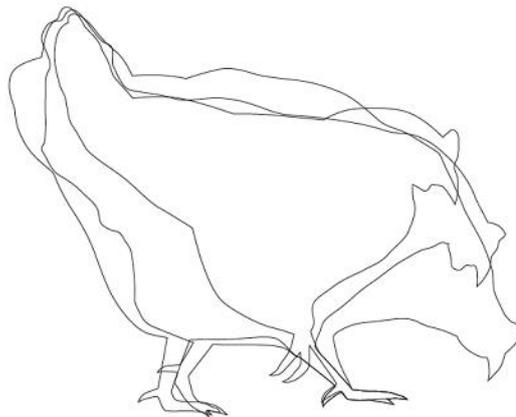


Introduction essay to PhD studies

Hatchery stress in laying hens

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1 Introduction and aim

In Swedish industrial egg production, layer chickens are during their first day in life exposed to several events which are potentially stressful. In a range of species, including chickens, early stress has been shown to affect physiology and behaviour in a short as well as long term perspective. However, it is not very well investigated whether the procedure in commercial hatcheries is a stressful event for the day-old chicks and to what extent it can further affect their behaviour, physiology and gene expression.

The aim of this essay is to clarify the stress concept and discuss behavioural and physiological effects of stress in general and early stress in chickens in particular. Further, to put stress in a context of industrial animal production and discuss its possible negative influence on production animal welfare.

2 The origin and breeding of the chicken

The ancestor of the domestic chicken, the Red Junglefowl, was domesticated already about 8000 years ago in south-east Asia for meat, egg and cock fighting. In the beginning of the 20th century, a direct selection for production traits in farm animals commenced and the poultry, that was not an exception, became intensively bred for either meat or egg production. The meat producing broiler chicken is today full grown at 35 days of age with a slaughter weight of 1,9 kg (Jordbruksverket 2017), whereas the egg laying hybrids lays about 300 eggs per year (Jordbruksverket, 2005). This can be compared with the Red Junglefowl which as full-grown weighs about 500-1000 g and lays 4-8 eggs per year.

90-95 % of the animals used in Swedish egg industry are hybrids of the breed White Leghorn, whereas the rest are brown hens mostly of the breed Rhode Island Red. The choice of breed is not so much a question of difference in production capacity as it is a cultural question. In Sweden, the White Leghorn has traditionally been used, whereas for example in England, the request for brown eggs are higher than for white. Some countries help consumers distinguish between caged and free-ranging hens by using white hens for cage and brown breeds for free-range.

3 Stress in animals

There are several definitions of stress but not really any consensus about how to define it. When Hans Selye (1973) first introduced the stress concept, he explained it as the body's response to external challenges. Dantzer and Mormède (1983) expanded upon this definition: “[Stress is a] *reflex reaction that occurs ineluctably when animals are exposed to adverse environmental conditions, and which is the cause of many unfavourable consequences, ranging from discomfort to death*”.

Another well-used model of describing stress is ‘body’s reaction to something threatening homeostasis’. The stress response starts when a potential threat is perceived and can be either behavioural, autonomic, neuroendocrine or immune, however in most cases, a combination of those (Moberg, 2000).

3.1 Physiological effects

Stimulation of an acute stressor activates the sympathetic nervous system which causes a release of adrenaline and noradrenaline in the blood. These hormones immediately increase heart rate and blood pressure and prepare the body for fight or flight. Blood is allocated primarily to brain, heart and muscles whereas blood to other organs, for example the skin, is unprioritized. Adrenaline and noradrenaline increase muscle strength and heart rate in order to make you able to run faster or fight harder. The sympathetic nervous system is a powerful tool in an acute situation, however, has a very short duration (Hemsworth and Barnett, 2000).

In parallel with activation of the sympathetic nervous system, the body activates the hypothalamic-pituitary-adrenal-axis (HPA-axis), which is here further described based on Matteri et al., 2000. The HPA-axis involves hypothalamus (H), the pituitary gland (P) and the adrenal glands (A), which together regulate several processes in the body such as digestion, immune system and energy storage.

