608 ^b	8.69 \pm 0.644 ^b	11.11 \pm 0.635 ^a
Latency to cross (s)	549.6 \pm 133.42 ^a	249.9 \pm 43.90 ^b	375.9 \pm 59.26 ^{ab}	255.5 \pm 38.80 ^b

¹ BW from focal birds tested in the latency to lie test. Birds were weighed on the same day the test was conducted.

² Latency-to-lie per focal bird. Number of birds per category tested at a similar age: CONV: n = 54, FAST: n = 106, MOD: n = 123, SLOW: n = 115.

³ BW from focal birds tested in the group obstacle test. Birds were weighed on the same day the test was conducted.

⁴ Number of obstacle crossings per focal bird. Number of birds per category tested at a similar age: CONV: n = 73, FAST: n = 130, MOD: n = 144, SLOW: n = 144.

^{a-c} Different superscripts within the same row represent differences among categories (P < 0.05).



Figure 6.1: Broiler chickens during the latency-to-lie test. The tank was placed on top of heating mat to help maintain the water at 30-32°C. The plexiglass portion of the tank was covered with a wood lid to prevent birds from flying out during the test. The birds were video recorded continuously during the test. The numbers on each side of the tank were used for identification purposes.



Figure 6.2: Broiler chickens during the obstacle test. The obstacle (white wooden barrier) was placed horizontally across the pen for 5 hours, during which the birds were continuously recorded. Birds had to cross the obstacle to obtain access to the feeder (located in the front of the pen) or drinker (located in the back of the pen). Focal birds were identified by livestock paint on their backs.

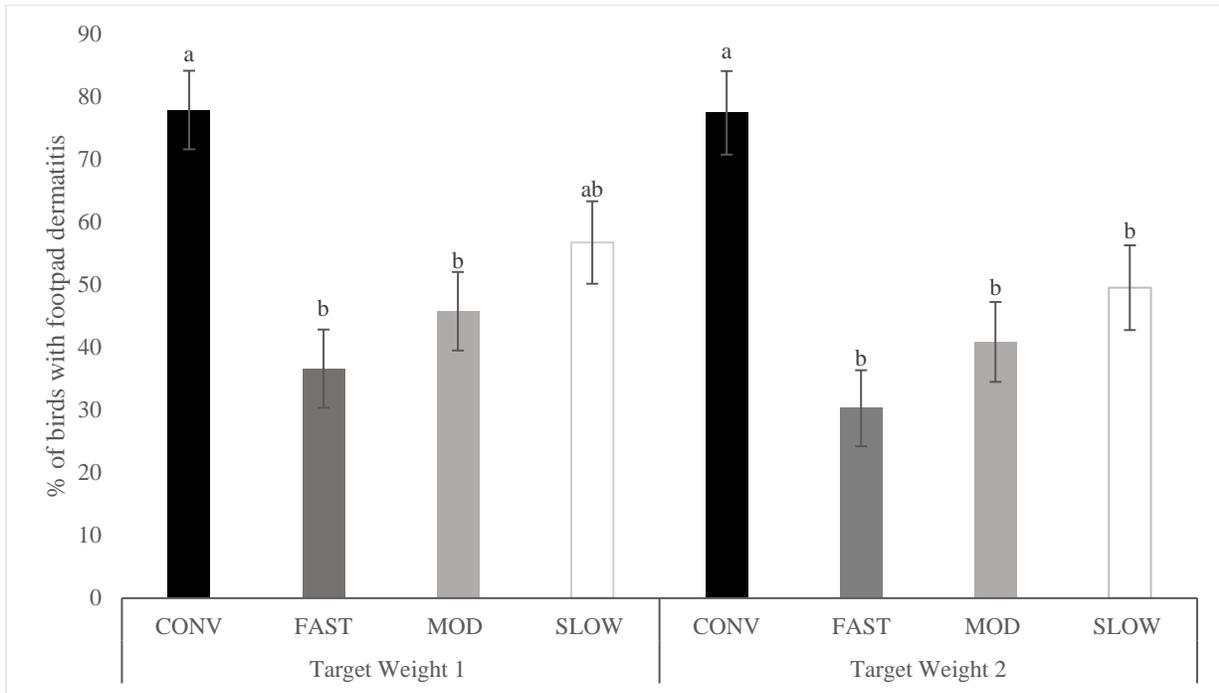


Figure 6.3: Effects of category on the total incidence of footpad dermatitis (LS-means \pm SEM) at Target Weight 1¹ and Target Weight 2². At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively. Within Target Weight, columns with different superscripts differ ($P < 0.05$).

¹ Number of birds per category evaluated at Target Weight 1: CONV: n = 273, FAST: n = 487, MOD: n = 547, SLOW: n = 528.

² Number of birds per category evaluated at Target Weight 2: CONV: n = 220, FAST: n = 460, MOD: n = 500, SLOW: n=504.

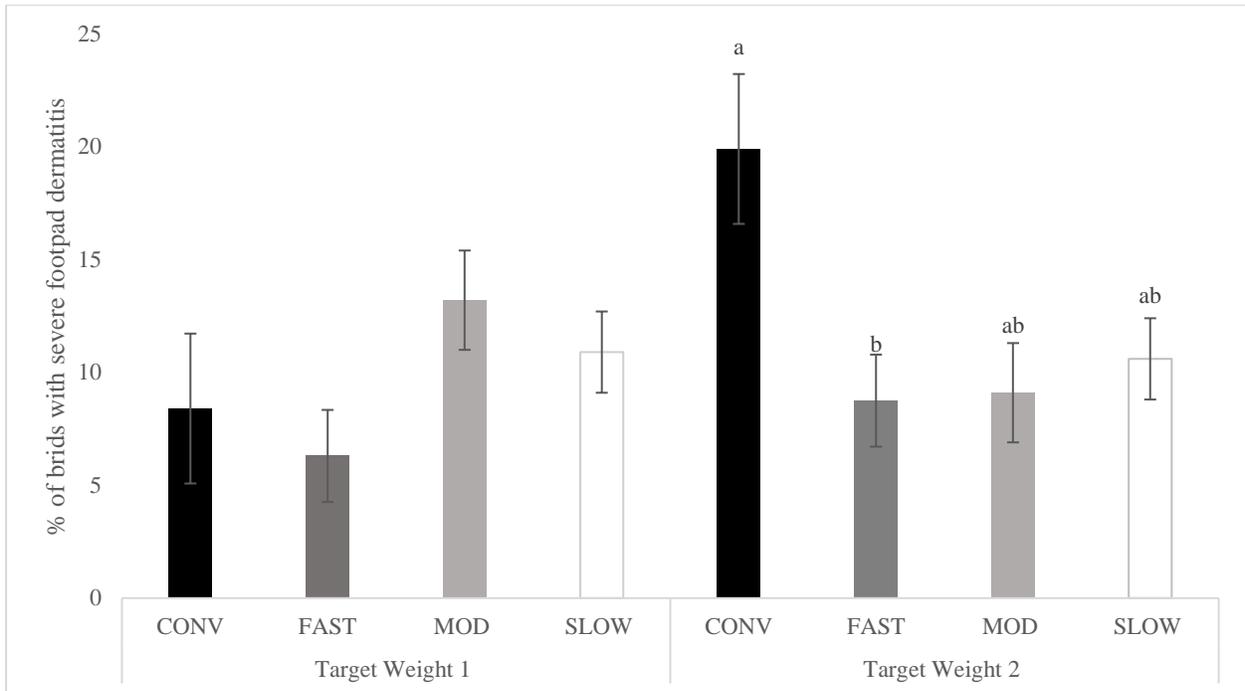


Figure 6.4: Effects of category on the total incidence of severe scores of footpad dermatitis (LS-means \pm SEM) at Target Weight 1¹ and Target Weight 2². At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively. Within Target Weight, columns with different superscripts differ ($P < 0.05$).

¹ Number of birds per category evaluated at Target Weight 1: CONV: n = 273, FAST: n = 487, MOD: n = 547, SLOW: n = 528.

² Number of birds per category evaluated at Target Weight 2: CONV: n = 220, FAST: n = 460, MOD: n = 500, SLOW: n=504.

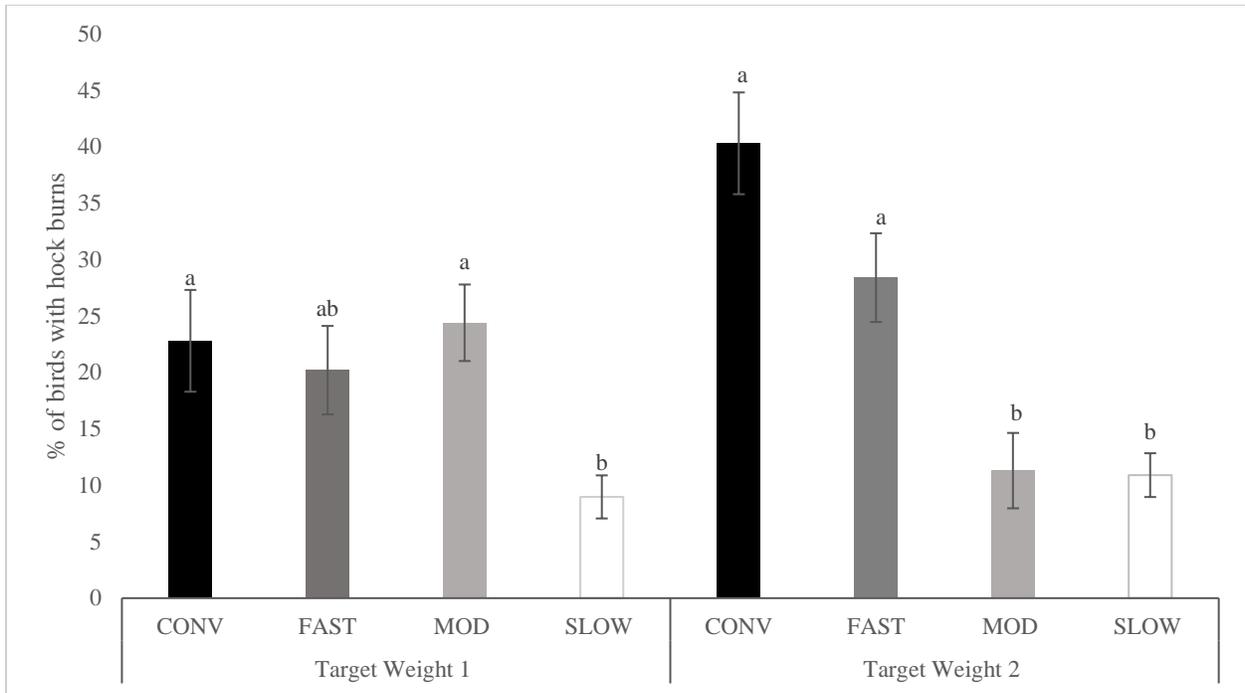


Figure 6.5: Effects of category on the total incidence of hock burns (LS-means \pm SEM) at Target Weight 1¹ and Target Weight 2². At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively. Within Target Weight, columns with different superscripts differ ($P < 0.05$).

