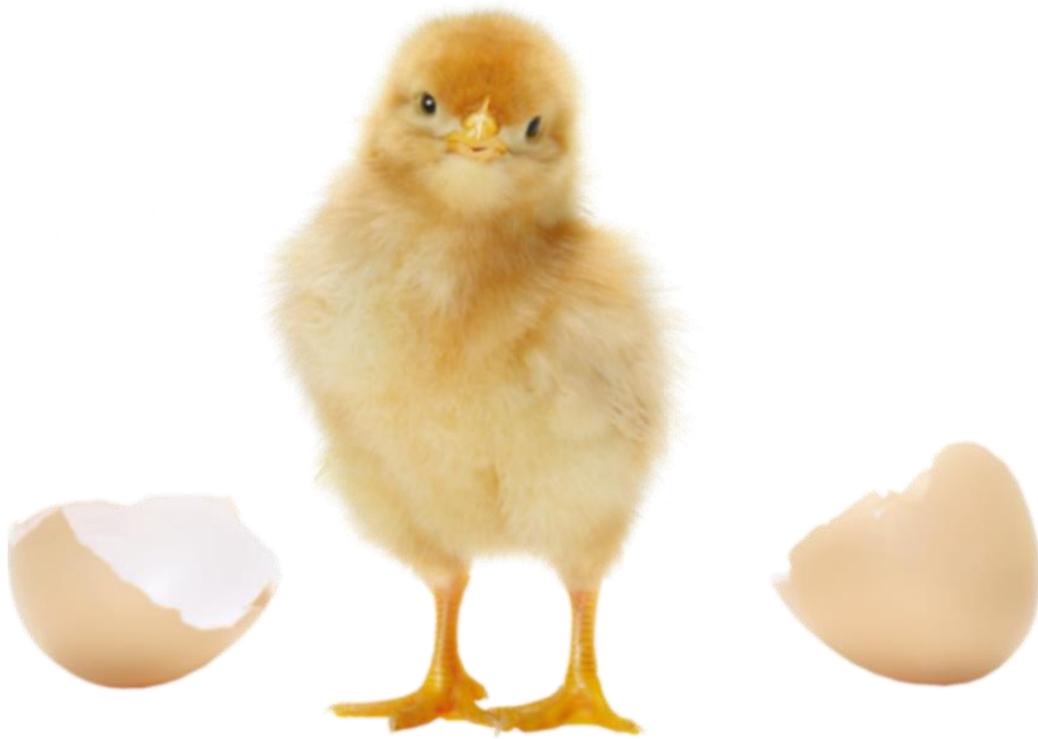


Effect of hatchery, breed and breeder flock age on egg weight loss and hatchability



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Preface and acknowledgements

As part of my European Engineer Degree course with a Livestock Production option, I had the opportunity to go on a 14-week internship in Belgium at the company Pluriton bvba. During this placement, I had the opportunity to discover an unknown aspect of the poultry chain. I was given the chance to conduct a research study for the hatchery sector in an aim to evaluate and understand hatchery technical factors and results.

I would first like to thank Marlies Broeckx who had the great kindness to accept me in this company as well as Inez who was always available to answer to my multiple question about my work or the everyday life around.

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Summary

During incubation, an embryo is developing in the egg for a period of 21 days. During the incubation process air exchange occurs between the egg and its external environment during which it loses humidity due to water diffusion through the shell. This loss in humidity can affect chicks' quality and, in case of extreme weight loss it increases embryonic mortality and thus may affect hatchability. To emphasize the importance of the before mentioned, hatchability is the biggest economic value for hatcheries. By controlling the egg weight loss factor, hatchery can ensure good quality chick and better hatchability.

The objective of this research was to investigate the influence of breeder age flock, breed and hatchery on egg weight loss and hatchability in order to give information as a guide for hatcheries and help them to possibly adapt their incubation profile to breed or age and improve their results.

This research has been realized in two hatcheries with two different incubation profiles on breeder flock age ranging from younger than 40 weeks old to older than 60 weeks old and on seven different breeds. The egg weight loss has been measured by weighing all eggs individually before setting and at transfer. Hatchability has been found by counting the difference between the quantity of non-hatched eggs and the total of eggs counted at set for each tray.

It has been found that all independent variables such as the hatchery, the breed and the age of breeder flocks have an effect on both egg weight loss and hatchability. Hatchery in Arendonk showed a more beneficial incubation program than Afferden. Furthermore, all results accords that an egg weight loss ranging between 8% and 16% allows a hatchability higher than 80% and that productivity significantly decrease for loss in humidity lower than 8% or higher than 16%. For two breeds out of the seven, egg weight loss lower than 8% and higher than 16% does not affect the hatchability. Also, egg weight loss from hens younger than 40 weeks is mostly distributed between 8% and 12% while the distribution is wider for the hens older than 60 weeks. According to the literature hatchability decreases in hens older than 60 weeks while loss in humidity increases.

In conclusion, it is recommended to manage egg weight loss in to order to keep it in a 8% to 16% range. Nevertheless, according to the literature, this range would provide a good hatchability but would not guaranty a good quality of chicks, as long as the recommendations are 11% to 12% of humidity loss to get a healthy chick

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1. Introduction

The supply of good quality day-old chicks is a key for the success of the poultry production chain. Indeed, commercial hatcheries are at the first position of production chains' link for eggs or chicken meat. It is a big responsibility to supply viable chicks able to express as much as possible their genetic potential. At the hatchery, fertilized eggs are placed in incubators for an incubation period of 21 days. A successful production of day-old chicks starts with the proper selection and management of layers breeding stock, proper post-lay handling of fertile eggs and the correct incubation process by the hatchery. From the many factors that influence hatcheries' performance, for example egg age (Tarongoy et al., 1990), storage conditions (Brah and Sandhu, 1989), system of husbandry and rearing technology (Weis, 1991), age of flock (Buhr, 1995) and incubation humidity and eggs turning angle (Permsak, 1996), the hatcheries are only able to manage the ones acting on the egg when they get. This means mostly good storage conditions and good incubation process.

The main factors that the hatchery can manage precisely during incubation period are air velocity, relative humidity, eggs turning angle and the temperature of the incubator. Over 97% of the variation in chick weight at hatch can be explained by two factors, fresh egg weight and weight loss during incubation (Tullett and Burton, 2008). Egg weight loss (EWL) is one of the most important factors to manage to ensure good hatchability and quality day-old chicks. The hatchability is the biggest economic value for hatcheries but to know exactly the quality of the incubation process the best reference is the hatchability from fertile eggs (Personal communication, Kevin Clijmans). The second attention point for hatcheries is the quality of the chicks. Indeed a chick that did not lose enough weight during the incubation period will be more passive and more reluctant to feed and drink when placed in the farm (Aviagen, 2013). Opposite to the previous, a too light chick will be dehydrated and have a little yolk reserve. Both would compromise a good start in the rearing farm and so on, later in the laying farm in case of laying hens (Personal communication, Guillaume Lafargue).

There are as many incubation programs as there is different incubating equipment, but it has been reviewed that a constant temperature of 37,8°C is the thermal homeostasis in the chick embryo and give the best embryo development and hatchability (King'ori, A.M., 2011).