When facing a stressful event, the hypothalamus releases vasopressin and corticotropin releasing hormone (CRH), which causes the pituitary gland to start producing adrenocorticotrophic hormone (ACTH). ACTH, also known as corticotropin, causes a release of glucocorticoids, mainly cortisol and corticosterone (CORT) from the adrenal glands which are hormones we sometimes describe as ‘stress hormones’. Also adrenaline

from the sympathetic nervous system stimulates the release of ACTH and thereby the activation of HPA-axis. Circulating glucocorticoids in turn act at receptors (GR) in the hypothalamus and pituitary to suppress the production of CRH and ACTH in a negative feedback loop (fig. 1).

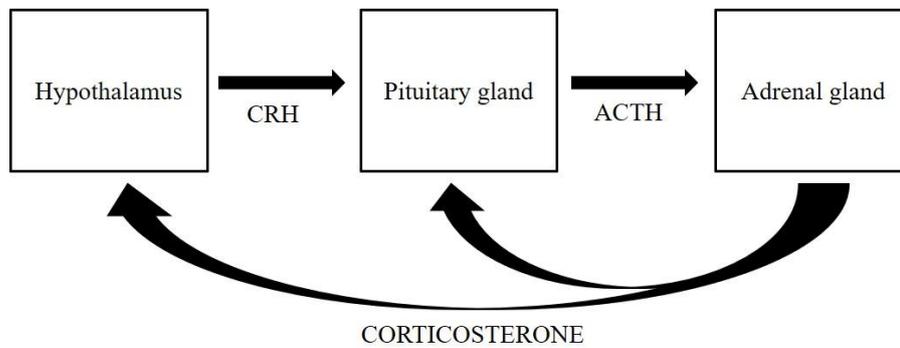


Fig 1. The hypothalamic-pituitary-adrenal-axis (HPA-axis) which are activated during stress

Cortisol and corticosterone (CORT) have several direct impacts on the body in order to create an energy boost, for example by increasing blood pressure and blood sugar. These hormones are in research regularly measured via blood sampling and used as indicators of stress.

However, high CORT level does not automatically equal experience of stress. The release of CORT is regulated by several underlying mechanisms explained in for example Kappler and Meaney, 2010 and Keller-Wood, 2015. In general, stress enhance the production of CORT but individuals experiencing the same stressor can produce more or less CRH and/or ACTH which in turn leads to individual differences in the production of CORT. There is also individual variation in the number of glucocorticoid receptors in the hypothalamus and/or pituitary where individuals with more receptors has a more effective feedback loop and take shorter time to suppresses the production of CORT.

It has been shown that the effectiveness of the HPA-axis feedback loop is not only heritable but also dependent on life experiences. Kappler and Meany (2010) showed that offspring to mothers that performed more grooming behaviour show increased

glucocorticoid receptor expression, enhanced negative feedback sensitivity to glucocorticoids, reduced CRF expression in the hypothalamus and lower pituitary-adrenal responses when stressed. The two offspring groups in this study differed in behaviour responses to stress later in life as well as gene expression. Goerlich et al. (2012) conducted a stress study on birds and showed that offspring to stressed parents differ from offspring to non-stressed birds in ability to cope with stress, also suggesting a transgenerational suppression of the HPA-axis.

To conclude this paragraph, CORT level is often used as an indicator of stress in animals but there are reasons to question the reliability of using only CORT results to prove stress. We now know that a high level of CORT not always correlates with the individuals *experience* of stress. Measuring stress hormones should therefore be combined with e.g. behaviour studies, physiological parameters such as heart rate or measurements of glucocorticoid receptors in the brain.