¹ Number of birds per category evaluated at Target Weight 1: CONV: $n = 112$, FAST: $n = 354$, MOD: $n = 352$, SLOW: $n = 396$.

² Number of birds per category evaluated at Target Weight 2: CONV: $n = 131$, FAST: $n = 352$, MOD: $n = 353$, SLOW: $n = 395$.

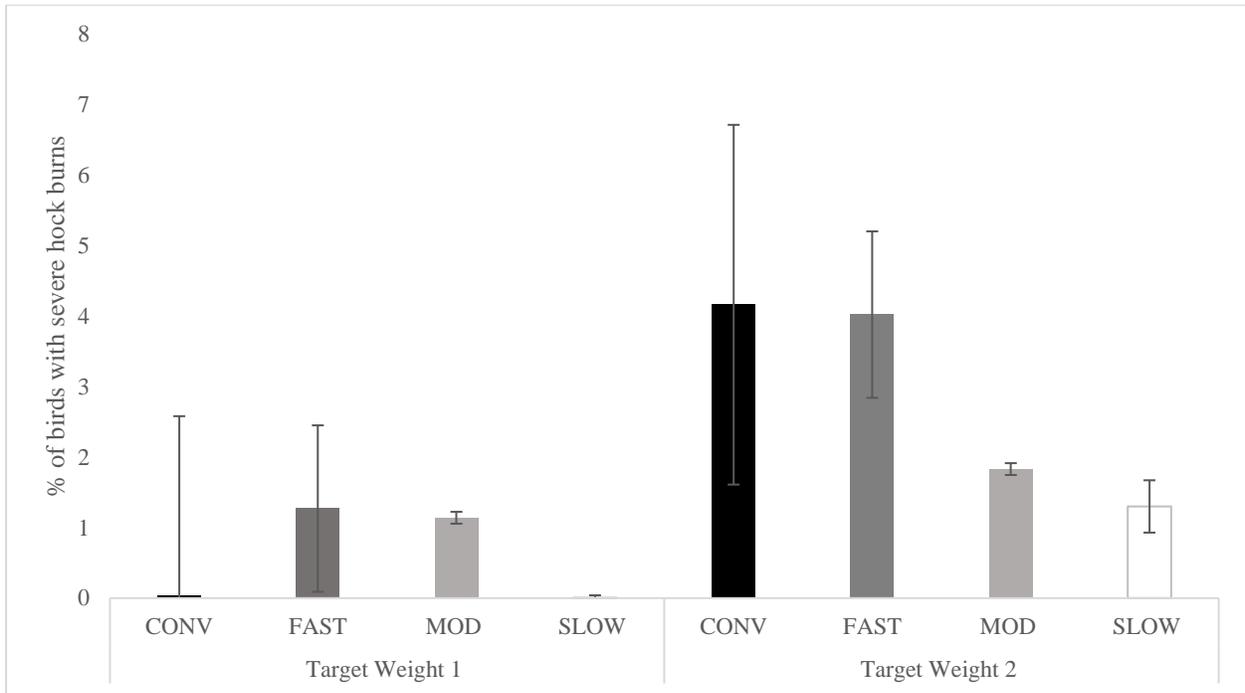


Figure 6.6: Effects of category on the total incidence of severe scores of hock burns (LS-means \pm SEM) at Target Weight 1¹ and Target Weight 2². At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

¹ Number of birds per category evaluated at Target Weight 1: CONV: n = 112, FAST: n = 354, MOD: n = 352, SLOW: n = 396.

² Number of birds per category evaluated at Target Weight 2: CONV: n = 131, FAST: n = 352, MOD: n = 353, SLOW: n = 395.

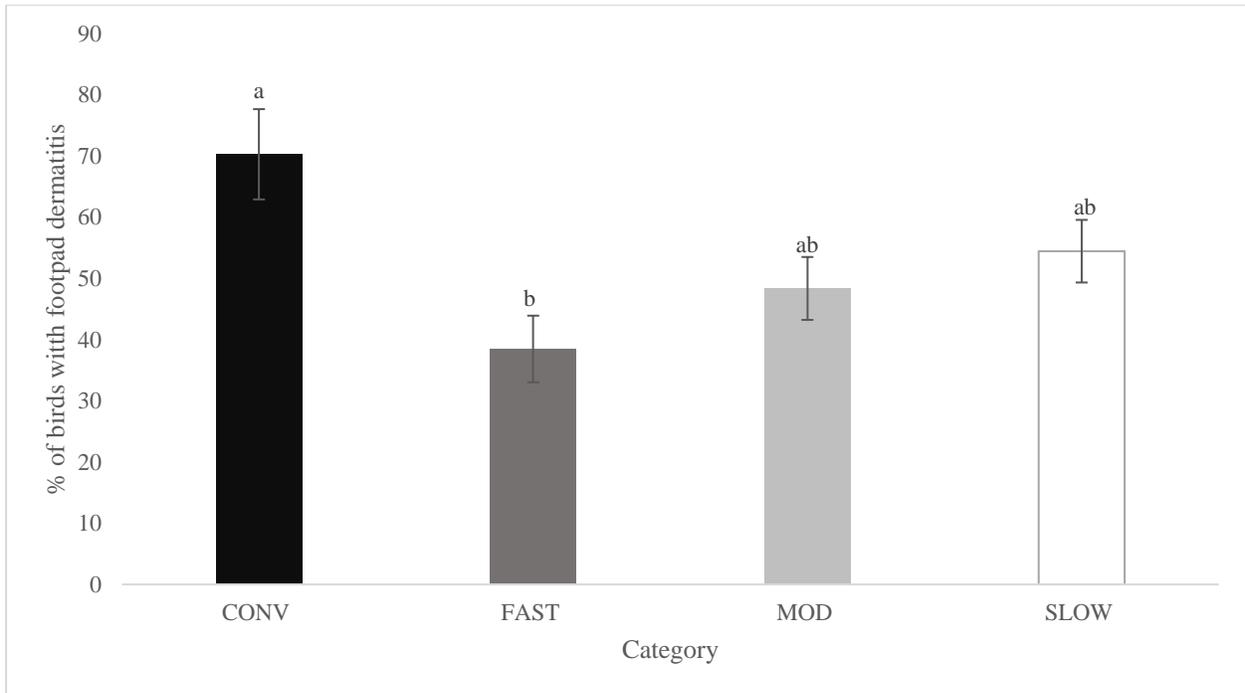


Figure 6.7: Effects of category on total incidence of footpad dermatitis (LS-means \pm SEM) of broiler chickens at 48 d of age¹.

¹ Number of birds per category evaluated at a similar age: CONV: n = 220, FAST: n = 487, MOD: n = 547, SLOW: n = 528.

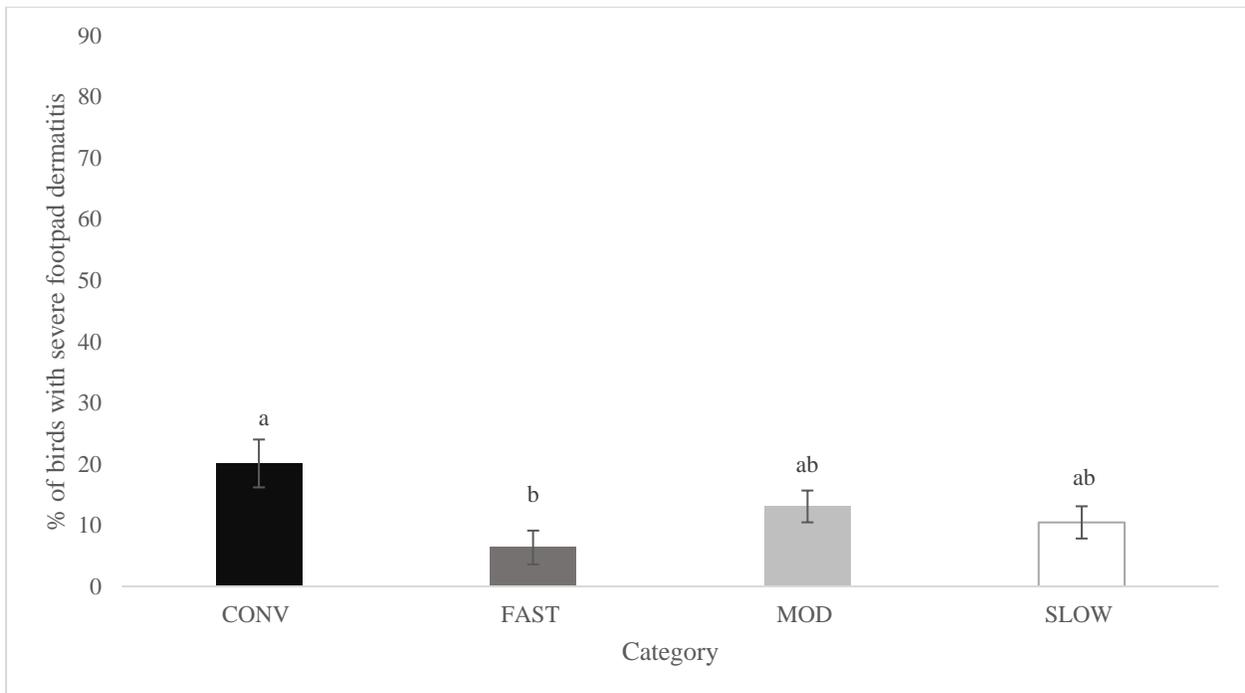


Figure 6.8: Effects of category on total incidence of severe footpad dermatitis (LS-means \pm SEM) of broiler chickens at 48 d of age¹.