Egg Weight Loss and embryonic development

The factor that affects EWL the most during the incubation period is the relative humidity. In each egg, between 7,000 and 17,000 pores are involved in the gas exchange, in which these pores facilitate the egg's water vapour conductance. During incubation, an egg loses a certain amount of weight mostly due to water diffusion through the shell (Rahn et al., 1977). This water loss is due to the energy needed for the embryonic development. The energy is taken from the fat stores of the yolk and for every gram of fat burned, an almost equal mass of

metabolic water is generated (Tona et al., 2001). Losing water through the egg shell allows the embryo to build up an internal oxygen air cell that is used for the transition to lung respiration after internal pipping. The average EWL that the hatchery try to reach is between 11% and 12% of humidity lost from the fresh eggs at 18th days of incubation. The embryonic mortality increases when humidity loss is lower than, 9.1% or higher than 18.5% (Buhr, 1995). Furthermore, the water content of the hatched chicks and the residual yolk sac are both affected by water loss from the egg during incubation and account for the differences in the weight of the whole chick at hatch from eggs of the same size (Tullett and Burton, 2008).

Age and breed influences

The efficiency of reproduction of broiler breeders decreases with age that is related to the internal egg composition or ratio, larger egg weight, poor shell quality, increased early and late embryo mortality (Elibol and Brake, 2003; Tona *et al.*, 2004; Joseph and Moran, 2005)

The eggs' quality depends mostly of the age of laying breeder hens in which the albumen and eggshell quality differs between young hens and old hens (Tona et al., 2001). Young hens will give eggs with fewer pores compared to eggs from older hens. Therefore, the gas exchange between the air and eggs will be more important in eggs from old chickens, resulting in increased egg weight loss (Zakaria et al., 2009). However, Roque and Soares (1994) noticed that weight loss percent during incubation was higher for eggs produced by young breeders than that of eggs produced by old breeders disregarding shell thickness. Trehan and Bajwa (2001) did not notice any significant difference in chick weight of 42 and 50 weeks old breeders, whereas Abanikannda et al. (2007) showed that in broiler breeders, older hens produced relatively smaller chicks despite their relatively larger eggs.

Other factors affecting day-old chick quality and EWL are embryonic weight and eggshell quality. The weight of embryos from the same size eggs were found to depend on the age of parent flock, with the largest increase in embryo weight occurring in eggs from flocks between 28 and 32 weeks old (Shanawany, 1982). Chistensen (1983) and Bennet (1992) showed a positive correlation between the thickness and strength of the eggshell and the number of hatched chicks, which can be explained by the fact that a thicker eggshell has lower water vapour conductance.

Most of the studies done regarding a potential relation between the breed and the egg weight loss were done with broiler breeders. Abanikannda et al. (2007), showed that the breed significantly affected chick weight loss in broiler breeders. Also Bayel et al. (2012) showed that breed and broiler breeder's age significantly affected egg weight loss percentage and young breeders had significantly the highest egg weight loss percentage. Lately, Grochowska et al. (2019) concluded that in broilers the genotype was the most important determinant of egg weight loss percentage.

Hatchability

Hatchability is directly related to egg fertility and embryonic mortality. However, many factors influencing it have been reported. King'ori (2011) reviewed the factors that influence hatchability in poultry (turkey) and concluded that different type of factors were affecting the hatchability such as environmental and genetic factors. From the breeder factors that affect hatchability, the strain, health, nutrition, age of the flock, egg weight, egg quality, egg size and egg storage duration and conditions are the mains. Grochowska et al. (2019) that the breeder flock age and egg storage time were the most important factors responsible for variability in the percentage of fertile hatchability.

As previously seen, EWL is the main factor in the incubation process playing on day-old chick quality. Nevertheless, most of the hatcheries have only one program for all settings. In addition, even if they reach the objective of 11 to 12% in average of EWL there still might be a big difference between the highest and the lowest EWL within one setting. Fluctuating EWL can result in low hatchability, low homogeneity of the chicks and a high 7-day mortality and suboptimal flock performance.

There is limited research literature available on the on the influence of age on EWL in layer breeders as most of the published literature concerns broiler breeder research and even less research about the influence of layer breed on EWL. This created the opportunity to investigate the effects of those factors in layer breeders at Pluriton (Arendonk, Belgium). Measuring the individual percentage of EWL for different breeds at different ages will allow the hatchery to formulate precise profiles of EWL per breed and age in both hatcheries.

This way, hatcheries could adapt their incubation program, get better hatchability results and therefor, provide more homogenate and better quality DOC.

The present study aims to investigate the relationship between layer breeder's age and breed on egg weight loss during incubation and hatchability measured at two different hatcheries.

Main question:

How does breeds and breeder flock age influence the egg water loss and hatchability between hatcheries?

To answer the main question, three sub-questions have been formulated:

- I. What is the effect of the hatchery on the EWL and hatchability?
- II. What is the effect of the breed on the EWL and the hatchability?
- III. What is the effect of the breeder flock age on the EWL and the hatchability?

2. Material and Methods

In order to answer the main question, research was carried out through the analysis of gathered data. These data are qualitative data regarding the hatcheries, layer breeder flock age and breeds, and quantitative data from weighing eggs and hatchability. As all the data that were collected are for all the sub-questions, the material and methods are presented by explaining the flock and hatchery profiles, the measurements that were done and the organization and use of the data to answer the sub-questions.

Farm profiles

The measurements were done on hatching eggs of 10 different parent stock farms with, in total 24 flocks. The age of the flocks varied from 28 to 68 weeks old. The flocks consisted of several breeds. Five flocks of Hy-Line Brown, three flocks of Hy-Line W36, five flocks of H&N Super Nick, eight flocks of H&N Brown Nick, one flock of H&N Nick Chick, one flock of Lohmann Brown Classic, and one of Lohmann LSL Classic.

Hatcheries and incubation profile

Table 1. Incubation program in Arendonk

The measurements were done in two different hatcheries. The first hatchery is located in Arendonk (Belgium) and is equipped with Petersime incubators and hatchers.

The incubation program was the regular program used for all the other eggs. The eggs have been put in the incubator and warmed up to 77°F for four hours. Then the machine started at day 0, hour 0. The program has been divided in 21 temperatures steps. The total incubation program lasted 21 days and 6 hours. Nevertheless, the chicks might have stayed few hours longer in the hatchers, as it is not possible to be exactly at the same moment in both hatcheries for the last measures.

Arendonk: incubation program		
Time (Day: hour)	Temperature (°F)	Relative humidity (%)
0:00	100,1	59,8
1:00	99,8	60,4
4:00	99,7	60,6
6:00	99,7	57,6
8:00	99,6	57,8
10:00	99,5	55
12:00	99,4	55,2
13:00	99,0	54
13:12	99,0	52
14:00	98,8	48,4
14:12	98,8	45,9
15:00	98,6	44,8
16:00	98,4	45,2
18:00	98,3	45,4
18:12	98,1	40,6
19:00	97,9	40,4
19:12	97,7	42
19:16	97,6	44,4
20:16	97,1	Off
20:18	96,9	Off
20:20	96,7	Off
20:22	96,5	Off
21:00	96,3	Off
21:02	96,1	Off

The second hatchery, located in Afferden (The Netherlands), is equipped with HatchTech incubators and hatchers and has a different incubation profile compared to the Petersime machinery in Arendonk. The temperatures of the incubation program are decreasing on 15 times.