3.2 Behavioural effects

From a behavioural point of view, an animal is defined as ‘stressed’ when it performs behaviours that differ from normal (Mason, 1991). These behaviours are usually termed ‘abnormal’ or ‘stereotypic’. ‘Abnormal behaviours’ is an umbrella-term which cover all behaviours that are not included in the animals’ usual behaviour repertoire, e.g. tail biting (Brunberg et al., 2011) and cannibalism (Keeling et al., 2004), but can also be normal behaviours expressed more frequently than usual, e.g. overgrooming (Amat et al., 2016) and polydipsia (Roberts, 1997).

Stereotypies are defined as repetitive behaviour patterns without goal or function, which are associated with past or present stress derived from a sub-optimal environment or management. Examples of behaviours included in this category are tongue rolling, weaving, crib biting, pacing and bar biting. Note that stereotypic behaviours always are abnormal whereas abnormal behaviours not necessary are repetitive and therefore per definition not stereotypic (for further reading, see Mason 1991).

Behaviour abnormalities are more or less consistent within species and can be measured by systematic ethological studies. For example, minks develop pacing or other heterogeneous movements (Mason, 1993), pigs tail- and bar-biting (Brunberg et al., 2011; Schroder-Petersen and Simonsen, 2001), ruminants oral behaviours such as tongue rolling and

polydipsia (Roberts, 1997; Baxner and Plowman, 2001; Redbo et al., 2010) and chickens feather pecking and cannibalism (Keeling et al., 2004). Abnormal behaviours should always be interpreted as alarm calls for stress and/or frustration which derives from a stressful or barren environment. The behaviours are the animal's attempt to cope with a situation and should therefore when once developed not be hindered. Additionally, researchers agree that when stereotypic behaviours are performed, endorphin releases which makes the individual feeling better (Cronin et al., 1986). When once developed an abnormal behaviour, the animal tends to keep on to it even when moved to a new environment. The most effective way of treating, or preferably prevent, behaviour abnormalities is by environmental enrichment (Dallaire et al., 2012).

Stress may in the long run have several negative consequences for the animal and research has investigated this in a range of different aspects and species, for example changed behaviour (e.g. Lidfors, 1996; Stêhulová et al., 2008; Cirulli et al., 2009; Franklin et al., 2010), impaired immune function (Kelley, 1980), impaired cognitive functions (Li et al., 2008), affected HPA-axis and stress hormone levels (Shinder et al., 2002; O'Mahony et al., 2008 *in press*), decreased feed intake and growth (Yahav and Hurwitz, 1996; Altan et al., 2000 Krohn et al., 2010), disturbed reproduction (Shini et al., 2009) and affected parenting- (Stone and Bales, 2010) and sexual behaviour (Sachser and Kaiser, 1996).

4 Early stress

Early development is in the literature defined as the period from conception to maturity, which is quite a broad definition (Lindström, 1999). In this essay, early stress is regarded as a stressful occasion happen in the first weeks of an individuals' life. Most research on this period of life has focused on the effects of maternal separation since this is considered as a traumatic experience for a young animal. Further, most of this work is carried out on inbred strains of mice or rats in laboratory environments.

4.1 Early stress in animals in general

Studies in rodents have shown that early separation stress has consequences in the adult animal, for example in terms of increased alcohol consumption and increased startle response (reviewed in Cirulli et al., 2009). It has also been shown to increase the duration

of learned helplessness later in life, a behaviour interpreted as depression since it could be reversed by treatment with antidepressant agents (Franklin et al., 2010). Further, early weaned rats have higher levels of corticosterone at an age of 8 weeks compared to later weaned rats (O'Mahony et al., 2008).

In farm animals, early maternal separation is often a routine for management reasons and production efficiency. The stress that comes with this affects the offspring in a range of different aspects which some are described below.

Early-weaned calves are more inactive and less likely to engage in social play than later weaned calves (Lidfors, 1996; Stêhulová et al., 2008) whereas early weaned piglets are more active and aggressive compared to later weaned littermates (Fraser, 1978; Colson et al., 2006; Orgeur et al., 2001). Calves as well as piglets and mink pups show after early weaning more abnormal and other stereotypies than later weaned littermates (Lidfors, 1996; Jeppesen et al., 2000; Dybkjaer, 1992). Early weaning also affects growth and feed intake in farm animals such as pigs (Leibbrandt et al., 1975), calves (Krohn et al., 2010), goats (Louca et al., 1975) and rabbits (Xiccato et al., 2003).