¹ Number of birds per category evaluated at a similar age: CONV: n = 220, FAST: n = 487, MOD: n = 547, SLOW: n = 528.

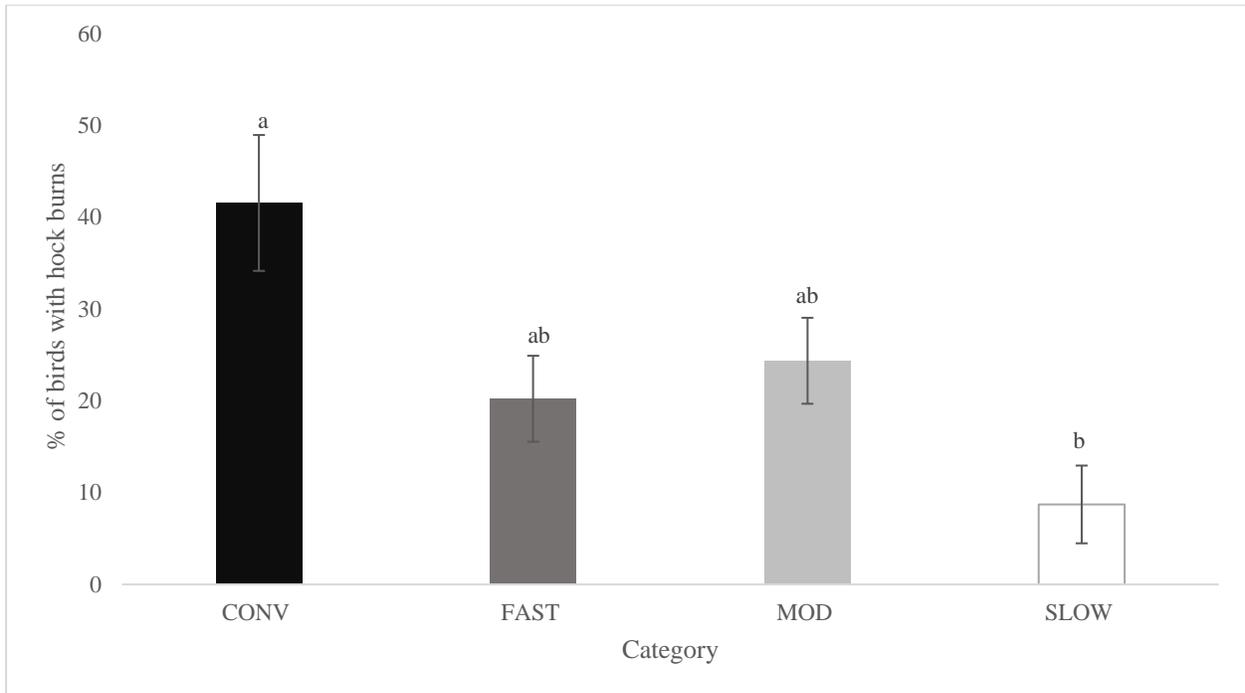


Figure 6.9: Effects of category on total incidence of hock burns (LS-means \pm SEM) of broiler chickens at 48 d of age¹.

¹ Number of birds per category evaluated at a similar age: CONV: n = 131, FAST: n = 354, MOD: n = 352, SLOW: n = 396.

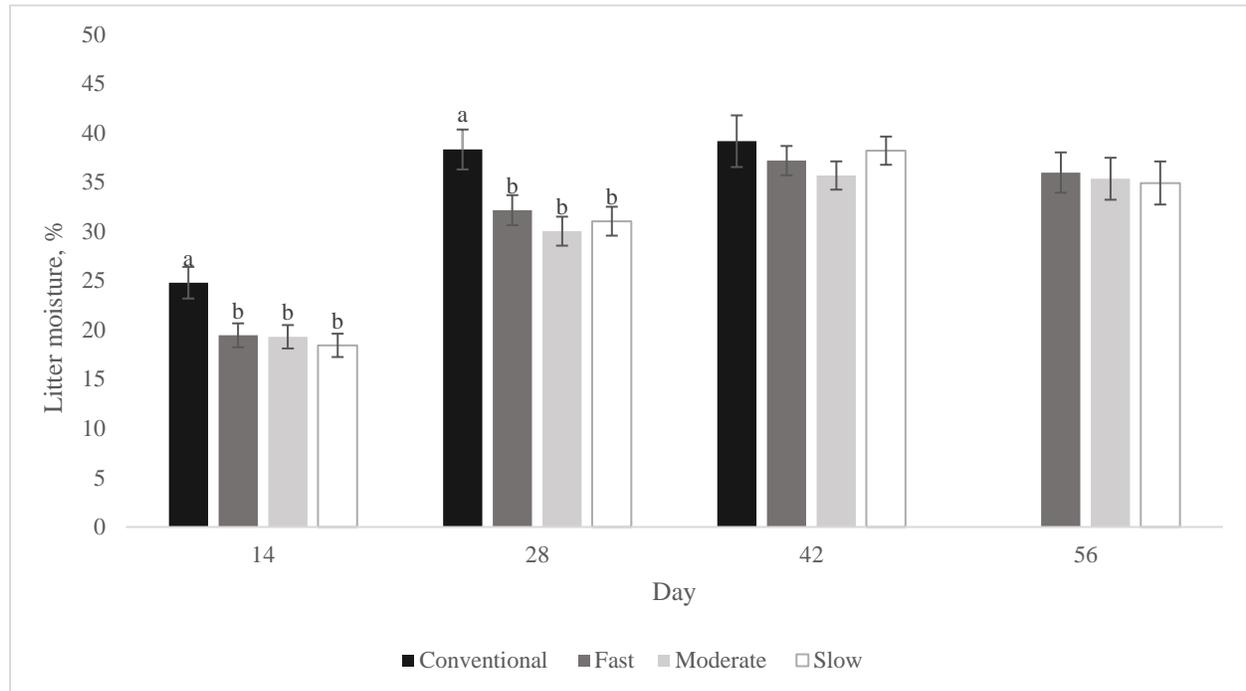


Figure 6.10: Effect of category on litter moisture (LS-means \pm SEM) on day 14¹, 28², 42³ and 56⁴. CONV birds are not represented on d 56 because birds were processed at 34 and 48 d. Within age, columns with different superscripts differ ($P < 0.05$).

¹ Number of pens per category at day 14: CONV: $n = 24$, FAST: $n = 44$, MOD: $n = 48$, SLOW: $n = 48$.

² Number of pens per category at day 28: CONV: $n = 24$, FAST: $n = 44$, MOD: $n = 48$, SLOW: $n = 48$.

³ Number of pens per category at day 42: CONV: $n = 12$, FAST: $n = 44$, MOD: $n = 48$, SLOW: $n = 48$.

⁴ Number of pens per category at day 56: FAST: $n = 22$, MOD: $n = 24$, SLOW: $n = 24$.

Chapter 7: General Discussion

7.1 Overall Findings

The early and accelerated growth of FG birds has been linked to several undesirable consequences that have economic and welfare implications to the poultry industry (Julian, 1998). In response to the growing consumer preference for products that promote improved animal welfare, hundreds of processors, retailers, and food-service companies have pledged to follow the standards of the “Better Chicken Commitment”. These standards encompass several practices to be adopted by the broiler industry aimed at the improvement of the welfare of broiler chickens, including the use of more robust breeds with better welfare outcomes, lower stocking densities, and better housing conditions (Better Chicken Commitment, 2021).

Although many studies have suggested better welfare for SG compared to FG birds, differences in BW when comparing the strains make it difficult to determine the effects of growth rate (Doğan et al., 2019; Mancinelli et al., 2020). Therefore, the main objective of this dissertation was to investigate the differences carcass traits, meat quality, and leg health amongst 2 FG and 12 SG strains. To the best of the author’s knowledge, there had been no published research evaluating these traits in such a wide variety of strains representing distinct growth rates and selection criteria.

To prevent the confounding of BW and rearing conditions on the response variables evaluated in this research, all the birds were raised under similar and controlled environmental conditions, fed a similar diet, and processed at two TWs of 2.1 kg (TW 1) and 3.2 kg (TW 2). This project was designed to provide a multidisciplinary approach to compare FG and SG strains due to the growing interest in SG strains as an alternative to mitigate the problems associated with accelerated growth.

Therefore, the results obtained in this research can be used to provide scientific-based recommendations about the use of SG strains raised under commercial rearing conditions (e.g., indoor systems with high stocking density and controlled temperature and lighting programs), which can be further exploited by primary breeding companies and animal welfare regulations to improve the welfare of broiler chickens.

Specifically, meat quality was assessed through the determination of the incidence of muscle myopathies including WB and WS (Chapter 4). Leg health was assessed by measuring bone quality indicators (Chapter 5), mobility, incidence and severity of contact dermatitis, and litter moisture content (Chapter 6). In addition, carcass characteristics were evaluated to provide an estimation of body conformation of strains selected for distinct growth rates (Chapter 4). Initially, the project was designed to compare all the strains tested in the study to each other. However, to simplify the statistical analysis, strains were grouped into four categories (CONV, FAST, MOD, and SLOW) based on their growth rate to reach TW 2. This categorization allowed comparisons among distinct growth rates to determine the impacts of selection for growth on the variables evaluated. Due to differences in selection criteria in strains selected for a similar growth rates, the research presented here also incorporated comparisons between strains within each category.