Table 2. Incubation program in Afferden

Afferden: incubation program		
Time (Day: hour)	Temperature (°F)	Relativity humidity (%)
00:00	101	80-55
3:00	101	80-55
3:05	101	75-55
3:10	101	70-55
3:16	101	65-55
3:20	101	60-55
4:00	100,8	55-40
6:00	100,6	55-40
8:00	100,4	55-40
10:00	100,2	50-40
12:00	100,0	50-40
14:00	99,8	45-40
15:00	99,6	45-40
16:00	99,4	45-40
18:00	99,2-98,0	45-40
19:12	97,5	65-45
20:00	97,0	65-45
20:12	97,0	60-45
20:18	96,5	60-45
21:00	95,5	55-45
21:03	95,0	55-45

The methods present the variables that were measured and how the measurements were realized. For this research, 7 824 eggs were followed during 48 incubation settings (450 to 530 eggs per setting).

Tray weight measurements

The hatchery in Arendonk used setter trays of 150 eggs, the one in Afferden used setter trays of 88 eggs. So to get at least the same quantity of eggs, two trays were used in Afferden for one in Arendonk. 450 or 530 eggs were weighed from each flock in both hatcheries. To measure the egg weight loss 3 empty trays were first weighed and then filled with eggs, after which they were weighed again and the result were recorded. The eggs were weighed again at 17 or 18 days of incubation depending of the hatcheries schedule. All the weighing results were recorded in an Excel file. In case of a transfer and candling on the 17th days, a calculation was made to get the equivalent for the 18th days. The quantity of hatched eggs was recorded per tray.

Individual egg weight measurements

From the three trays measured for the global egg water loss, all the eggs of one (or two tray for Afferden) were individually weighed. The eggs were marked from 1 to 150 for the trays of 150 eggs and 1 to 176 for the two trays of 88. All the results were recorded in an Excel file.

The egg weight loss was measured by weighing all the eggs a second time at 17 or 18 days, depending of the hatcheries' schedule. A break out analysis was performed on the non-hatched eggs to identify the reason of non-hatching.

Data analysis

Data was categorized by hatchery. The flocks were divided by group of age; less than 40 weeks old, 40 to 50, 50 to 60 and older than 60 weeks old, and by breed. EWL was also divided by class. Every 2% for each class, starting from less than 6% to more than 16%.

First of all, average hatchability was compared between hatcheries and then results were specified per breed and per age class and again compared between hatcheries.

Then EWL (as dependent variable) were compared between the independent variables, which are hatchery, breed and breeder flock age.

Finally, distribution graph were made to show the distribution of eggs weighed and hatchability per EWL class for all independent variable.

3. Results

3.1 Data collection

Data has been collected for a general analysis to generate descriptive statistics regarding egg weight loss and hatchability in laying hen hatching eggs and to analyse the possible factors involved, more specifically hatchery, breed and breeder age. Three to six trays were weighed per henhouse for 24 flocks in total. As shown in table 3, the quantity of eggs weighed was depending on the hatchery. In the first hatchery (Arendonk) three trays of 150 eggs each were weighed. In the second hatchery (Afferden), six trays of 88 eggs each were weighed to get similar total quantity of eggs as Arendonk. All trays were individually weighed at set and at transfer and were counted at hatch.

Table 3. Distribution of the quantity of eggs weighed per flock and per hatchery and the number of flocks.

Breed	N flocks	Hatchery		
		Afferden (N eggs)	Arendonk (N eggs)	Total (N eggs)
H&N Brown Nick	8	4224	3591	7815
H&N Nick Chick	1	528	450	978
H&N Super Nick	5	2640	2247	4887
HyLine Brown	5	2640	2247	4887
Hyline W-36	3	1584	1350	2934
Lohmann W LSL	1	528	450	978
Lohmann Brown	1	528	450	978
Total	24	12672	10785	23457

Hatchability has been calculated with the results from the all the trays whereas the EWL has been calculated with the results from the individuals egg weight. By using the individual egg weight to analyse the EWL it is possible to know the exact distribution of weight loss per egg instead of the average per tray. It also means that the hatchability has been calculated on all the eggs whereas the EWL results have been calculated on one third of the total eggs.

The independent variables have been selected regarding the literature and the available data in the company. As stated earlier, eggs data were collected in the same way in both hatcheries. This way, hatchery results could be compared with each. During the research period, all the eggs were separated by flocks in order to associate their results to their age and breed for the analysis. The age classes have been made to categories of different ages to be able to get relevant statistical analysis.

3.2 What is the effect of hatchery on EWL and hatchability?

Table 4 shows the EWL and hatchability results in both hatcheries. Independent one way ANOVA showed a significant effect of hatchery on EWL ($p < 0.001$) and on the hatchability ($p = 0.025$). Arendonk has a higher EWL and hatchability than Afferden.

Table 4. Average EWL and hatchability results in both hatcheries

	Arendonk	Afferden	Significance (p-value)
Egg weight loss (%±sem)	12,07 ± 0,17	10,52 ± 0,09	<0,001
Hatchability (%±sem)	87,93 ± 0,58	83,53 ± 0,57	0,025

Figure 1 presents the distribution of the egg weighed per EWL class per hatchery. A difference on the distribution between the hatcheries can be seen. In Afferden, more than 74,2% of the eggs weighed had an EWL between 8% and 12% , whereas in Arendonk only 57.5% of the eggs weighed had an EWL between 8% and 12. The graph also shows that Arendonk has almost 40% of its eggs weighed in the categories higher than 12% EWL while in Afferden only 18% of its egg weighed had more than 12% EWL.

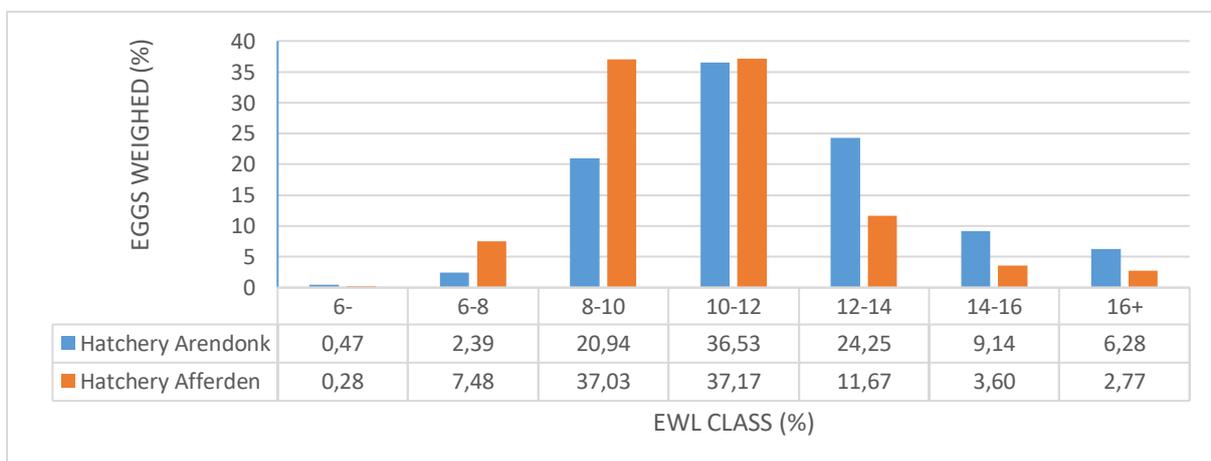


Figure 1: Distribution of the total eggs weighed in percentage per EWL class between hatcheries

