

Investigating the technology of Short Period of Incubation During Storage (SPIDES) to mitigate damage caused by mechanical impact

*Timea Ágnes Torma**, and *Katalin Kovácsné Gaál*

Széchenyi István University, Albert Kázmér Mosonmagyaróvári Faculty, Department of Animal Sciences, 9200 Mosonmagyaróvár, Vár square 2., Hungary

Abstract. Transporting hatching eggs on plastic setter trays is common due to the reduction in packaging materials compared to paper trays, favouring sustainability. However, the plastic setter trays convey mechanical effects more strongly, negatively impacting eggshell integrity and blastoderm viability. On the other hand, Short Period of Incubation During Storage (SPIDES) proven to increase the liveability of the blastoderm, if applied at the right time. This experiment investigated the mitigating effects of SPIDES during modelled egg transport conditions on vibration machine. Data showed that moderate mechanical effects, simulated by a vibrating modelling machine, resulted in better hatchability of live embryos compared to setups where SPIDES was also applied.

1 Introduction

It is a well-known fact that shaking during transport has a negative effect on hatchability, but few studies have been aimed at establishing this more precisely [1, 2].

At the end of the 19th century, Dareste [1] conducted pioneering experiments on chicken embryos, using various noxious stimuli, physical impacts, and heat. His research concluded that the timing of these impacts was more critical than the type, as it determined the nature of the deformities in the embryos. Landauer [2] expanded on this by showing that allowing eggs to rest for 48 hours post-transport reduced the incidence of abnormal embryos compared to immediate incubation. De Lange [3] confirmed the significance of a resting period, although his study did not specify the proportion of abnormal embryos.

In our earlier series of experiments, [4, 5] a significantly increased level of abnormal embryos was determined when eggs were incubated less than 24 hours after modelled transport conditions on a vibrating platform. But the calculation from the data measured by the piezo sensor logger data was not solved. Until Gebresenbet [6] and Nazareno [7] introduced the calculation of the RMS (root mean square in different impact direction) and RSS (root sum of squares) to quantify and compare the impacts of different vibration conditions.

* Corresponding author: tormatimeagnes@gmail.com

Donofre et al [8] research highlighted, high levels of vibration (RSS, Root sum of Squares = 7.5 m/s^2) resulted in worse hatching results, longer hatch time and higher mid-term embryonic deaths compared to lower levels (RSS, Root Sum of Squares = 2.5 m/s^2).

While in our earlier works [4-5, 9] the hatchability decrease was mostly due to the significantly increased level of embryonic death in the early stage of embryonic development.

However, techniques like Short Period of Incubation During Storage (SPIDES) have shown [10] promise in reducing both early and late embryonic mortality, potentially counteracting the adverse effects of mechanical impact.

Proudfoot [11] stated, the daily moderate vibration of eggs, during storage, had no detrimental effect on hatchability and the dormant embryo was normally positioned on the yolk surface in the equatorial region of the egg and that the yolk turns so that the blastoderm lies uppermost and close to the shell in the large end up orientation. Practice shows, without regular turning in this position, blastoderm could stick to the shell membrane result in higher early dead embryos. Turning during storage decrease the level of early [12] and late embryonic mortality [10, 12].

Meanwhile in our other work [15] moderate, 10 Hz vibration resulted higher hatchability than the control group, due to the lower late dead embryo level.

Because we also considered the possibility to apply SPIDES on the egg truck during long transport, the other the aim of this trial to investigate the effect of modelled, moderate vibration and SPIDES applied directly next to each other.

The other objective of this trial is to observe the extent to which the blastoderm adheres to the shell membrane in the moderate shaking groups, and to determine if this method reduces the rate of early embryo mortality. The purpose of this investigation is to explore alternatives for egg storage when setter trolleys, which facilitate turning during storage, are unavailable. Moderate shaking using self-supporting setter trays or pallets equipped with an automatic shaking device could potentially serve as a substitute, aiming to maintain hatchability when traditional turning methods are not possible.

2 Materials and methods

2.1 Experimental design

Egg transport was modelled on a Crazy Fit Massager vibration machine (CFM machine). The machine is equipped with a vibration platform, moving in in two dimensions. The setting of the machine allows to vibrate the platform on 10, 20 or 30 Hz, ensuring consistent and controllable mechanical impacts.

The applicability of the CFM machine was evaluated by comparing its acceleration and deceleration measurements with those obtained under field transport conditions [9] and in previous trials [4-5, 9].