4.2 Early stress in chickens in particular

From what we know about maternal separation stress in other animals, it is somewhat surprising there is no existing debate about that chickens in poultry farming are not only raised separated from the hen but never even get to meet her. Neither the chicken industry, nor the poultry research has focused on the possible stress of this, probably because this is perceived to be of little relevance from a production point of view. However, from what we already know about other animals, one can assume that hatching and raising in absence of a mother affects chickens, although we still don't know how.

In terms of early stress, poultry research has rather than separation stress focused on temperature- and feed stress, probably because these are parameters relevant when raising broiler chickens for meat. Poultry are sensitive to temperature stress and affected by this in several ways (e.g. Faisal et al., 2008). Early heat stress in broiler is shown to result in depressed weight gain (Yahav and Hurwitz, 1996) which is not yet compensated for at slaughter 35 days later (Altan et al., 2000). As well as heat stress, repetitive cold stress in early age (3-4 days) elevates plasma corticosterone concentration and affects weight gain (Shinder et al., 2002).

Early food restriction causes stress in birds and has been shown to affect body weight in for example Japanese quail (Gebhardt-Henrich and Marks, 1995; Hassan et al., 2003), broiler chickens (Urdaneta-Rincon and Leeson, 2002; Yumova et al., 2002), turkeys (Tumová et al., 2002), zebra finches (Krause et al., 2009) and White leghorns (Mashaly et al., 2004). However, quails (Gebhardt-Henrich and Marks, 1995) as well as broilers and turkeys (Tumová et al., 2002) have been shown to compensatory grow after feed restricted. Zebra finches raised under poor nutritional conditions were later in life more likely to engage in foraging behaviour and other exploratory behaviours compared to birds raised under normal conditions (Krause et al., 2009).

There are some studies investigating other aspects of early-life experiences in chickens. For example, birds kept under dark brooders after hatching show lower levels of feather pecking than those kept under light brooders (Johnsen and Kristensen, 2001; Jensen et al., 2006). Chickens brooded by hen during rearing period increase feeding activity and perform more ground pecking than non-brooded chicks (Roden and Wechsler, 1998; Wauters et al., 2002; Riber et al., 2007). There are also lower mortality levels due to cannibalism and feather pecking when the hen is present (Riber et al., 2007).

4.3 Prenatal stress

There is a range of scientific studies on different species that show how several types of prenatal stress affects the foster in different aspects. For example, chronic maternal stress in humans during pregnancy is associated with increased plasma levels of CRH, ACTH and cortisol which may increase preterm birth, delayed development and behaviour abnormalities in the offspring (Weinstock, 2001). Repeated transportation stress in cows during pregnancy alters the calves' physiological response to stress which assumable has impact on the animals' ability to cope with stress (Lay et al., 1997). In rats, a short period of restraint during pregnancy elevates ACTH (McCormick et al., 1995) and increase basal corticosterone level (Szuran et al., 2000) in female offspring, but not in males. An unstable social environment for a pregnant guinea pig is shown to affect social- and mating behaviour in the daughters, which display high amounts of male-typical courtship, play and social orientation (Sachser and Kaiser, 1996).

The timing of maturation of the HPA-axis is species specific and is related to brain development. In the group of precocial species, in which we include the domestic chicken,

the maximal brain growth and neuroendocrine maturation takes place before birth or egg laying, and therefore, prenatal stress has been shown to have a great impact on the offspring in several aspects (reviewed by Matthews, 2002). Early-life history, laying, brooding and rearing conditions in chickens are investigated by several scientific studies and shown to have profound effects on behavioural development in the bird (reviewed by Rodenburg et al., 2008).