The results from this research indicate differences in most of the variables evaluated among categories, while fewer differences were observed for strains within the same category. Differences between sexes and TWs were also observed, but they were mostly dependent on the BW of the birds, as males were heavier than females and birds processed at TW 2 were heavier than those processed at TW 1, which is reflected in differences for most of the variables described in the previous chapters.

Despite the original plan to evaluate and process the birds at a similar BW, due to project logistics, the BW before processing differed among categories. In Chapter 6, at TW 1, CONV birds were lighter than FAST and MOD birds, which is reflected in their longer time standing, lower percentage of birds lying, and fewer number of times lying in water in the LTL test compared to FAST birds. These results are consistent with other studies that indicate the deleterious effects of the increase in BW on leg health (Weeks et al., 2000; Kestin et al., 2001; Bokkers et al., 2007; Nääs et al., 2010; Wilhelmsson et al., 2019), suggesting that at TW 1, BW rather than the growth rate likely influenced the differences obtained in the LTL test among categories. At TW 2 in the LTL test, CONV and SLOW birds had similar BW, which was lighter than MOD and FAST birds. However, CONV and FAST birds laid down in water sooner and more often compared to SLOW birds. While the heavier BW of FAST birds compared to SLOW may explain the differences obtained in the LTL test at TW 2, the similar BW of CONV and SLOW suggests that growth rate was likely the major contributor to the differences in leg strength between these categories at TW 2. Moreover, unlike the remaining categories, CONV birds showed a decrease in time spent standing and an increase in the percentage of birds lying down in water from TW 1 to TW 2, indicating that leg strength decreased as the birds grew heavier. The degeneration of leg health as the birds age is well established in broiler chickens, with previous studies emphasizing the negative effects of the increase in BW on walking ability (Sørensen et al., 2000; Venäläinen et al., 2006; Brickett et al., 2007). However, this decrease in leg strength assessed through the LTL test was more evident in CONV than SG birds. It is worth mentioning that the increase in BW from TW 1 to TW 2 for CONV, FAST, MOD, and SLOW was 63%, 46%, 48%, and 45%, respectively.

Therefore, the sharp increase in BW from TW 1 to TW 2 in CONV birds compared to the other categories may have exacerbated the differences obtained in the LTL test.

In the group obstacle test, CONV and FAST birds had a similar number of obstacle crossings at TW 1 and TW 2 despite the lower BW of CONV birds at both TWs compared to FAST. This similarity in the number of obstacle crossings may relate to the negative effects of growth rate on walking ability. Moreover, previous studies have demonstrated a gait alteration in FG birds in response to their distinct body conformation, due to selection with emphasis on large breast muscle compared to SG birds. These changes in body conformation were evident in the present study, in which CONV birds had greater breast yield (Chapter 4) yet shorter tibiae (Chapter 5) compared to the other categories at both TWs.

Tibiae from CONV birds were 1.62 to 2.05 cm and 1.07 to 1.30 cm shorter than SG birds at TW 1 and TW 2, respectively. The CONV birds were 2 wk younger than the remaining categories at both TWs, which may explain their shorter tibial length, as bone length increases as the birds grow (Lilburn, 1994). Because the obstacle had similar dimensions (160 × 9 × 10 cm; length × width × height) for all the strains despite the differences in body conformation, it is unknown if CONV birds were more affected by the presence of the obstacle due to their shorter legs compared to the SG birds. However, an earlier study conducted to compare differences in walking ability between FG and SG strains (fed *ad libitum* or restricted) showed that despite having longer legs at a similar age, FG birds fed *ad libitum* walked slower compared to SG birds and feed restricted FG birds (Corr et al., 2003a). The researchers also observed several gait alterations (e.g., shorter steps, longer step width, and longer double contact) in FG birds fed *ad libitum* compared to the other birds. These findings led the authors to conclude that these changes in gait may be an attempt of

FG birds to cope with the apparent instability, likely caused by the displacement of the centre of gravity cranially as a result of their larger breasts.

Because crossing the obstacle required the birds to step one foot at a time on and off the barrier, it is possible that CONV birds found this task more difficult compared to SLOW birds and as difficult as FAST birds despite the greater BW of FAST compared to CONV birds at both TWs when the group obstacle test was performed. In barnacle geese, an artificial increase in breast mass by applying loads weighing up to 15% of the body mass was twice as energetically expensive compared to carrying a similar load on the back (Tickle et al., 2010), suggesting high metabolic costs associated with moving a heavy sternum. Thus, the low locomotor activity commonly observed in FG birds and fewer obstacle crossings found in this study may be to compensate for the higher metabolic demand of standing and moving, partially due to their large breasts.

Despite the differences in body conformation (Chapter 4), tibial length, (Chapter 5), and walking ability (Chapter 6), CONV birds had similar or greater tibial breaking strength, ash, and organic content compared to SG birds (Chapter 5). Overall, the incidence of tibial dyschondroplasia was low in all categories, especially in CONV birds, indicating the successful incorporation of leg health and bone strength in breeding programs. However, CONV and SLOW birds had the lowest tibial dry matter, ash, and organic matter weight, with CONV also exhibiting the lowest values relative to the BW *vs.* SG birds at TW 2. The lower values obtained in CONV birds were likely due to their shorter tibiae and heavy BW at processing at TW 2. Although this may be an indicator of excessive body mass inadequately supported by light and short bones, the differences in bone characteristics may not necessarily reflect differences in walking ability in broiler chickens. A lack of relationship between bone parameters (e.g., bone strength, ash content, mineral density, bone

weight, and size traits) and walking ability has been reported by many researchers (Yalcin et al., 2001; Bizeray et al., 2002a; b; Venäläinen et al., 2006). While Talaty et al.(2010) found differences in gait scores among four commercial lines of broiler chickens, the strains did not differ in bone mineral density, bone length, diameter, or area. In addition, gait scores were not correlated to bone mineral content or tibial dimensions. However, a positive correlation between gait score and BW was found for all the strains, suggesting better walking ability in lighter birds. This agrees with many studies that reported a decrease in walking ability as the BW increases (Sørensen et al., 1999; Su et al., 1999; Venäläinen et al., 2006; Brickett et al., 2007). Nonetheless, because CONV birds were lighter than FAST birds in the LTL and group obstacle tests yet performed similarly or better (LTL test at TW 1) to this category, both BW and growth rate may have influenced the results obtained in these tests.

It was demonstrated by Caplen et al. (2014) that the presence of FPD and HB were explanatory variables in the LTL and group obstacle test. In addition, the incidence and severity of these lesions are commonly assessed as welfare outcomes in broiler chickens (Bessei, 2006). Therefore, the incidence and severity of these skin lesions were evaluated in this present study. Results obtained in Chapter 6 indicate no clear pattern of growth rate on the incidence of contact dermatitis at TW 1, as CONV and SLOW had a similar incidence of FPD, while the incidence of HB of CONV birds did not differ from those of FAST and MOD birds. However, at TW 2, the effects of growth rate were more evident among categories, as demonstrated by the highest incidence of FPD in CONV birds, whereas the incidence of HB was higher in CONV and FAST compared to MOD and SLOW birds.

Litter moisture content was high throughout the study, exceeding 30% from d 28 onwards for all categories. The results obtained in Chapter 6 indicate that the high litter moisture and to a lesser extent growth rate influenced the incidence of contact dermatitis in FG and SG birds. Correlation analyses between the presence of contact dermatitis and the results obtained in the LTL and group obstacle tests were overall weak and inconsistent among categories, indicating that the differences in FPD and HB were likely not the determining factor in leg strength and ability to cross the obstacle.

As described in Chapter 6, the large breast muscle of FG birds likely influenced the ability of birds to remain standing or to cross the obstacle in the LTL and group obstacle tests, respectively. Interestingly, strain F, which had the larger breast yield among FAST strains (Chapter 4), also had lowest number of obstacle crossings compared to the other FAST strains at both TWs (Chapter 6), though not statistically different. The breast yield of strain F was as high or higher than CONV birds at both TWs. The high breast yield of these strains was associated with a high incidence and severity of WB and WS, suggesting that the emphasis on larger breast muscles may play a crucial role in the occurrence of muscle myopathies, which has been described by other researchers (Kuttappan et al., 2013b, 2017; Alnahhas et al., 2016). Even though the increase in BW contributes to the development of muscle disorders, CONV birds had the highest incidence of WB at TW 1, despite having the lowest BW. Although the underlying causes and mechanisms of WB and WS are yet to be determined, it has been suggested that these myopathies may influence the welfare of affected broilers. Due to the high prevalence of WB and WS in FG birds, the effects of these muscle abnormalities on welfare should be investigated in future studies.

7.2 Limitations of the Project

Some limitations occurred during this study, which may have affected the variables measured, to some extent. The effects of BW on walking ability, leg strength, and the incidence of myopathies have been well documented. However, BW differed among categories at both TWs, with CONV and SLOW birds being, in general, lighter than FAST and MOD at TW 1, while at TW 2, SLOW birds were lighter than the other categories. Because the strains used in the present study represents a wide range of growth rates, multiple behavioural tests and processing dates would be required to accommodate such differences in potential for growth to evaluate the birds from all the strains at a similar BW. As this was not possible due to project logistics, CONV and SG birds were grouped to be evaluated and processed at specific ages, resulting in a variation in BW among categories at both TWs. Therefore, it is unknown if the findings of the study would be different if birds from all categories and strains were evaluated at more similar BWs. However, some of the variables such as carcass and tibial traits were corrected for BW (Chapter 4 and 5). In addition, as previously described, differences in BW could not explain some of the results obtained in the incidence of muscle disorders (Chapter 4), LTL and group obstacle tests (Chapter 6), suggesting the effects of growth rate and breast size on some of the variables measured in this study.