The experiment followed a completely randomized 3×2 factorial design (Table 1.), involving 12 repetitions of 150 eggs from Line A and 4 repetitions of 150 eggs from Line B per group. To minimize environmental and nutritional variations, eggs from both lines originated from the same farm and were handled identically until the SPIDES and vibration treatments.

Each setter tray (with the capacity of 150 eggs) served as observation unit.

Egg-breakout and hatchability data were collected for each tray separately. Eggs were placed on a Petersime self-supporting setter tray and exposed to 10 Hz vibration for 5 minutes on the CFM machine. HOB0 loggers monitored the mechanical impact during vibration treatment.

Eggs were stored for less than seven days in a climatized (16°C) egg storage room without turning. SPIDES treatment was applied according to established technology [13], with egg shell temperatures monitored using Tinytag-TK-4023 temperature loggers to ensure a consistent 32°C for 3 hours.

Table 1. Arrangement of the experimental groups.

	1.	2.	3.	4.	5.	6.
SPIDES	-	-	+	-	-	+
Vibration (CFM)	-	+	+	-	+	+
Line	A	A	A	B	B	B

2.2 Logger data and calculation

The mechanical impact from vibration was tracked using HOBO Pendant® G Data loggers [14], recording acceleration and deceleration in three directions (x, y, z) every second. The values, measured in m/s² or as gravitational acceleration equivalents ($g = 9.81 \text{ m/s}^2$), were used to calculate the Root Mean Square (RMS) and Root Sum of Squares (RSS) for all directions during vibration.

The value can be measured in m/s² or in terms of gravitational acceleration (g), which is 9.81 m/s². Logger data can be calculated using the following formulas [2, 15]:

$$\text{RMS}_j = \left(\frac{\sum_{t=1}^N a_j(t)^2}{N} \right)^{1/2}$$

where $a_j(t)$ is the observed instantaneous acceleration in the direction of axis j (x, y and z), and t is the time ($t=1, 2, \dots, N$); while N is the number of observations in the direction of the given axis;

The RSS (Root Sum of Squares) can then be calculated using the following:

$$\text{RSS} = (\text{RMS}_x^2 + \text{RMS}_y^2 + \text{RMS}_z^2)^{1/2}$$

where RSS is the sum of the RMS mean square values in all directions, calculated by taking the square root of the sum of the mean square values measured in all directions during the vibration (RMS; m/s²).

2.3 Egg-breakout and hatchability assessment

Live embryo proportions were determined by candling on the 10th day of incubation. The timing of embryonic deaths was assessed by egg-breakout analysis [16] of candled eggs and those left on the hatch tray post-hatching. The level of blastoderm or embryo stick to the shell membrane was also recorded.

Hatchability metrics included HOF% (hatched chicks as a percentage of fertile eggs) and HOV% (hatched chicks as a percentage of live embryos), along with early, middle, and late embryo deaths and abnormal or malpositioned embryos [16].

On the day of hatch, the number and quality of the chicks were determined.

2.4 Statistical analyses

MANOVA was employed to analyse the influence of two factors and their interactions on various hatchability traits, with a significance threshold of 5%. Normality of the data was evaluated using the Shapiro-Wilk test, excluding non-normally distributed categories from

further analysis. Statistical analyses were conducted using IBM SPSS Statistics (SPSS Inc., Chicago, IL, USA).

3 Results and discussions

Data calculated and measured by HOBO loggers for this moderate vibration: maximum acceleration of 14.22 m/s² in the x-axis direction, with an Root Mean Square in x direction of 9.5 m/s² and an Root Sum of Square of 10 m/s².

Eggs subjected to this moderate vibration (10 Hz for 5 minutes) showed the best hatchability of fertile (HOF, %) in the vibrated but non-SPIDES treated groups for both lines, though significance was above 0.05. No improvement in early dead levels was noted, and no embryos exhibited shell membrane sticking.

However, this moderate vibration on the CFM machine improved the hatchability of live embryos in both lines compared to the SPIDES-treated groups, though lines reacted differently., as shown on Figure 1-2.