Exposure of the egg to light is one of the factors affecting the chicks development. Research has shown that chickens exposed to 2 h light on incubation day 19 show higher levels of early feather pecking than chickens brooded under darkness (Riedstra and Groothuis, 2004). Feather pecking is suggested to be a behaviour of exploration which is a lateralized behaviour and the lateralization of brain functions is shown to be stimulated by exposure to light in late embryonic development (Deng and Rogers, 2001).

Not only light affects the chicken embryo. For example, research show that temperature influences both hatchability and plasma corticosterone levels (Yahav et al., 2004). Reduced gas exchanges during incubation impair hatchability drastically to about 10 % (Rodricks et al., 2004) as well as the chickens body weight (Camm et al., 2001) and cognitive functions such as learning and memory (Camm et al., 2001). Exposure of the egg to corticosterone may have negative influence on imprinting (Nordgreen et al., 2006), growth and mortality (Eriksen et al., 2005), whereas exposure to testosterone increase growth (Schwabl, 1996), boost muscular growth (Gil, 2003), begging rate (Schwabl, 1996; Gil, 2003), social rank (Schwabl, 1993), nuptial plumage (Eising et al., 2006) and aggressive and competitive behaviour (Schwabl, 1993; Eising et al., 2006).

4.4 Hormesis – what does not kill you makes you stronger

There are two different but somehow related definitions of hormesis: (1) Exposure of certain substances or stressors can be beneficial at low exposure levels while toxic at higher, and (2) exposure of these stressors early in life can improve functioning later in life (e.g. Rostagna, 2009, Costantini et al., 2010). The later definition is the one used in this essay.

The theory behind hormesis is that exposure to a certain stressor, for example a dose of toxin or a behavioural stress, that in higher dose would be harmful or even lethal, has a beneficial effect since the individual somehow copes better when facing low levels of the

same stressor later in life. Sometimes hormesis is referred to as ‘priming’ or ‘conditioning’ (Costantini et al., 2010). Another way of thinking about it is as ‘stress hardening’ (Schreck, 2010).

Most of the hormesis research is to be found in fields of toxicology but has later been applied in ecology as well as evolutionary biology, however primarily on temperature stress and starvation (Table 1). The theory of hormesis enable scientists to regard environment not only as permissive for development but crucial to calibrate and program the animals’ stress system.

Species	Stressor	Hormetic response	Reference
Broiler chickens	Heat	Increased weight gain and feed intake. Decreased mortality rate during heat stress	Yahav, 1999
Broiler chickens	Heat	Increased ability to cope with heat stress	Yahav and Hurwitz, 1996
Broiler chickens	Heat	Increased body weight	Yahav and Mc Murtry, 2001
Broiler chickens	Cold	Increased ability to cope with cold stress	Shinder et al., 2002
Rodents	Feed restriction	Longevity	Reviewed by Le Bourg, 2003
Mice	Radiation	Longevity	Caratero et al., 1998
Drosophila Melanogaster	Heat	Increased ability to cope with cold stress	Bruton et al., 1988
Caenorhabditis elegans	Heat	Longevity	Olsen et al., 2006
Zebra finches	Feed restriction	Increased body weight, increased exploratory and foraging behaviour	Krause et al., 2009

Table 1. An overview of hormesis studies in animals

Hormetic responses seem to occur for some traits but not for other, even when measured at the same time point. Costantini et al. (2010) discuss that this might be due to that some traits simply do not exhibit this kind of response. Further, it might be obvious but yet worth mentioning that the same stressors’ hormetic effect differs between species. Further, the timing of the first exposure may be important, and when planning an experiment, it is worth considering if there is a known specific stage or time window when the species in question is more sensitive to a certain type of stressor.