While providing similar incubation, diet, and rearing conditions to all the strains allowed comparisons under a standard and controlled laboratory environment, these conditions may have affected the strains differently, with some strains performing better than others. Previous studies have demonstrated the relevance of ideal incubation and/or rearing conditions and diet on muscle and bone development and integrity (Waldenstedt, 2006; Oviedo-Rondón et al., 2009; Nääs et al.,

2012; Cruz et al., 2017; Clark et al., 2017). Because a large number of strains were tested in this research trial, providing ideal incubation and rearing conditions for each strain was not feasible. However, due to the potential impacts of these factors on meat quality, walking ability, health, and overall welfare, the optimization of specific conditions to each strain should be taken into account in other studies comparing FG and SG strains. As an example, differences in heat tolerance was observed between 4 FG strains of broiler chickens, indicating possible strain-specific requirements for rearing conditions (Chand et al., 2018). Similarly, differences in microclimatic ammonia levels were observed between 2 FG strains (Soliman et al., 2017), which could lead to differences in ventilation to optimize the performance and growth of each strain.

The high litter moisture throughout the experiment may be an indicator of suboptimal environmental conditions, which may have affected the incidence and severity of contact dermatitis, especially in CONV birds, due to the higher litter moisture in pens from CONV birds compared to SG strains at d 14 and d 28. The litter was only replaced at the beginning of each trial to simulate Canadian commercial broiler production, where this practice is commonly performed. However, the use of solid walls to separate adjacent pens in the room, combined with a high stocking density (30 kg/m²) probably affected the efficacy of the ventilation in the room and at the pen level, leading to a rapid increase in litter moisture content. Furthermore, some SLOW strains perched on the drinker line, which likely contributed to an increase in litter moisture content due to water spillage from the drinkers, reflected in the high incidence of FPD observed in these strains. Thus, the prevention of this behaviour and better litter quality management should be considered in other studies to assess differences in litter moisture content and contact dermatitis in strains differing in growth rate.

Another limitation of this study was caused by the low statistical power ($1-\beta < 0.70$) found in some variables, especially for comparisons among strains within a category, which likely occurred due to the high variation and small sample size of the present study. Thus, large differences among strains within categories were not statistically detected, possibly preventing the determination of peculiarities in breeding goals and selection criteria for strains selected for a similar growth rate. The budget, timeline, and number of strains evaluated in this present research, prevented the use of a larger sample size to account for a large variation. Therefore, the use of more experimental units per strain to account for such variation would lead to a better understanding of the differences among categories and strains within categories.

7.3 Next Steps

As previously mentioned in this thesis, this study was part of a multidisciplinary project that investigated behavioural, physiological, production, health, bone traits, meat quality, and welfare differences among strains selected for distinct growth rates. While this thesis focused on carcass traits, incidence of contact dermatitis and muscle myopathies, bone attributes, and walking ability, the incorporation of other traits collected throughout the study (e.g., behaviour, production parameters, meat quality, plasma attributes, inactivity) can provide a deeper understanding of differences among strains differing in growth rate. Furthermore, combining these variables can reveal some relevant relationships that can be further investigated in other studies and potentially provide information for future breeding programs, aimed at improving broiler production and welfare. For this purpose, a principal component analysis is being conducted with all the variables

evaluated in this multidisciplinary study. This analysis is classified as exploratory statistics due to the possibility of exploring if any relationship exists among variables collected.

Though not presented in this thesis, meat quality traits, including pH, colour, drip loss, cook loss, and shear force were also evaluated in the study. Because the muscle myopathies investigated have been shown to alter meat quality in FG birds (Zanetti et al., 2018), I will be able to evaluate if similar modifications are observed in SG birds affected by WB or WS. In addition, these results can be used to identify differences in meat quality that may have effects on consumer acceptance, especially considering the growing demand for meat from SG birds.

Another interesting test performed that was not included in this thesis is the novel object test. This test is used to assess fear responses to novelty in several species, including poultry. As suggested by the name, in this test a novel object to which the birds have not been previously exposed is placed in their home pen (Keer-Keer et al., 1996). The latency to approach and the number of birds close to the object are recorded, with longer latency and fewer birds approaching the object being associated with higher degrees of fear (Graml et al., 2008). Because age is known to affect the fear response (Albentosa et al., 2003), this test was performed at two ages (d 11 and d 38) to investigate strain differences as the birds grew. The results from this test may be of particular relevance to investigate if some strains are more fearful than others, which could lead to welfare implications depending on the conditions in which the birds are raised.

The studies presented in this thesis along with the other variables collected by other researchers provided some answers, but more importantly, more questions than I originally had about the use of FG and SG strains in commercial conditions. Hopefully, these findings can be further

investigated in future studies to provide an in-depth approach about the impacts of growth rate on the aforementioned traits and other variables not encompassed in this study yet of relevance to the broiler industry.

7.4 Final Considerations and Conclusion

Selection for growth rate and large breast muscles have been linked to skeletal and muscle disorders in FG birds, which ultimately have economic and welfare implications for the poultry industry. The results obtained in this research confirm the findings from earlier studies, suggesting that selection for growth and distinct body conformation, with emphasis on breast yield, have negative impacts on leg strength, walking ability, and incidence of muscle disorders. However, CONV birds exhibited similar or higher bone breaking strength and mineral content compared to the other SG birds. Furthermore, the low incidence of leg disorders observed in this study indicates the successful inclusion of leg health in more holistic breeding strategies practiced in FG broiler chickens. This indicates the potential of selecting for robustness concomitantly with production to improve birds' health and welfare. Even though CONV birds had, in some cases, a higher incidence of contact dermatitis than SG birds, the development of FPD and HB seemed to be more related to litter moisture content than differences in growth rate.

This research provides a better understanding of the welfare and meat quality of strains differing in growth rate raised under similar conditions. This is especially important due to the growing interest in the use of SG strains to meet the growing consumer demand for products that promote better animal welfare. The strains used in this study encompass a wide range of growth rate, with SG birds representing strains with ADG ranging from to 43.6 to 55.5 g/d. In addition to the

differences in growth rate, a great variation in several traits evaluated in this study were observed among strains classified as SG strains, suggesting that not all SG strains have better welfare than FG strains.

Further research is needed to investigate if the results obtained in this study can be extrapolated to commercial broiler houses. In addition, the economic and environmental implications should be evaluated in future studies comparing FG and SG strains in order to investigate if the latter can be used as an alternative to decrease the aforementioned disorders associated with growth and improve broiler welfare while considering the sustainability and productivity of the poultry industry.

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APPENDIX A.1

Effects of Category and Sex on Tibial Traits at Target Weight 1 and 2

Table A.2.1. Effect of category and sex on tibial traits (LS-means \pm SEM) at Target weight 1 and 2. At Target Weight 1, conventional and slower strains birds were 34 and 48 d of age, respectively. At Target Weight 2, conventional and the remaining categories were 48 and 62 days, respectively.

Variable	Sex	Category			
		CONV	FAST	MOD	SLOW
Target Weight 1					
Length (mm)	F	93.8 \pm 0.89 ^c	112.91 \pm 0.73 ^{az}	109.9 \pm 0.69 ^{bz}	108.76 \pm 0.68 ^{bz}
	M	97.5 \pm 1.05 ^c	119.4 \pm 0.81 ^{ay}	115.1 \pm 0.72 ^{by}	114.88 \pm 0.72 ^{by}
Target Weight 2¹					
Length (mm)	F	113.5 \pm 1.05 ^{bz}	125.1 \pm 0.83 ^{az}	123.3 \pm 0.78 ^{az}	123.2 \pm 0.77 ^{az}
	M	120.6 \pm 0.88 ^{by}	135.0 \pm 0.81 ^{ay}	133.4 \pm 0.74 ^{ay}	132.2 \pm 0.74 ^{ay}
Dry wt (g)	F	8.78 \pm 0.294 ^z	9.60 \pm 0.203 ^z	9.02 \pm 0.222 ^z	8.87 \pm 0.220 ^z
	M	11.17 \pm 0.243 ^{cy}	13.64 \pm 0.183 ^{ay}	12.67 \pm 0.198 ^{by}	11.90 \pm 0.205 ^{bcy}
Ash wt (g)	F	3.45 \pm 0.128 ^z	3.76 \pm 0.099 ^z	3.54 \pm 0.101 ^z	3.52 \pm 0.091 ^z
	M	4.41 \pm 0.106 ^{cy}	5.47 \pm 0.089 ^{ay}	5.06 \pm 0.089 ^{by}	4.85 \pm 0.085 ^{by}
Organic matter (g)	F	5.31 \pm 0.185 ^z	5.85 \pm 0.125 ^z	5.48 \pm 0.147 ^z	5.34 \pm 0.148 ^z
	M	6.77 \pm 0.511 ^{cy}	8.17 \pm 0.111 ^{ay}	7.61 \pm 0.129 ^{by}	7.07 \pm 0.137 ^{bcy}
Ash:BW (g/kg)	F	1.16 \pm 0.023 ^b	1.23 \pm 0.021 ^{bz}	1.25 \pm 0.027 ^{bz}	1.37 \pm 0.025 ^{az}
	M	1.23 \pm 0.018 ^c	1.41 \pm 0.019 ^{by}	1.41 \pm 0.024 ^{by}	1.53 \pm 0.023 ^{ay}
Ash:Length (g/mm)	F	0.31 \pm 0.009 ^z	0.30 \pm 0.006 ^z	0.29 \pm 0.007 ^z	0.29 \pm 0.007 ^z
	M	0.36 \pm 0.007 ^{by}	0.40 \pm 0.006 ^{ay}	0.38 \pm 0.006 ^{aby}	0.36 \pm 0.006 ^{by}

¹ Tibial dry matter, organic and inorganic content were only obtained at Target Weight 2.

^{a-c} Different superscripts within the same row represent significant differences between categories for each sex (P<0.05).

^{y-z} Different superscripts within the same column for the same parameter represent significant differences between sexes in each category (P<0.05).

Body Weight and Tibial Traits of Strains A and T

Table A.2.3. Body weight (BW), tibial breaking strength (TBS), tibial morphology, and tibial ash and organic content at Target Weights 1 and 2. Only descriptive data included.