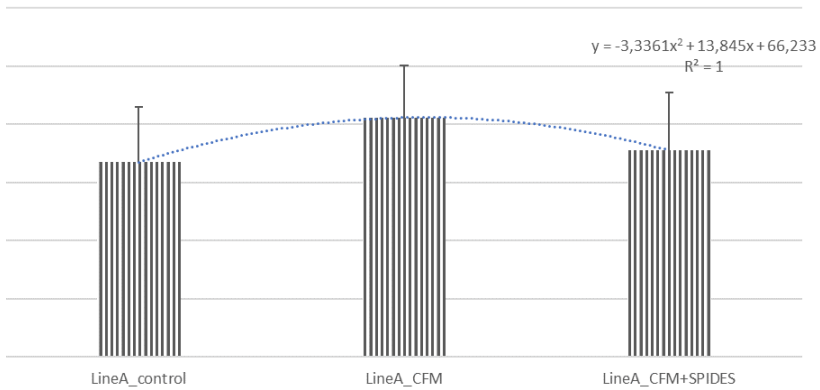


Fig. 1. Hatch of viable embryos in line A

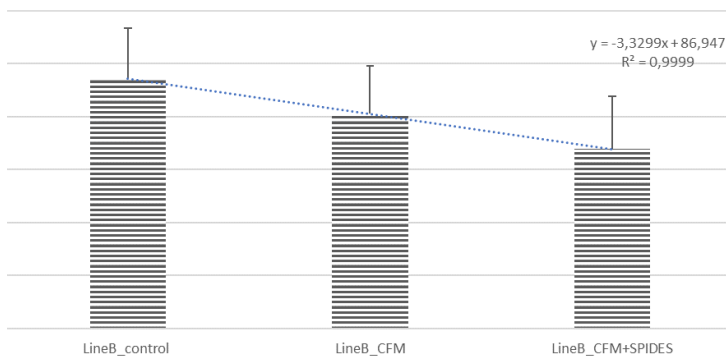


Fig.-2. Hatch of viable embryos in line B.

Vibration prior to incubation significantly affected hatchability (HOV%) and cull chick proportions (P=0.037 and 0.042), with marginal significance for abnormal embryos (P=0.054).

The hatchability result is in contradiction with Proudfoot [11] observation, although he used 0.8 mm amplitude for 15 minutes and 6 times higher frequency of vibration than our

settings with 12 mm for 5 minutes. Difference can be also due to the genetic difference in lines, since line in Proudfoot trials in 1969, show more resemblance to the slow growth birds in hatchability and durability of the germinal disc. That is also draw the attention to the different sensitivity of different genetic lines.

In Donofre et al [8] trial eggs from modern broiler line was used and 7,5 RSS (Root Sum of Squares) m/s^2 (180 min) impact caused lower hatchability due to the increased rate of embryos died on the middle phase in incubation.

In the current trial Vibration x SPIDES interaction gave a significant difference in the middle dead embryos ($P=0.029$) only in Line B and abnormal embryo level ($P=0.048$) in both lines.

This latest implies the similar observation when eggs are incubated directly after transport on the field and under experimental conditions when eggs were incubated less than 24 hours after mechanical impact resulted in lower hatchability and higher occurrence of malformed embryos [4-5].

SPIDES had a significant beneficial effect on the proportion of cull chicks ($P=0.045$) in both lines, despite of the short time between the treatment and the moderate mechanical impact.

This is in accordance with Dymond et al [15] and Maman et al [17] findings besides field observation.

Significant differences were noted for late dead embryos in Vibration x Line and SPIDES x Line interactions ($P=0.024$ and $P=0.031$), but not for the three-way interaction ($P=0.058$).

In Line B, the interaction between Vibration and SPIDES produced a significant result, but only for the most common malposition (Head over wing, $P = 0.024$). This still highlights the difference in sensitivity between the lines.

Part of the difference in hatchability due to the change in early embryonic mortality.

In our study early embryonic mortality showed 2 % decrease (Line A) in the 10 Hz vibrated and not SPIDES treated group, while in Line B increased but the difference is above the statistically significantly level in both cases.

4 Conclusions

We were unable to prove the beneficial impact of moderate shaking on early embryonic mortality. However, we did not observe a significant increase in this category when SPIDES and moderate vibration (Root Mean Square in x direction of $9.5 m/s^2$, Root Sum of Square of: $10 m/s^2$, 5 minutes) were applied consecutively.

The increase in hatchability in Line A due to moderate shaking was attributed to a decreased level of late dead embryos. Additionally, the interaction of different lines with SPIDES and vibration, even at this moderate level, underscores the need to investigate the sensitivity of different lines.

Based on the data, the moderate mechanical effect applied by the modelling machine resulted in better hatchability of viable embryos in both examined lines compared to the setup when SPIDES treatment was also applied. The lower hatchability when mechanical impact from vibration and SPIDES were applied closely together in time can be explained by the same symptoms observed when eggs are incubated immediately after transport to the hatchery without a resting period.