Figure 2 shows the distribution of the hatchability results from the individual measurements per EWL class between hatcheries. ANOVA showed a significant effect of the EWL class on the hatchability ($F_{1,6000} = 10.956$; $p=0.003$) for this sample. Hatchability from the EWL class 8% to 16% was higher than 85% in both hatcheries. Hatchability is significantly lower in the EWL class 6-8% ($64.3 \pm 2.8\%$) than 8-10% ($87.3 \pm 0.6\%$, $F_{1,1000}=129.703$; $p=0.008$), it is also lower in the EWL class >16% ($66.1 \pm 1.6\%$) than 14-16% (87.9 ± 3.4 ; $F_{1,1000}=68.421$; $p=0.014$). For the groups 6-8% and >16% the hatchability is between 60% and 70% also in both hatcheries. For the EWL Class <6%, more than 70% of the eggs weighed hatched in Arendonk while only 50% hatched in Afferden.



Figure 2: Distribution hatchability in percentage per EWL class between hatcheries

3.3 What is the effect of breed on EWL and hatchability?

Table 5 shows that the F-statistic is significant ($p < 0.001$) for the average hatchability per breed. PostHoc testing showed that the hatchability of H&N Brown Nick ($81.9 \pm 0.7\%$) is significantly lower than Hyline W-36 ($93.0 \pm 0.6\%$; $p < 0.001$), H&N Super Nick ($85.9 \pm 0.8\%$; $p = 0.004$), H&N Nick Chick ($90.9 \pm 1.0\%$; $p < 0.001$). The lowest hatchability is 81.0% ($\pm 1.8\%$) for the Lohmann W LSL and is only significantly lower from Hyline W-36 ($93.0 \pm 0.6\%$; $p < 0.001$) and H&N Nick Chick ($90.9 \pm 1.0\%$; $p = 0.003$). There is no significant difference between Lohmann Brown ($86.9 \pm 1.4\%$) and all the other breeds.

Table 5 also shows a strong significant difference of hatchability for H&N Brown Nick between hatcheries (Arendonk: $86.0 \pm 0.8\%$; Afferden: $79.9 \pm 0.9\%$; $p < 0.001$). Hatchability of HyLine Brown between the hatcheries is also different (Arendonk: $87.4 \pm 1.2\%$; Afferden: $81.5 \pm 1.2\%$; $p = 0.019$). There is no other significant difference of result between the hatcheries.

Within the hatcheries, the results range between 79.8% and 94.8% hatchability for Arendonk and 79.9% and 92.1% hatchability for Afferden. In Arendonk, the highest hatchability is found in the breed HyLine W-36 ($94.8 \pm 0.8\%$) and is significantly higher than the hatchability of H&N Brown Nick ($86.0 \pm 0.8\%$; $p < 0.001$), H&N Super Nick ($88.4 \pm 1.0\%$; $p = 0.004$), HyLine Brown ($87.4 \pm 1.2\%$; $p < 0.001$) and Lohmann W LSL ($79.8 \pm 2.2\%$; $p < 0.001$). The breed with the highest hatchability in Afferden is also Hyline W-36 ($92.1 \pm 0.8\%$) and is significantly higher than Lohmann W LSL ($81.6 \pm 2.5\%$; $p = 0.002$) and all the other breeds with $p < 0.001$, except with H&N Nick Chick ($91.5 \pm 1.4\%$).

Table 5. Hatchability per breed, in average and per hatchery

Breed	Hatchability (% \pm sem)			p-value between hatcheries
	Average	Arendonk	Afferden	
H&N Brown Nick	81.9 ± 0.7^a	86.0 ± 0.8^{bc}	79.9 ± 0.9^a	<0.001
H&N Nick Chick	90.9 ± 1.0^b	90.0 ± 1.2^{acd}	91.5 ± 1.4^{bd}	1.000
H&N Super Nick	85.9 ± 0.8^{bc}	88.4 ± 1.0^{cd}	84.6 ± 1.0^{bc}	0.519
HyLine Brown	83.5 ± 1.0^{ac}	87.4 ± 1.2^c	81.5 ± 1.2^{ac}	0.019
HyLine W-36	93.0 ± 0.6^b	94.8 ± 0.8^a	92.1 ± 0.8^d	0.989
Lohmann W LSL	81.0 ± 1.8^{ac}	79.8 ± 2.2^b	81.6 ± 2.5^{ac}	0.995
Lohmann Brown	86.9 ± 1.4^{abc}	89.8 ± 1.9^{ac}	85.4 ± 1.5^{ab}	1.000
F-statistic	$F_{1,6000}=16.697$	$F_{1,6000}=8.247$	$F_{1,600}=13.786$	
p-value	<0,001	<0.001	<0.001	

(Superscript shared letters within a column means no significant difference)

Table 6 shows that the F-statistic is significant ($p < .001$) for the average EWL per breed. PostHoc testing using Tukey's correction revealed that H&N Brown Nick ($10.0 \pm 0.1\%$) had a lower EWL than H&N Super Nick ($11.5 \pm 0.2\%$; $p = 0.009$), HyLine Brown (11.4 ± 0.2 ; $p < 0.001$), Hyline W-36 (12.0 ± 0.2 ; $p < 0.001$) and Lohmann W LSL (11.2 ± 0.3 ; $p = 0.001$). All the other breeds showed no significant difference between them.

A comparison of EWL by breed within and between the hatcheries is also presented in table 4. There is a highly significant effect of the breeds in Arendonk ($p < 0.001$) and a low significance in Afferden ($p = 0.048$).

Also the PostHoc comparison showed that the EWL between each individual breed is not significantly different in Afferden ($p > 0.05$) while in Arendonk the EWL of H&N Brown Nick ($10.8 \pm 0.2\%$) is only similar to H&N Nick Chick ($12.7 \pm 0.8\%$). The EWL of Lohmann W LSL ($13.5 \pm 0.2\%$) is significantly higher than Hyline W-36 ($13.2 \pm 0.3\%$; $p = 0.013$) and all the other breeds with $p < 0.001$.