Furthermore, it is shown that hormetic responses can generalize across stressors, meaning that one early-life stressor does not only help the individual to later in life better cope with this particular stressor but also with other stressors (Le Bourg and Minois, 1997; Cypser and Johnson, 2002; Hall et al., 2002; Le Bourg et al., 2004; Le Bourg, 2005). Yahav and McMurtry (2001) investigated the sensitivity to thermal conditioning in chickens depending on age and temperature. They exposed different groups of broiler chickens to different temperature in different ages to study chickens’ ability to cope with acute heat

stress later in life. Yahav and McMurtry (2001) found that 36.0 to 37.5°C applied at 3 days of age is the optimum temperature and age for thermal conditioning in broiler chickens. These chickens had a growth retardation followed by a compensatory growth period which resulted in higher body weight than control chickens as well as chickens heat stressed at other temperatures and time points. Further, the study showed a non-significant trend that these chickens overall had lower feed intake than the other groups that together with a higher body weight suggest an improved feed efficiency.

Hormetic responses is shown to be inheritable. In the earlier briefly mentioned study of Goerlich et al. (2012) birds were stressed with isolation 1-3 hours per day between the age of 9-26 days of age. The results show no effects on baseline corticosterone level, however, there was a significant difference in comparison to control birds in suppression of the HPA-axis after acute stress. Similarly, the offspring of stressed parents recovered faster from acute stress than offspring to control parents, suggesting a transgenerational suppression of HPA axis.

Hormesis studies can be complicated since it is important that you know exactly how strong the early-life stressor has to be for making a difference, however not strong enough to be harmful (as discussed above in the paragraph 'Early stress'). Also, experiments need to be well designed since we yet know little about the costs and cons of stress priming. To consciously expose an animal to stress should always be regarded critically, especially in young age when the individual is potentially more vulnerable in several aspects, and these studies should therefore be discussed from an animal welfare point of view.

5 Egg production in Sweden

Commercial egg production is highly standardised. Companies such as Lohmann and Hy-line produce the birds used in egg production worldwide – no genetic selection is taking place in Sweden. Strains of animals are bred for specific traits such as egg productivity, fitness, life expectancy, behaviour, disease resistance and egg quality in order to, when crossed, produce a robust hybrid (Hy-line.com). These hybrids are imported to Sweden and reared at specialized so called 'grandparent farms'. The offspring of these birds are usually reared at yet another farm in a 'parental flock'. The eggs from these parental birds are transported to commercial hatcheries and the chickens hatched in incubators at these are the birds used for consumption

egg production. After the hatchery process, in more detail described below, birds are transported to pullet farms which keep the animals up to sexual maturity at an age of 16-18 weeks. After sexual maturity, these birds are transported to egg farms. The egg laying hens are slaughtered at about 75 weeks of age.

5.1 The hatchery process

In Sweden, there are two commercial hatcheries for laying hens; Swedfarm in Lingham and Gimranäs AB in Herrljunga. Together, these two companies produce almost 8,2 million animals per year distributed on 2897 egg farms in Sweden (Svenska ägg, 2016).

The hatchery procedure is briefly described by Mauldin (2002), however, the information in the following passage derives mainly from personal observations as well as personal communication with Gimranäs AB.

The hatchery process starts when fertilized eggs arrive to the hatchery and are inserted in incubators. The incubators are calibrated for optimal hatchability which includes turning of the eggs as well as temperature and humidity fluctuating over the day. At day 18, the eggs are moved to hatching racks for the last days of incubation. The major part of the eggs hatch at day 21, but commercial hatcheries usually leave the eggs until day 22 to maximise hatchability rate.

When hatched, the racks with chickens are tilted on a conveyer belt and the shells are removed, either by hand or mechanical. The chickens are conveyed to a sex sorting station where they are manually sorted by either colour (only brown breeds), wing or vent sexing. Venting chickens involve squeezing out any feces remain in the intestines which opens up the chicken's cloaca. With left thumb, the left side of the vent can be lifted up and over while right thumb and index finger pulls back the right side of the vent which makes the inside of the cloaca and the sexual organs visible. A chicken sexer is expected to sex a substantial number of animals per minute.