The fact that even a short (5 minutes), gentle (10 Hz) vibration can impact hatchability highlights the importance of transport and egg handling conditions, as well as the timing of SPIDES in relation to mechanical impacts.

References

1. C. Dareste, Research on the artificial production of monstrosities, or essays on experimental teratogenesis, Camille Dareste Published by C. Reinwald & cie in Paris (1877) <https://doi.org/10.5962/bhl.title.45945>
2. W. Landauer, L. Baumann, Rumplessness of chicken embryos produced by mechanical shaking of eggs prior to incubation. *J. Exp. Zool.* **93**: 51-74 (1943)
3. G. de Lange, Hatching egg transport IHP 10 vol. **22** no. 4. (2008)
4. T. Torma, K. Kovácsné Gaál, The influence of mechanical effects on the damages of broiler breeder hatching eggs and hatchability results different types of egg trays. *Animal Welfare, Etológia és Tartástechnológia*, **15**(2). pp. 64-72. ISSN 1786-8440 (2019)
5. Torma, T; Kovácsné, K. G. Effects of mechanical impacts on hatchability of broiler breeders MendelNet 2012: Proceedings of International Ph.D. Students Conference. ISBN:9788073756567 pp. 359-367 (2012)
6. G. Gebresenbet, S. Aradom, F.S. Bulitta, E. Hjerpe, Vibration levels and frequencies on vehicle and animals during transport. *Biosystem. Eng.*, **110**, 10–19. (2011) [10.1016/j.biosystemseng.2011.05.007](https://doi.org/10.1016/j.biosystemseng.2011.05.007)
7. A.C. Nazareno, A.C., I.J.O. Silva, A.M.C.Vieira, F.M.C. Vieira, K.O.S. Miranda, Transport of fertilized eggs: Influence of breeder age, storage period and road, *Rev. Bras. Eng. Agríc. Ambient* **18**(3) (2014) <https://doi.org/10.1590/S1415-43662014000300014>
8. A. C. Donofre, I. J. O Silva, A. C. Nazareno, I. E. Ferreira, Mechanical vibrations in the transport of hatching eggs and the losses caused in the hatch and quality of broiler chicks. *J Agric Eng* 48, No **1**(2017) <https://doi.org/10.4081/jae.2017.593>
9. T.Á. Torma and K.G. Kovácsné (2024) Induced and field mechanical effects on the hatchability of broiler breeder hatching eggs. *Euro. Poult. Sci.*, **88**. 2024, ISSN 1612-9199, © Verlag Eugen Ulmer, Stuttgart. DOI: 10.1399/eps.2024.397
10. S. Özlü, A. Ucar, T. Erkus, A. Nicholson, O. Elibol, 2021: Research Note: Effects of turning and short period of incubation during long-term egg storage on embryonic development and hatchability of eggs from young and old broiler grandparent flocks. *Poult. Sci.* **100**(4), 101026.
11. F.G. Proudfoot, Effect of packing orientation, daily positional change and vibration on the hatchability of chicken eggs stored small end up to four weeks *Can. J. Anim. Sci.*, **49** (1969), 29-35 (1969) <https://doi.org/10.4141/cjas69-005>
12. Elibol O, Peak SD, Brake J. Effect of flock age, length of egg storage, and frequency of turning during storage on hatchability of broiler hatching eggs. *Poult Sci.* 2002 Jul; **81**(7):945-50. doi: 10.1093/ps/81.7.945. PMID: 12162354.
13. D. Nicholson, N. French, S. Tullett, E. Lierde, G. Jun, Short Periods of Incubation During Egg Storage – SPIDES, *Lohman information* **48**(2):51. (2013)
14. HOBO® Pendant® G Data Logger (UA-004-64) White Paper 16920n
15. J. Dymond, Vinyard B, Nicholson AD, French NA, Bakst MR. Short periods of incubation during egg storage increase hatchability and chick quality in long-stored broiler eggs. *Poult Sci.* **92**(11):2977-87, 2013 Nov. doi: 10.3382/ps.2012-02816. PMID: 24135602.
16. S.G. Tullett, Investigating Hatchery Practice Ross Tech 98/35. Aviagen Ltd, Newbridge, Scotland (2009).

17. A.H. Maman and Yildirim I. The effect of short periods of incubation during egg storage (SPIDES) on internal egg quality, hatchability and chick quality of long stored old layer breeder eggs. *Euro. Poult. Sci.*, **86**. 2022, ISSN 1612-9199, © Verlag Eugen Ulmer, Stuttgart. DOI: 10.1399/eps.2022.353