Variable	Strain ¹	
	A	T
Target Weight 1²		
BW (g)	1,652	N/A
TBS (N) ³	313.3	N/A
TBS:BW (N/kg) ⁴	188.2	N/A
Diameter (mm)	7.41	N/A
Length (mm)	91.88	N/A
Diameter:BW (mm/kg)	4.49	N/A
Length:BW (mm/kg)	55.87	N/A
Length:Diameter	12.50	N/A
Target Weight 2⁵		
BW (g)	3,070	1,019
TBS (N)	371.9	185.3
TBS:BW (N/kg)	120.9	183.4
Diameter (mm)	9.00	6.78
Length (mm)	118.12	104.9
Diameter:BW (mm/kg)	2.95	6.75
Length:BW (mm/kg)	39.14	104.7
Length:Diameter	13.23	15.51
Dry matter wt (g) ⁶	9.96	4.87
Ash wt (g)	4.13	1.79
Organic matter wt (g)	5.83	4.11
Dry matter wt:BW (%)	3.23	4.84
Ash wt:BW (%)	1.34	1.79
Ash content (%)	41.50	36.98
Organic matter (%)	58.49	66.21
Organic:Inorganic	1.41	1.95
Ash: Length (g/mm)	0.347	0.171

¹ Due to the reduced sample size, only descriptive statistics are provided for strains A (Fast growing; ADG₀₋₄₈ = 62.65 g/d) and T (Slower growing; ADG₀₋₆₂ = 19.78 g/d). Because strain T had the lowest ADG among the strains, leg traits were only obtained at TW 2.

² Number of birds per strain at Target Weight 1: A: n= 6.

³ Absolute tibial breaking strength (TBS). Maximum TBS expressed in newtons (N).

⁴ Relative tibial breaking strength (TBS). Maximum TBS was obtained in newtons (N) and adjusted for the BW.

⁵ Number of birds per strain at Target Weight 2: A: n= 7; T: n=11.

⁶ Tibial dry matter, and organic, and inorganic content were only obtained at Target Weight 2.

Comparison of Loading Rate

In order to test if the higher loading speed adopted in our study interfered with the results found, the right tibia samples from 40 birds, representing 10 strains (4 samples per strain) were analyzed using a loading rate of 50 mm/min and compared to the left tibia of the same birds, which were analyzed using a higher loading rate of 20 mm/s. The loading speeds tested had no effect on TBS (Low speed = 369.6 ± 16.60 N; High speed = 359.8 ± 16.67 N, $P= 0.383$), suggesting that the high loading speed used in our study did not significantly impact our results. However, future studies should use low loading speed, due to the possible impacts on bone mechanical properties and to allow comparisons with other studies that adopted a lower speed.

APPENDIX A.2

Effect of Strain on Latency-to-Lie and Group Obstacle Test at Target Weight 1 and 2.

Table A.2.1. Differences in body weight (BW), latency-to-lie (LTL), and group obstacle tests (LS means \pm SEM) among CONV strains in the week prior to Target Weights 1 and 2. At Target Weight 1 and 2, CONV birds were 34 and 48 days, respectively.

Variable	Strain	
	B	C
Target Weight 1		
BW (g)- LTL test ¹	1,726 \pm 79.6	1,735 \pm 81.3
LTL (s)	549.2 \pm 39.75	460.9 \pm 42.07
% of birds lying per pen	17.1 \pm 9.45	35.5 \pm 11.34
Lying events per bird	0.42 \pm 0.155	0.77 \pm 0.215
Target Weight 2		
BW (g)- LTL test	2,992 \pm 88.1	2,733 \pm 85.3
LTL (s)	340.6 \pm 45.76	379.8 \pm 44.08
% of birds lying per pen	58.2 \pm 10.83	60.1 \pm 11.88
Lying events per bird	1.04 \pm 0.326	1.62 \pm 0.421
BW (g)- obstacle test	2,794 \pm 73.8	2,485 \pm 66.4
No. of obstacle crossings ³	4.60 \pm 1.002	5.85 \pm 0.976
Latency to cross (s)	556.8 \pm 187.78	536.1 \pm 181.06

¹ BW from focal birds tested in the latency-to- lie test. Birds were weighed on the same day the test was conducted.

² BW from focal birds tested in the group obstacle test. Birds were weighed on the same day the test was conducted.

³ Number of obstacle crossing per focal bird.

Table A.2.2. Differences in body weight (BW), latency-to-lie (LTL), and group obstacle test (LS means \pm SEM) among FAST strains in the week prior to Target Weights 1 and 2. At Target Weight 1 and 2, FAST birds were 48 and 62 days, respectively

Variable	Strain			
	F	G	I	M
Target Weight 1				
BW (g)- LTL test ¹	2,357 \pm 79.5 ^a	2,442 \pm 99.7 ^a	2,413 \pm 99.0 ^a	1,911 \pm 85.5 ^b
LTL (s)	425.2 \pm 40.18	403.7 \pm 47.03	342.6 \pm 46.17	403.8 \pm 46.03
% of birds lying per pen	51.1 \pm 10.73	56.9 \pm 11.08	75.0 \pm 11.08	48.0 \pm 10.66
Lying events per bird	1.96 \pm 0.447	1.03 \pm 0.336	1.09 \pm 0.342	0.62 \pm 0.229
Target Weight 2				
BW (g)- LTL test	3,485 \pm 84.1	3,396 \pm 99.0	3,339 \pm 99.0	3,109 \pm 104.9
LTL (s)	404.7 \pm 42.83	372.5 \pm 46.17	357.8 \pm 46.17	410.7 \pm 55.97
% of birds lying per pen	56.3 \pm 11.82	58.3 \pm 11.08	58.3 \pm 11.08	50.6 \pm 13.21
Lying events per bird	1.47 \pm 0.369	1.32 \pm 0.397	1.42 \pm 0.420	0.73 \pm 0.304
BW (g)- obstacle test	3,154 \pm 82.2	2,990 \pm 79.3	2,968 \pm 79.3	2,964 \pm 97.1
No. of obstacle crossings ³	4.72 \pm 1.122	5.56 \pm 1.053	6.78 \pm 1.053	8.54 \pm 1.290
Latency to cross (s)	439.0 \pm 173.8	428.8 \pm 155.9	182.8 \pm 66.48	400.1 \pm 180.96

¹ BW from focal birds tested in the LTL test. Birds were weighed on the same day the test was conducted.

² BW from focal birds tested in the group obstacle test. Birds were weighed on the same day the test was conducted.

³ Number of obstacle crossing per focal bird.

^{a,b} Different superscripts within the same row represent differences among categories ($P < 0.05$).

Table A.2.3. Differences in body weight (BW), latency-to-lie (LTL), and group obstacle test (LS means \pm SEM) among MOD strains in the week prior to Target Weights 1 and 2. At Target Weight 1 and 2, MOD birds were 48 and 62 days, respectively.

Variable	Strain			
	E	H	O	S
Target Weight 1				
BW (g)- LTL test ¹	2,220 \pm 79.6 ^a	1,735 \pm 72.1 ^b	2,398 \pm 99.0 ^a	2,208 \pm 99.0 ^a
LTL (s)	328.6 \pm 39.75	486.1 \pm 36.46	435.2 \pm 46.17	414.0 \pm 46.17
% of birds lying per pen	64.5 \pm 9.45	36.8 \pm 9.88	50.0 \pm 11.08	50.0 \pm 11.08
Lying events per bird	0.87 \pm 0.247	0.62 \pm 0.717	0.96 \pm 0.313	0.69 \pm 0.249
Target Weight 2				
BW (g)- obstacle test ²	2,160 \pm 77.2 ^a	1,821 \pm 72.9 ^{ab}	1,868 \pm 77.2 ^{ab}	1,735 \pm 77.2 ^b
No. of obstacle crossings ³	8.75 \pm 1.278	8.91 \pm 1.267	7.11 \pm 1.278	9.88 \pm 1.278
Latency to cross (s)	339.8 \pm 122.9	410.0 \pm 145.9	497.7 \pm 178.3	285.6 \pm 102.3
Target Weight 2				
BW (g)- LTL test	3,350 \pm 88.1 ^a	2,884 \pm 73.9 ^b	3,386 \pm 99.0 ^a	3,098 \pm 99.1 ^{ab}
LTL (s)	397.1 \pm 45.76	489.8 \pm 41.95	360.5 \pm 46.17	449.2 \pm 46.20
% of birds lying per pen	61.7 \pm 10.83	29.2 \pm 11.04	58.3 \pm 11.08	41.7 \pm 11.08
Lying events per bird	0.79 \pm 0.267	0.57 \pm 0.186	0.69 \pm 0.250	0.71 \pm 0.257
BW (g)- obstacle test	2,916 \pm 77.2	2,683 \pm 72.9	2,893 \pm 77.2	2,694 \pm 77.5
No. of obstacle crossings	9.64 \pm 1.278	6.94 \pm 1.279	5.67 \pm 1.278	6.48 \pm 1.278
Latency to cross (s)	425.0 \pm 152.2	359.86 \pm 129.3	274.2 \pm 98.2	294.8 \pm 105.6

¹ BW from focal birds tested in the LTL test. Birds were weighed on the same day the test was conducted.

² BW from focal birds tested in the group obstacle test. Birds were weighed on the same day the test was conducted.

³ Number of obstacle crossing per focal bird.

^{a,b} Different superscripts within the same row represent differences among categories ($P < 0.05$).

Table A.2.4. Differences in body weight (BW), latency-to-lie (LTL), and group obstacle test (LS means \pm SEM) among SLOW strains in the week prior to Target Weights 1 and 2. At Target Weight 1 and 2, SLOW birds were 48 and 62 days, respectively.