Male chickens are culled immediately after this procedure since they are of no use for the production, traditionally through maceration by using a high-speed grinder but hatcheries in Sweden have in recent years moved to asphyxiation by carbon dioxide.

After sex sorted, females are transported via conveyer belts to the vaccination station. Vaccination can be either manual or mechanical. In most countries, de-beaking is also a

part of this procedure, however not legal in Sweden. Further, the birds are via conveyor belts transported, automatically counted and packed into boxes which are transported with truck to breeding farms. Before the counting station, the conveyor speed is drastically increased to increase distance between individual birds and thereby increasing the precision in the automatic counters. Sometimes the transport time exceeds 14 hours.

5.2 Housing systems

Sweden has four housing systems for egg laying hens; furnished caged, free-ranging indoor, free-ranging outdoor and organic production. Un-furnished caged are since 1997 not legal in Sweden. Pullets at breeding farms shall according to Swedish legislation be held in a similar system to which they are aimed to be housed as adults. Swedish legislation referred to in this paragraph is to be found in Statens jordbruksverks föreskrifter och allmänna råd [SJV 2010:15] om djurhållning inom lantbruket m. m., saknr L 100.

The legislation in terms of space varies between the different systems. For birds in cages, the minimum area per animal is 0,06 m², however, the cage should not hold more than 16 birds. Free-ranging systems consist of either one or several floor levels with a maximum of three. In one level system, you can have 9 birds per m² whereas in two or three level systems, you can house no more than 7 hens per m² of the total area and no more than 20 animals per m² floor area.

In organic farming, you can have no more than 6 birds per m² and no more than 3000 animals per stable, according to the Swedish organization KRAV (2018). Hens in organic farming should be free-ranging and be given the opportunity to go outside whenever the weather allows.

Over the last years, the demand for organic eggs has increased in Sweden. At the same time, several of Sweden's biggest grocery chains stop selling eggs from caged hens and these two factors had led towards a trend for farmers to switch to free-ranging systems. In the end of 2016, the distribution between the different housing systems were 64,9 % free-ranging indoors, 3 % free-ranging outdoors, 15,4 % furnished cages and 16,7 % organic (Svenska ägg, 2016).

6 Possible welfare implications

The layer chicks hatched in commercial hatcheries are during the first hours in life exposed to a procedure potentially stressful to them, which is what is discussed in this paragraph. The paragraph, and this essay, finishes off with question formulations aimed to be investigated and answered during the following five years of my PhD.

To start with, incubators in which chickens are hatched are programmed for optimal hatchability, however not for animal welfare. There are few studies on incubator conditions' effect on chickens, however parameters such as light (Archer et al., 2009), temperature (Ande and Wilson, 1981; Iqbal et al., 1990) and turning of eggs (Elibol and Brake, 2003) have been shown to affect feeding activity (Archer et al., 2009), development (Archer et al., 2009), hatchability (Ande and Wilson, 1981; Elibol and Brake, 2003) and body weight (Iqbal et al., 1990). These results indicate that conditions in incubators and hatcheries to some extent are affecting chickens in several ways, however the magnitude of this is not yet fully investigated.

After hatching, the treating of animals is optimized for optimal survival rate rather than welfare. The procedure, which includes vaccination and sex sorting by vent or wing, might not only be stressful but also painful to the animal.

The male chickens are immediately after sex sorting culled by use of carbon dioxide. Carbon dioxide is in several studies shown to be aversive and is a stressful as well as painful way of anaesthetising and/or euthanising animals (e.g. Conlee et al., 2004; Nowak et al., 2007). However, Blackshaw et al. (1988) recorded behavioural patterns in chickens during euthanasia by chloroform and carbon dioxide respectively to investigate stress levels in the animals. The study showed that behavioural patterns were similar in carbon dioxide and chloroform, however, the animal collapsed and reached dead faster in carbon dioxide why this gas was recommended. Coenen et al. (2007) compared chicken behaviour and time to reach death in a gas mixture of carbon dioxide-oxygen-nitrogen with a mixture of carbon dioxide-argon with regard to animal welfare. Time to reach unconsciousness was longer in carbon dioxide, however in argon, even though they died faster, birds immediately showed signs of agitation and distress such as muscle contractions, why the authors also in this study suggested use of carbon dioxide when anaesthetising chickens. In the poultry industry, the practical alternative to carbon dioxide is maceration which means culling by use of a high-