Table 6. EWL results per breed in average and per hatchery

Breed	EWL (% \pm sem)			p-value between hatcheries
	Average EWL	Arendonk	Afferden	
H&N Brown Nick	10.0 ± 0.1^a	10.8 ± 0.2^a	9.6 ± 0.1^a	0.178
H&N Nick Chick	11.0 ± 0.5^{ab}	12.7 ± 0.8^{ab}	10.2 ± 0.1^a	1.000
H&N Super Nick	11.5 ± 0.2^b	12.9 ± 0.4^c	10.7 ± 0.2^a	<0.001
HyLine Brown	11.4 ± 0.2^b	12.1 ± 0.3^{bc}	11.1 ± 0.2^a	0.438
HyLine W-36	12.0 ± 0.2^b	13.2 ± 0.3^c	11.4 ± 0.1^a	0.745
Lohmann W LSL	12.0 ± 0.4^b	13.5 ± 0.2^d	11.3 ± 0.2^a	0.518
Lohmann Brown	11.2 ± 0.3^{ab}	12.5 ± 0.2^{bc}	10.6 ± 0.2^a	1.000
F-statistic	$F_{1,6000}=7.608$	$F_{1,6000}=40.679$	$F_{1,600}=2.121$	
p-value	<0,001	<0.001	0.048	

(Superscript shared letters within a column means no significant difference)

Figure 4 present the distribution of the egg weighed in percentage per EWL class per breed. It shows that all breeds except H&N Brown Nick and Lohmann Brown have most of their eggs losing between 10% and 12% of their egg weight. In general EWL class <6%, 14-15% and >16% represent per class, less than 10% of the eggs weighed, except for the H&N Brown Nick where the EWL class 6-8% represent 11.5% of the eggs weighed for this breed and also expect for Lohmann W LSL where 10.7% of the eggs weighed lost between 14% and 16% of EWL.

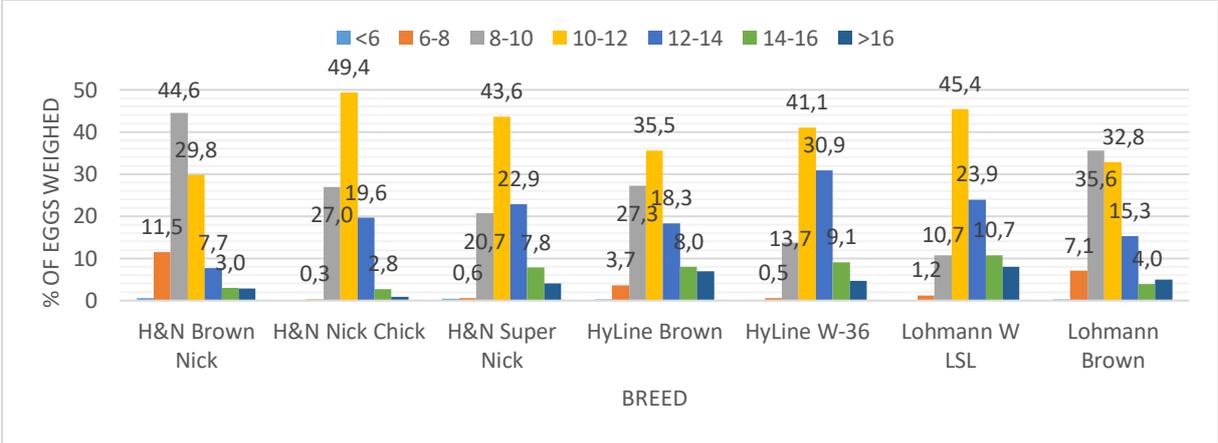


Figure 4: Distribution of the egg weighed in percentage per EWL class per breed

Figure 5 shows the distribution of hatchability in percentage per EWL Class per breed. Hatchability remain higher than 80% for all breed in the EWL class from 8% to 16%. For the same EWL class, hatchability can vary from 53.4% to 100% depending on the breed. Moreover, this wide variation occurs for the EWL class <6%, 6-8% and >16%. Except for the breeds H&N Nick Chick and HyLine W-36, there is a significant difference in average in hatchability between EWL class 6-8% ($65.4 \pm 6.6\%$) and 8-10% ($86.4 \pm 0.6\%$; $F_{1,1000}=50.379$; $p<0.001$). Also for the same breeds, hatchability is significantly lower for the EWL class >16% (62.0 ± 5.3) than the class 14-16% (88.3 ± 2.8 ; $F_{1,1000}=95.527$; $p<0.001$).

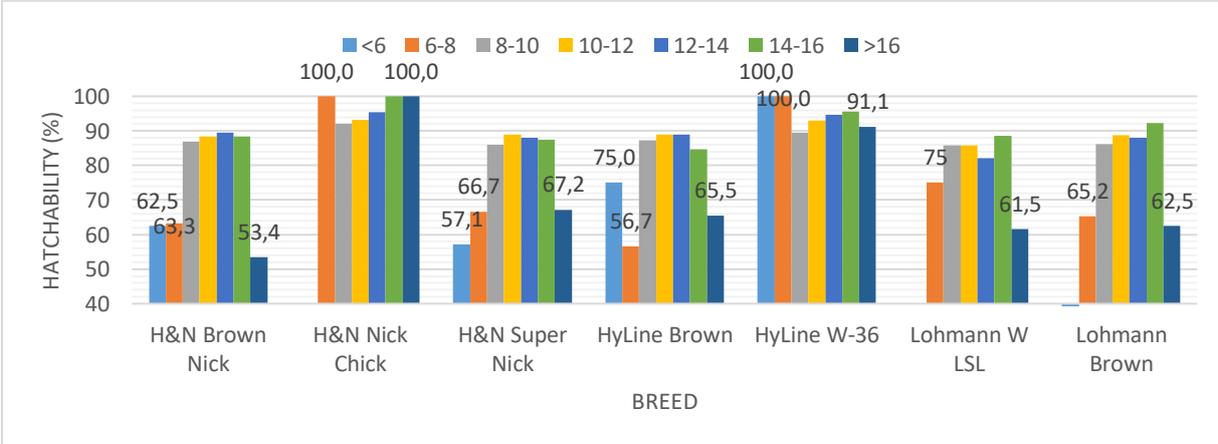


Figure 5: Distribution of hatchability in percentage per EWL Class per breed

3.3 What is the effect of breeder flock age on EWL and hatchability?

Table 7 presents the hatchability results per age category of the breeder flock in average and per hatchery. It shows that there is a significant effect of the age category on hatchability ($p < 0.001$).

The highest hatchability is found in the age class 40-50 weeks ($89.7 \pm 0.8\%$) and is significantly different from all the others age class; <40 weeks ($84.6 \pm 0.9\%$; $p = 0.002$), 50-60 weeks ($81.9 \pm 1.1\%$; $p < 0.001$) and >60 weeks ($84.1 \pm 0.7\%$; $p < 0.001$). The lowest hatchability is found in the age category 50-60 weeks old ($81.9 \pm 1.1\%$) but is significantly different only from the age class 40-50 ($89.7 \pm 0.8\%$; $p < 0.001$).

There is a clear effect of the age class on the hatchability in Afferden ($p < 0.001$) while the significance is lower in Arendonk ($p = 0.042$). In Afferden, the highest hatchability is from the age category 40-50 weeks ($89.2 \pm 1.1\%$) and is significantly different from all the other age categories ($p < 0.001$). In Arendonk, the highest hatchability is also from the 40-50 weeks category ($90.8 \pm 1.0\%$) and is only significantly different from the category >60 weeks ($86.7 \pm 1.0\%$; $p = 0.033$). PostHoc testing revealed that the results between hatcheries from hens younger than 50 weeks old are not significantly different while there is a significant difference between the hatcheries for the age groups 50-60 weeks (Arendonk: 87.0 ± 0.7 ; Afferden: 79.3 ± 1.4 ; $p = 0.005$) and >60 weeks (Arendonk: 86.7 ± 1.0 ; Afferden: 82.8 ± 0.8 ; $p = 0.038$).