Variable	Strain			
	D	J	K	N
Target Weight 1				
BW (g)- LTL test ¹	1,671 \pm 67.1	2,082 \pm 102.3	1,935 \pm 99.0	1,936 \pm 99.0
LTL (s)	460.6 \pm 34.68	493.5 \pm 49.83	481.8 \pm 46.17	516.8 \pm 46.17
% of birds lying per pen	36.1 \pm 9.22	54.2 \pm 11.08	29.2 \pm 11.08	20.8 \pm 11.08
Lying events per bird	0.51 \pm 0.138	0.79 \pm 0.292	0.46 \pm 0.189	0.46 \pm 0.190
Target Weight 2				
BW (g)- LTL test	2,797 \pm 74.1	2,990 \pm 99.0	2,824 \pm 99.0	2,603 \pm 99.6
LTL (s)	514.5 \pm 41.82	493.3 \pm 46.17	485.0 \pm 46.17	435.3 \pm 46.96
% of birds lying per pen	18.0 \pm 11.47	37.5 \pm 11.08	33.3 \pm 11.08	54.2 \pm 11.08
Lying events per bird	0.29 \pm 0.117	0.43 \pm 0.182	0.48 \pm 0.193	0.93 \pm 0.312
BW (g)- obstacle test	2,383 \pm 64.6	2,545 \pm 71.5	2,443 \pm 78.3	2,322 \pm 71.5
No. of obstacle crossings ³	10.07 \pm 1.248	7.44 \pm 1.259	9.70 \pm 1.380	9.39 \pm 1.259
Latency to cross (s)	311.5 \pm 104.1	364.5 \pm 120.57	216.5 \pm 78.44	220.7 \pm 73.00

¹ BW from focal birds tested in the LTL test. Birds were weighed on the same day the test was conducted.

² BW from focal birds tested in the group obstacle test. Birds were weighed on the same day the test was conducted.

³ Number of obstacle crossing per focal bird.

Effect of Category and Sex on Latency-to-Lie Test at Target Weight 1 and 2.

Table A.2.5. Effect of category and sex and category on body weight (BW) and latency-to-lie (LTL) test (LS means \pm SEM) within a week prior Target Weight 1 and Target Weight 2

Variable	Sex	Strain			
		CONV	FAST	MOD	SLOW
Target Weight 1					
BW (g)- LTL ¹	F	1,619 \pm 72.4 ^{bz}	2,058 \pm 54.1 ^{az}	1,933 \pm 51.1 ^{az}	1,732 \pm 53.5 ^{bz}
	M	1,843 \pm 60.0 ^{cy}	2,504 \pm 50.5 ^{ay}	2,347 \pm 48.4 ^{ay}	2,085 \pm 51.0 ^{by}
LTL (s)	F	521.3 \pm 45.77	453.3 \pm 32.13 ^y	453.3 \pm 29.80 ^y	483.9 \pm 30.98
	M	488.9 \pm 33.05 ^a	334.4 \pm 28.60 ^{bz}	378.6 \pm 26.66 ^{bz}	492.4 \pm 28.03 ^a
% of birds lying per pen	F	25.0 \pm 10.49	44.5 \pm 7.26 ^z	41.0 \pm 6.83 ^z	36.8 \pm 6.95
	M	27.5 \pm 8.50 ^b	71.0 \pm 6.71 ^{ay}	59.7 \pm 6.41 ^{ay}	33.3 \pm 6.58 ^b
Target Weight 2					
BW (g)- LTL	F	2,579 \pm 75.8 ^{bcz}	2,960 \pm 56.5 ^{az}	2,785 \pm 53.2 ^{bz}	2,475 \pm 54.1 ^{cz}
	M	3,077 \pm 66.1 ^{by}	3,704 \pm 53.3 ^{ay}	3,574 \pm 50.0 ^{ay}	3,087 \pm 51.5 ^{by}
LTL (s)	F	426.5 \pm 47.04 ^y	514.3 \pm 33.45 ^y	534.3 \pm 31.81 ^y	532.9 \pm 31.71 ^y
	M	293.9 \pm 38.38 ^{bz}	258.5 \pm 30.30 ^{bz}	314.0 \pm 28.68 ^{bz}	431.1 \pm 28.87 ^{az}
% of birds lying per pen	F	44.7 \pm 10.77 ^z	29.1 \pm 7.67 ^z	20.9 \pm 7.25 ^z	26.1 \pm 7.27 ^z
	M	73.6 \pm 9.77 ^{ay}	82.6 \pm 7.32 ^{ay}	74.5 \pm 6.81 ^{ay}	45.4 \pm 6.94 ^{by}

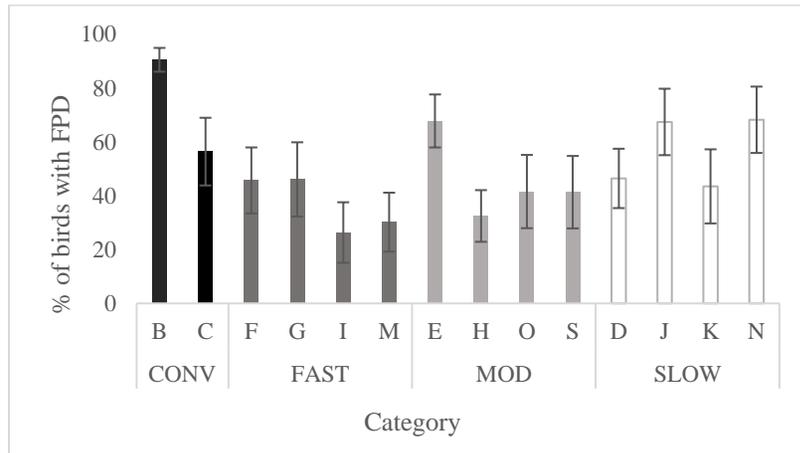
¹ BW from focal birds tested in the LTL test. Birds were weighed on the same day the test was conducted.

^{a-c} Different superscripts within the same row represent significant differences between categories for each sex (P<0.05).

^{y-z} Different superscripts within the same column for the same parameter represent significant differences between sexes in each category (P<0.05).

Effect of Strain on Incidence and Severity of Footpad Dermatitis (FPD) and Hock Burns (HB) at Target Weights 1 and 2.

A)



B)

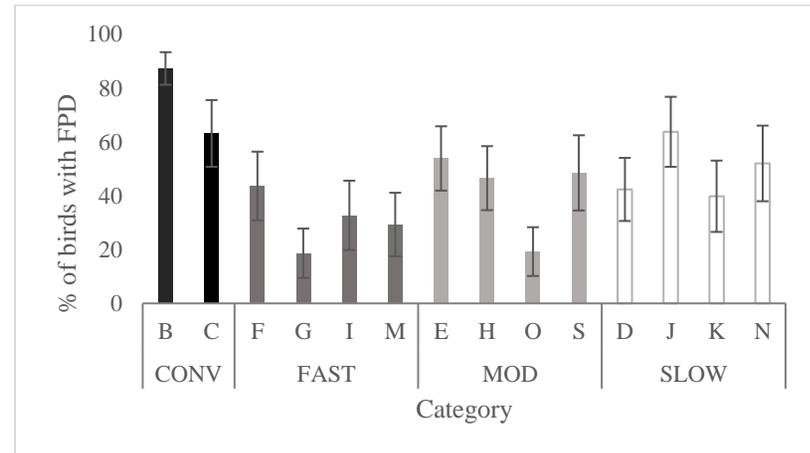


Figure A.2.1. Effects of strains (within category) on total incidence of footpad dermatitis (FPD) (LS-means \pm SEM) at Target Weights 1 (A) and 2 (B). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

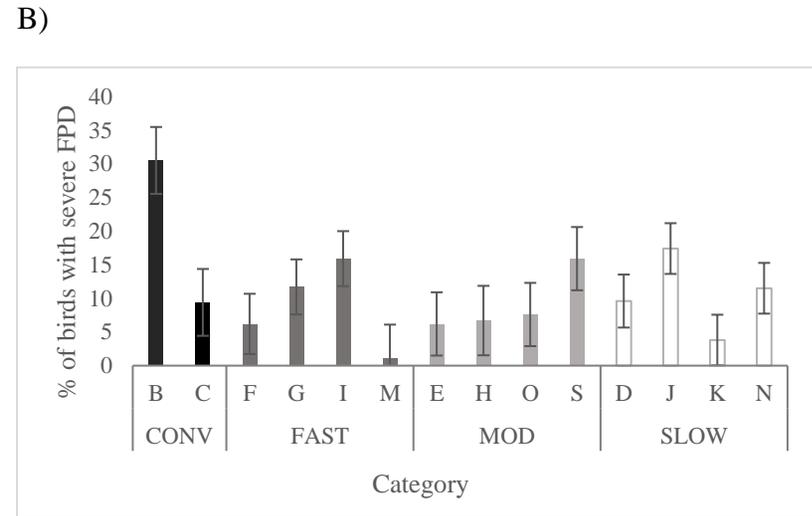
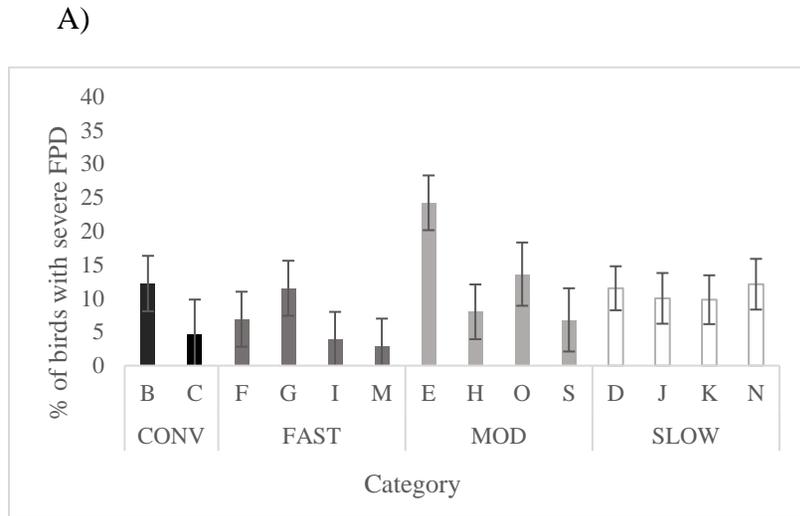


Figure A.2.2. Effects of strains (within category) on total incidence of severe footpad dermatitis (FPD) (LS-means \pm SEM) at Target Weights 1(A) and 2 (B). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