speed grinder. The welfare of this is to my knowledge not investigated nor discussed, however, the fast death provided by a high-speed grinder might be the quickest and least painful alternative for culling.

According to a publication from Agriculture Canada (1989), high-speed maceration of chicks is a practical and humane method since, when properly designed, death occurs instantaneously. Carbon dioxide might be suitable for chicks but high concentrations are required because day-old chicks are resistant to the gas (Agriculture Canada, 1989).

The female chickens on the other hand, are further processed. Handling of birds is in research generally regarded as a welfare problem since it includes moments of fear, frustration, pain and discomfort, and there are even studies arguing that conveying is potentially less stressful than human handling (reviewed by Scott, 1993). One of these, a study of Scott and Moran (1993) investigated fear level by tonic immobility test on five groups of hens: Group 1 was transported on a flat conveyor belt; Group 2 was conveyance by hand; Group 3 was conveyance by a processing shackle; Group 4 was exposed to noise from a conveyor and Group 5 was a control group exposed to tonic immobility only. The results show that inversion and conveyance by human handling or a shackle is significantly more stressful than transporting by a mechanical conveyor belt. The authors suggest that using a carefully designed conveying system might improve animal welfare.

In many countries, however not in Sweden, laying hens are beak-trimmed or debeaked as a part of the hatchery procedure to prevent feather pecking and cannibalism later in life. One third of the upper beak is removed by use of a blade or a machine that punch a hole which then causes the end of the beak to fall off in a few days. Regardless of method, this is performed without anaesthesia. The beak is innervated, and it has been shown that when amputated, the severed nerves form neuromas which spontaneously send pain signals to the brain (Beward and Gentle, 1985). This can be compared with phantom limb pain in human. Additionally, the acute pain when debeaked causes behavioural changes that indicates pain lasting at least 5-6 weeks after trimming (Duncan et al., 1989). Beak trimming is however, as already mentioned, not a part of the hatchery process in Sweden since it is illegal due to the animal legislation (10 § Djurskyddslagen [1988:534]).

Because there are only two commercial hatcheries that supply Sweden with layers, some of the animals are transported all the way from south of Sweden to north. Most birds from

Swedish hatcheries are transported by road but live animals are also exported from Sweden to other countries, why other modes of transportation can occur as well.

All transporting of animal involves placement of animals in containers. The animals are then loaded on the transport vehicle, transported and unloaded when arriving to the end station. Poultry are during transport exposed to a number of different stressors that may influence the welfare of the animals, for example thermal challenges, withdrawal of food and water, social disruption, noise motion, vibrations and acceleration (reviewed by Mitchell and Kettlewell, 2009). These factors may all increase stress levels in the birds, however, heat stress is the parameter which is regarded as the major threat to both well-being and productivity (reviewed by Mitchell and Kettlewell, 2009). Solutions to welfare issues with transportations could be for example stricter regulations of maximal transport time and redesigning of transport containers and vehicles, such as example by changing the ventilation system.

From this introduction essay, following questions are formulated:

1. Are newly hatched chickens at Swedish commercial hatcheries stressed after the hatchery procedure?
2. Can the handling of chickens at these hatcheries be regarded as a welfare issue?
3. Is the assumed stress at these hatcheries something that affects the animal later in life?
4. With regard to the concept of hormesis, is it possible that handling during the first hours in life might prepare the animal to stress and primes it to better cope with stress later in life?
5. Which are the physiological and epigenetic mechanisms that modify any possible later responses to hatchery stress?

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