Table 7. Hatchability per age class, in average and per hatchery

Age (weeks)	Hatchability (% \pm sem)			p-value between hatcheries
	Average	Arendonk	Afferden	
<40	84.6 ± 0.9^a	88.7 ± 1.2^{ab}	82.6 ± 0.9^a	0.062
40-50	89.7 ± 0.8^b	90.8 ± 1.0^b	89.2 ± 1.1^b	0.984
50-60	81.9 ± 1.1^a	87.0 ± 0.7^{ab}	79.3 ± 1.4^a	0.005
>60	84.1 ± 0.7^a	86.7 ± 1.0^a	82.8 ± 0.8^a	0.038
F-statistic	$F_{1,3000}=12.809$	$F_{1,3000}=2.882$	$F_{1,3000}=12.626$	
p-value	<0,001	0.042	<0.001	

(Superscript shared letters within a column means no significant difference)

Table 8 shows the average EWL results per age class. There is a strong significance on the EWL between age classes ($p < 0.001$). The EWL of the age class of <40 weeks ($9.6 \pm 0.2\%$) and 40-50 weeks (10.8 ± 0.2) are significantly lower than the EWL of the class >60 weeks ($11.8 \pm 0.1\%$; $p < 0.001$; $p = 0.005$). There is no significant difference between the EWL of the group younger than 40 weeks old (9.6 ± 0.2) and the groups 40-50 weeks (10.8 ± 0.2) and 50-60 weeks (10.7 ± 0.2).

Table 8 also shows the EWL results per age within and between hatcheries. There is a strong significant effect of age on EWL in Arendonk ($p < 0.001$) while the age had no significant effect on the EWL in Afferden ($p = 0.274$). For Arendonk, the PostHoc testing using Tukey's correction revealed that each age class is significantly different from each other with $p < 0.001$ except between the class 40-50 weeks ($12.1 \pm 0.3\%$) and 50-60 weeks ($11.6 \pm 0.3\%$) which have a significance of $p = 0.039$. The results in Afferden are from $9.3 \pm 0.2\%$ for the youngest to $11.2 \pm 0.2\%$ for the oldest chickens but no significant difference has been shown.

ANOVA PostHoc testing between hatcheries revealed that there is no significant difference on EWL between hatcheries for the age groups younger than 50 weeks old. In Arendonk EWL was higher than in Afferden for the groups 50-60 weeks (Arendonk: $11.6 \pm 0.3\%$; Afferden: $10.2 \pm 0.2\%$; $p = 0.032$) and >60 weeks (Arendonk: $12.8 \pm 0.2\%$; Afferden: $11.2 \pm 0.2\%$; $p < 0.001$).

Table 8. EWL results per age class in average and per hatchery

Age (weeks)	EWL (% \pm sem)			p-value between hatcheries
	Average	Arendonk	Afferden	
<40	9.6 ± 0.2^a	10.3 ± 0.1^a	9.3 ± 0.2	0.994
40-50	10.8 ± 0.2^{ab}	12.1 ± 0.3^b	10.2 ± 0.2	0.255
50-60	10.7 ± 0.2^{abc}	11.6 ± 0.3^c	10.2 ± 0.2	0.032
>60	11.8 ± 0.1^c	12.8 ± 0.2^d	11.2 ± 0.2	<0.001
F-statistic	$F_{1,3000}=12.239$	$F_{1,3000}=81.436$	$F_{1,3000}=1.295$	
p-value	<0.001	<0.001	0.274	

(Superscript shared letters within a column means no significant difference)

Figure 5 presents the percentage of eggs weighed per EWL class per age category. As it can be seen, almost 90% of the eggs weighed lost between 8% and 12% of humidity. For the category of 40-50 weeks old EWL class 10-12% stay above 41.8% whereas the class 8-10% decreased to 28.7% hatchability and the class 12-14% of EWL increased by 13.4% to reach 18.6% of egg weighed in this class. In the age category 50-60 weeks, EWL class 8-10% is the one with the most of eggs weighed, followed by EWL class 10-12% with 30.1% and EWL class 12-14% with 14% of the eggs weighed. Percentage of eggs weighed in the class higher than 14% increased

slowly from the category younger than 40 weeks to older than 60 weeks. Starting with 0.9% for the EWL class 14-16% and 0.5% for the class >16%, in the oldest group, the percentage of these classes represent respectively 9.1% and 7.3%

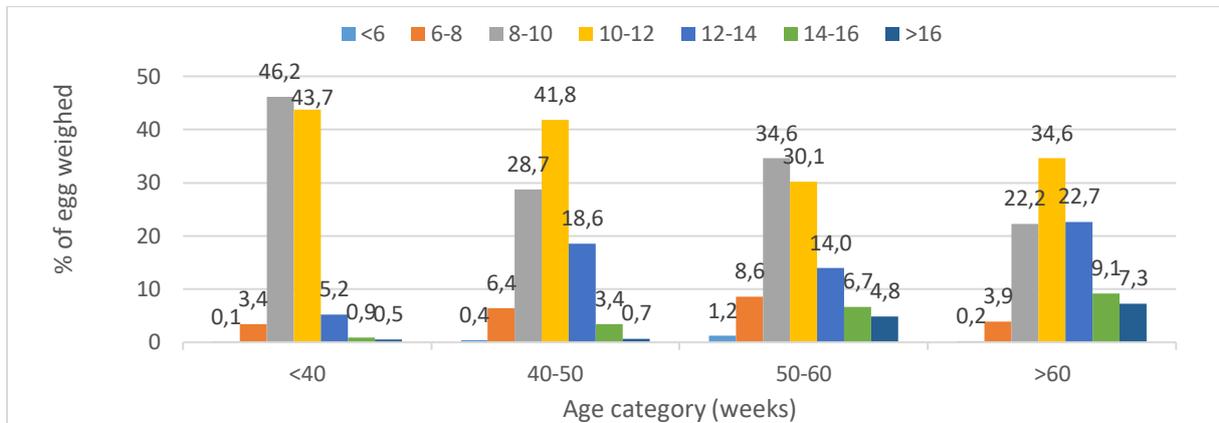


Figure 5: Distribution of the egg weighed in percentage per EWL class per age category

Figure 6 shows the distribution of hatchability per EWL Class for each age category. Hatchability looks remain higher than 82% in all age category for EWL between 8% and 16%. It is also and only higher than 80% for the EWL class >16% in the age category 40-50 weeks. On the other hand, hatchability from the EWL group 6-8% is zero for age group younger than 40 weeks and seems decrease between 40 weeks old and the group older than 60 weeks. In all cases, EWL class <6% has its hatchability lower than 70%.

For all categories of age, analysis of variance showed that hatchability is lower for the EWL Class >16% ($66.7 \pm 11.0\%$) than the for the EWL class 14-16% ($90.0 \pm 6.5\%$; $F_{1,1000}=13.287$; $p=0.011$). In addition, Anova test revealed that hatchability from the category 6-8% ($65.8 \pm 9.4\%$) EWL is lower than the one in 8-10% category ($87.4 \pm 2.5\%$) in average for all age groups ($F_{1,1000}=19.884$; $p=0.004$). This test did not show difference between EWL classes from 8% to 16% in all age categories.