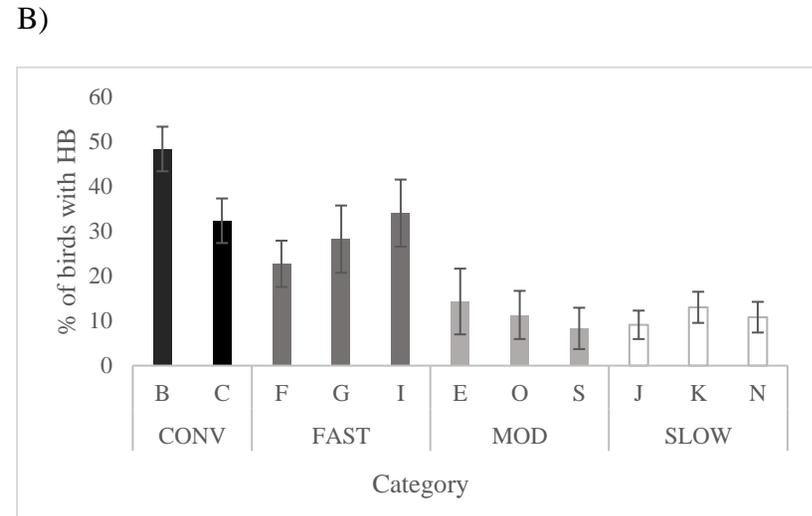
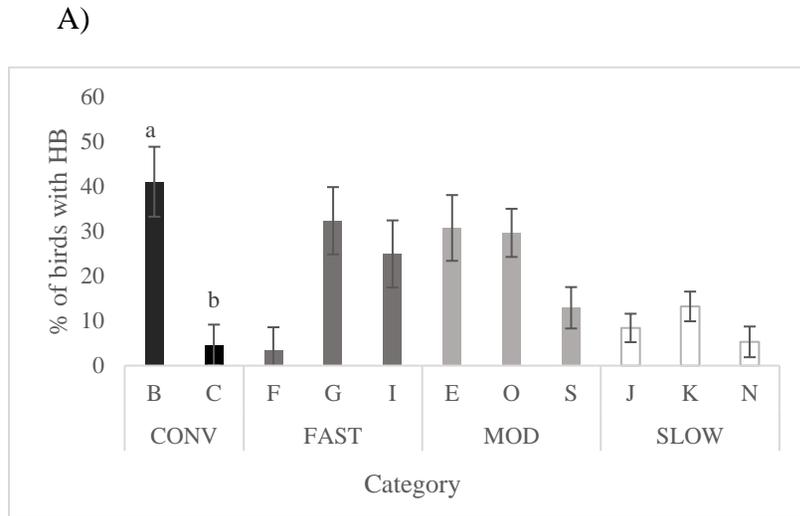


Figure A.2.3. Effects of strains (within category) on total prevalence of hock burns (HB) (LS-means \pm SEM) at Target Weights 1 (A) and 2 (B). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively. Within category, columns with different superscripts differ ($P < 0.05$).

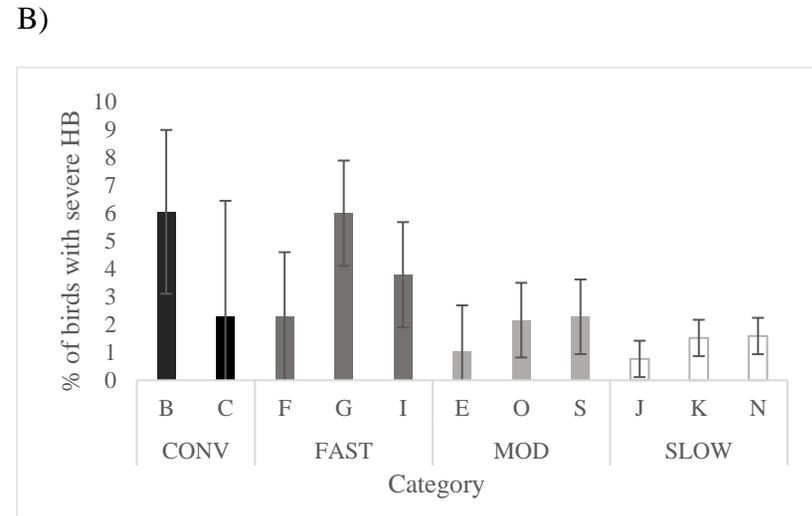
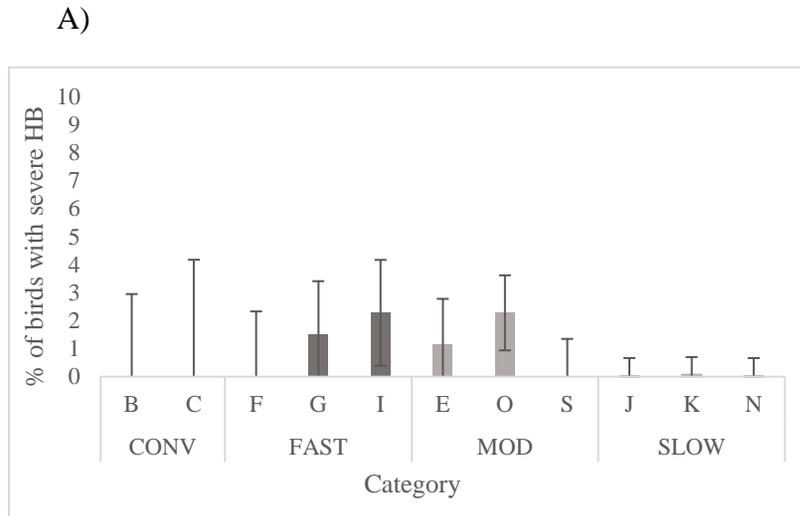


Figure A.2.4. Effects of strains (within category) on total incidence of severe scores of hock burns (HB) (LS-means \pm SEM) at Target Weights 1 (A) and 2 (B). At Target Weight 1, CONV and other categories were 34 and 48 d of age, respectively. At Target Weight 2, CONV and other categories were 48 and 62 days, respectively.

Differences in Litter Moisture among Strains from Day 14 to Day 56.

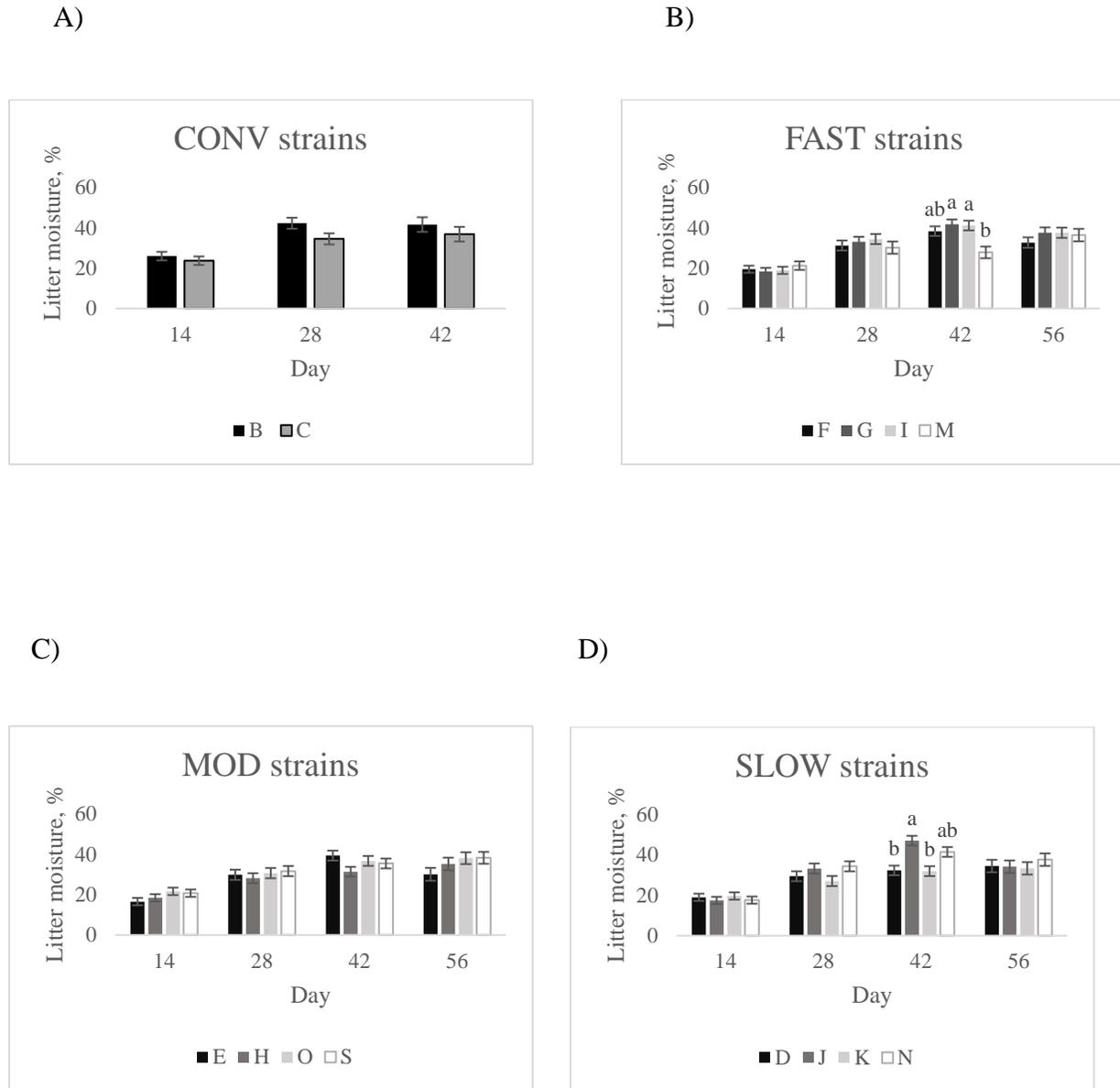


Figure A.2.5. Litter moisture (LS-means \pm SEM) among CONV (A), FAST (B), MOD (C) and SLOW (D) birds from 14 to 56 d. Litter moisture content from CONV birds at 56 d is not presented because pens containing CONV birds were processed at 34 and 48 d.