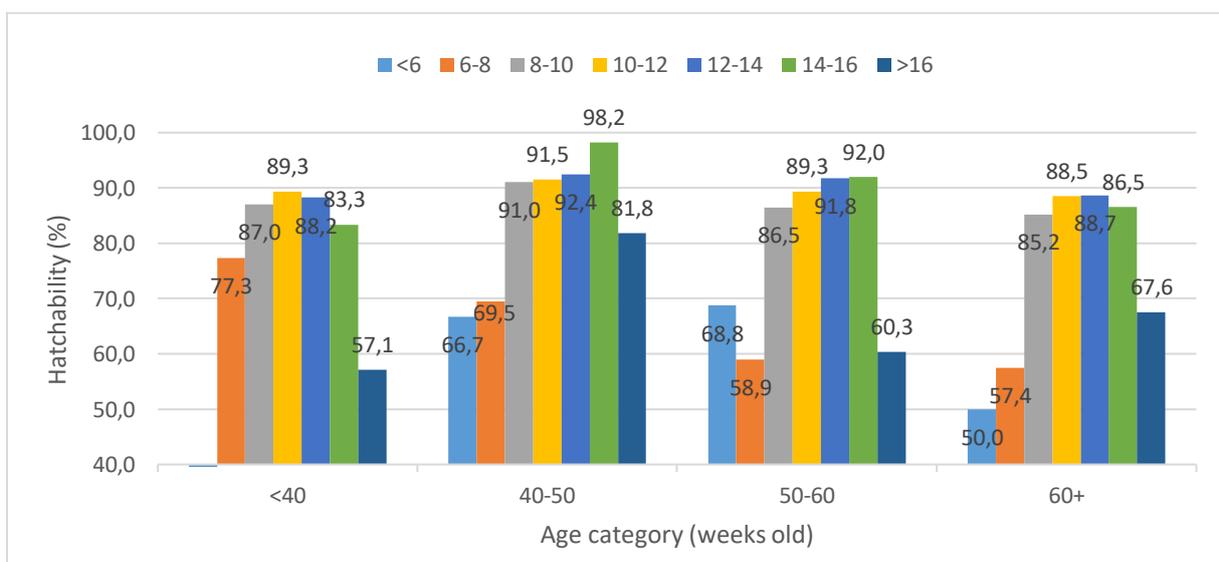


Figure 6: Distribution hatchability in percentage per EWL Class per age category

4. Discussion

The main aim of this study was to investigate factors that can influence egg weight loss and hatchability such as age, breed and hatchery in order to improve hatcheries' performance.

4.1 What is the effect of hatchery on EWL and hatchability?

There was a strong effect of hatchery on egg weight loss and hatchability. With a higher EWL, Arendonk had also a higher hatchability. In line with our results, Yassin et al. (2008) also concluded that the hatchability was significantly related with hatchery.

Figure 1 showed a difference in the distribution of the EWL between hatcheries. When Arendonk's distribution was mostly like a bell curve, Afferden showed two peaks with almost the same percentage. Buhr (1995) explained that the average that hatcheries try to reach is between 11% and 12%. Both hatcheries do not comply with this target, but Arendonk is closer with 12.07% of EWL in average.

Compared to the hatchability results, EWL does not affect the hatchability for the eggs that lost between 8% and 16% weight. Nevertheless, the analysis of variance revealed a significant effect of the EWL on hatchability on the overall average. Buhr (1995) also explained that embryonic mortality increases when humidity loss is lower than 9,1% or higher than 18,5%. In the case of our results, groups of weight loss have been made per 2%. Therefore, it was possible to see the results for EWL lower than 8% or higher than 10% but not to see the difference between 9% and 8% of EWL.

Nevertheless, there was a significant difference in hatchability (Figure 2) lower by more than 20% between the EWL categories 6-8% and 8-10% and between the EWL class 14-16% and >16%.

4.2 What is the effect of breed on EWL and hatchability?

The results showed an effect of breed on hatchability in average and per hatchery. Nevertheless, they also showed some difference per breed. Indeed, the lowest hatchability was 81% whereas the highest reached 93%.

These differences in the results might be caused by the effect of the different flock ages. Not all the age categories were represented in each breed, Lohmann W LSL and Lohmann Brown were both more than 60 weeks old while H&N Nick Chick was present only in the age class 40-50 weeks.

Ultimately, it is hard to determine a typical profile per breed. Indeed the figure 5 showed that H&N Nick Chick and HyLine W-36 had a high hatchability even in extreme EWL percentage. However, for all breeds hatchability was higher than 80% for the EWL class 8 to 16%, which is

to a certain degree conform to literature (Buhr, 1995) stating that mortality increases for EWL lower than 9.1% and higher than 18.5%. Here, except for H&N Nick Chick and HyLine W-36, hatchability is significantly lower in the EWL class below 8% and above 16%. As far as EWL class between 8% and 16% is concerned, hatchability is higher than 80% for all breeds.

Nevertheless, it is interesting to evaluate the difference in hatchability between hatcheries. The breeds H&N Brown Nick and HyLine Brown had more than 5% of difference in hatchability between hatcheries although they were produced and incubated the same day with the same protocol. In comparison with the EWL, the breed that had the lowest hatchability in Afferden, had also the lowest EWL. Nevertheless, none of the results had a significant difference between each other in Afferden.

Despite this last point, analysis of variance revealed a significant effect of breed on EWL in average and per hatchery (Table 6). Which is similar to the results from Alsobayel et al (2012) in broiler breeders who showed that breed significantly affected EWL percentage. On top of that, more recently Grochowska et al. (2019) concluded that in broilers, the genotype was the most important determinant of egg weight loss percentage. The distribution of the eggs weighed (Figure 4) for this research showed that for all breeds more than 55% of the eggs lost between 8 and 12% of weight and between 7.7% and 30.9% of eggs lost 12 to 14% of weight.

In Arendonk, the breed with the highest EWL is Lohmann W LSL with 13.5% and is significantly higher than all the other ones while it lost 11.3% of egg weight in Afferden. In contrast to Arendonk that have no results in the range of 11-12% of EWL preconized by Buhr (1995), Afferden has the breeds HyLine Brown, HyLine W-36 and Lohmann W LSL that lost between 11.1 and 11.4% of EWL. Yet, HyLine Brown and Lohmann W LSL reached only 81.5% and 81.6% of hatchability while HyLine W-36 reached 92.1% of hatchability.

However, results might not reflect the real egg weight loss percentage or hatchability for Lohmann W LSL and Lohmann Brown as there was only one flock from each and they were infected by *Mycoplasma Synoviae*. Lohmann W LSL was infected by *Mycoplasma Synoviae* strain causing eggshell apex abnormalities, and Feberwee and Landman (2009) found on broiler breeder a negative effect on eggshell strength by this strain suggesting that it could have an effect on hatching egg quality. Moreover, Lohmann W LSL has the lowest hatchability with 81% in average and 79.8% on Arendonk, which is significantly lower than almost all the other breeds.

4.3 What is the effect of breeder flock age on EWL and hatchability?

The analysis of variance showed an effect of age on hatchability in average and per hatchery. This is in line with King'ori (2011), who reviewed that the age of the flock is a hatchability influencing factor. In all our results (Table 5), the highest hatchability is from the age category 40-50 weeks. Compared with each other, Arendonk has significant higher results for the hens older than 50 weeks old.

Elibol and Brake found (2003) in broiler breeder that the efficiency of reproduction decreases with age due to many factors. Hatch results in Arendonk are most likely conform to the literature with a trend growing until 40-50 weeks old and then slowly decreasing. Also the hatchability distribution (Figure 6) showed that results from EWL class higher than 16% have a lower hatchability than the class 14-16%, as well as EWL class 6-8% is lower than 8-10% in all age category. But hatchability from 8% to 16% of EWL remains higher than 82% for all age groups.

The average EWL seems to increase with the age of the breeder flock (Table 8). Starting with 9.6% of EWL for the hens younger than 40 weeks old, there is no significant difference with the age category 40-50 weeks old and 50-60 weeks old. However, EWL is significantly higher in the >60 weeks group than the groups younger than 50 weeks old. These results confirm the findings of Zakaria et al. (2009) that due to a higher gas exchange between the air and eggs from old chickens, the egg weight loss increases with the age. Moreover, it also contradicts the results from Roque and Soares (1994) and Alsobayel et al. (2012) who noticed the opposite. It also has been seen (Figure 5) that for hens younger than 40 weeks, the distribution of EWL regroup almost 90% of the eggs between 8% and 12% while results are more spread between EWL class for the hens older than 60 weeks.

Strangely, analysis revealed no effect of age on EWL in the hatchery in Afferden while it revealed a clear effect in Arendonk (Table 8). Also in Arendonk the EWL is significantly higher in the >60 weeks group than all the other groups but it is not continually increasing from the youngest to the oldest. Indeed the age group 40-50 weeks loose more egg weight than the older group 50-60 weeks. In comparison, in Afferden the lowest results is not significantly different from the highest but it looks increase with the age with an EWL stabilized between 40 and 60 weeks old.

Regarding the process, age and breed results could have been improved by doing more measurements to get enough data to have relevant results on hatchability and EWL. Indeed, collect data on the complete production cycle of all flocks would have given a better trend of the effect of age on EWL and hatchability. As well as for the breeds, this would have given a comparable age average and so, comparable results.

5. Conclusions and recommendations

Many factors are involving hatchability and EWL during incubation. The loss in humidity is one of the few that the hatchery can manage.

The objective of this research was to investigate the influence of the breeder age flock, breed and hatchery on egg weight loss and hatchability. In order to provide information as a guide for hatcheries and possibly help them to adapt their incubation profile to breed or age. This research has been realized in two hatcheries with two different incubation profiles.

The first sub-question was to know the effect of the hatchery on the egg weight loss and hatchability. Literature were Analysis of variance revealed a clear effect of hatcheries on egg weight loss and hatchability, which is conform to the literature.

It also showed difference in the results, Afferden had a lower egg weight loss and hatchability than Arendonk. Individual egg weight results were not exactly conform to the literature. Indeed, the distribution of the eggs weighed was following a bell curve in Arendonk with one peak at 10-12% of egg weight loss while Afferden was presenting two peaks at the egg weight loss class 8-10% and 10-12%. Which give an average over the recommendation of 11% to 12% from the literature for Arendonk and below for Afferden. Nevertheless, hatchability was over 85% for the egg weight loss class from 8% to 16%.

The second sub-question was to know the effect of the breed on egg weight loss and hatchability. Results from this research showed an effect of breed on hatchability and on EWL in average and per hatchery.

Hatchability results were spread from 81% to 93% in average. Given that not all group of age were represented for each breed, it is not possible to conclude a global better hatchability for one breed. However, for this sample the best hatchability results are from HyLine W-36 and H&N Nick Chick that are both higher than 90% of hatchability in both hatchery. What is more, the distribution of hatchability per egg weight loss category showed that these two breeds had their hatchability higher than 80% whatever the egg weight loss class. It also proved that hatchability was higher than 80% for all breeds in the egg weight class range 8% to 16%. Which contradicts in part the literature by reducing the advised maximum egg weight loss range from 18.5% to 16%.

Egg weight loss results did not show significant difference between breeds in Afferden while there was a difference in average and in Arendonk. Highest egg weight loss were from Lohmann W LSL and HyLine W-36 in all results. Distribution showed in all breed that more than 55% of the eggs weighed were losing between 8% and 12% of weight but none of them were following a similar diminution profile.

The third and last sub-question was about the effect of age on egg weight loss and hatchability. Results showed an effect of age on hatchability in average and per hatchery and an effect of age on egg weight loss in average and in one hatchery.

Significant highest hatchability have been reached for the category of 40-50 weeks old in all results. In Arendonk, hatchability trend increased to 40-50 weeks old category and then slowly decreased with older hens while Afferden showed results different from expected with the lowest but non-significant result at the age category 50-60 weeks. However, previous research showed that efficiency in reproduction supposed to decrease with the age.

On the other hand, hatchability distribution per egg weight loss category showed a hatchability higher than 82% for all age categories between 8% and 16% of egg weight loss. And, same results as for the breeds and comparison of hatcheries, it showed a significant lower hatchability for the eggs that lost more than 16% of humidity and less than 8%.

Some literature were contradictory about the effect of age on egg weight loss. Here, in average, egg weight loss is significantly higher in the >60 weeks category than the groups younger than 50 weeks old. Also hatcheries showed different trends, Arendonk revealed a clear effect of age on humidity loss whereas no significant effect have been found in Afferden. Despite unexpected results in the evolution on egg weight loss with the age, in Arendonk and in average, loss in humidity is lower for hens younger than 40 weeks old than older than 60 weeks old. These results were following the distribution of egg weighed per egg weight loss class. In fact, 90% of the lost in egg weight was between 8% and 12% for the hens younger than 40 weeks whereas percentage of eggs weighed for the hens older than 60 weeks had a wider distribution.

All the results combined answers to the main question: How does breeds and breeder flock age influence the egg water loss and hatchability between hatcheries?

In short, breed and breeder flock age have a clear effect on egg weight loss and hatchability. Results per hatcheries showed that effect of influencing factors was depending of the hatchery. According to the literature and the data, hatchability trends to decrease after 50 weeks old while egg weight loss increase with the age. Nevertheless, for hatcheries, breed or different breeder flock age, hatchability was always higher than 80% for the weight loss category between 8% and 16%, and results from below and above this range was always lower. Therefore, egg weight loss also has an effect on hatchability.

As all independent variable have a good hatchability between 8% and 16% of egg weight loss, it is recommended to Afferden to make some change on its incubation program in order to get a higher loss in humidity in average and so have an egg weight loss distribution with a higher trend.

This research could be improved by doing the measurement on one complete production cycle. Starting with breeder flock younger than 30 weeks old until older than 60 weeks and by making groups of 10 weeks range. Parent stock flocks should be healthy and do not present permanent disease (for example: *Mycoplasma Synoviae*). Same number of measurements should be done for each age category per breed.

Also this study does not take into account the quality of day-old chicks which is known as being influenced by the egg weight loss. Recommendations for a range from 8% to 16% in humidity loss guaranty a good hatchability but not a good quality of day-old chicks.

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