Boris Habermann and Jens Unrath, Berlin, as well as Peggy Braun and Noelia Fernandez Castro, Leipzig

Disinfecting Hatching Eggs with Ozonate Media

For optimal hygiene management in animal husbandry facilities, various disinfectants and disinfection methods, which start inactivating germs on hatching eggs at differing points, are used. In a research project a technology was developed, which fumigates hatching eggs with ozone and effectively reduces Salmonella Enteritis (S.E.) on the eggshell. Environmentally relevant emissions cannot arise, since ozone is unstable and decomposes into oxygen within a short period of time.

Dipl.-Ing. (FH) Boris Habermann is head of the department of the Institut für Agrar- und Stadtökologische Projekte of Humboldt-University (IASP), Invalidenstraße 42, D-10115 Berlin.
Dr. agr. Jens Unrath is member of the scientific staff of Frankenförder Forschungsgesellschaft mbH (FFG), Potsdamer Str. 18 a, 14943 Luckenwalde, Wissenschaftsbereich Berlin, Chausseestraße 10, D-10115 Berlin;
e-mail: *info@frankenfoerder-fg.de*.
PD Dr. Peggy Braun and Noelia Fernandez Castro are scientists at the Institut für Lebensmittelhygiene, Veterinärmedizinische Fakultät of Leipzig University , An den Tierkliniken 1, D-04103 Leipzig.

Keywords

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In the year 2005 the salmonellosis of man was the second most frequented infectious intestine disease conveyed to Robert Koch-Institute and still domestic poultry and eggs are considered to be the most important reservoir for salmonellae with regard to human nutrition. From great importance are the zoonotic salmonellae serovare, which frequently cause non- perceptible infections in poultry [1].

Particularly the intensively managed livestock with floor management are loaded with salmonellae. In order to displace this pathogen from the food chain it is necessary to concentrate on the most important entry source, the laying hen population [2, 3]. A first step takes place by vaccinating and control measures which are stipulated for breeding stocks. However, immunisations do not represent any guarantee for smaller salmonellae loads [4]. From the view of food-hygiene, the disinfection of eggs represents a problem. The ban of irradiation for hen's eggs may possibly result in antibiotics-dipping or scrubbing-procedures, in order to reduce the shell contamination. Both are not permitted for eggs used for human nutrition. An important measure for the control of salmonellas is therefore represented by the hatching egg disinfection. Today the most frequently practised method of the hatching egg disinfection is the fumigation with formaldehyde.

Due to the carcinogenic effect of formaldehyde a possible ban can be expected in the future, so that up to then an alternative method must be available.

Therefore, from environmental and occupational safety aspects it was necessary to develop a practice-suited alternative procedure with which the parameters germ reductions as well as process engineering are scientifically examined, the safety of application is given and the most possible disinfection effect is proven.

Problem definition

In the context of a research project a technical procedure solution should be developed by means of ozonation, in order to reduce salmonella enteritidis on hatching eggs. The following goals should be realised:

- Proof of the effectiveness of the ozone fumigation regarding a hundred per cent reduction of salmonellae.
- Proof of the safety of the procedure on the developing embryos as well as the conservation of the hatchability.
- Proof of the safety for the animals from fumigated eggs in the entire life phase.
- Development of a practice-suited fumigation procedure.

Performance of the task

In the first step an experimental set-up on a laboratory scale was prepared and converted. With this experimental set-up, which was flexibly modified for some analyses, individual tests could be accomplished over the entire period of the project (*Table 1*). During each test the shells of up to 50 eggs were artificially contaminated with different doses

Table1: Used measuring instruments and materials for the experiments

Term	Specification	Company
technical oxygen	20 l Gasflasche mit medizinischem Sauerstoff 200 bar	Messer-Grießheim
ozone measuring instrument	0,00-5,00 % wt / wt	BMT Messtechnik
ozone generator	4 g/h, BMT 802X	BMT Messtechnik
ozone generator	8 g/h, BMT 803	BMT Messtechnik
instrument for temp. and humidity	0-100 % r.F., -10…+80 °C Genauigkeit ± 1,5 % r.F., ± 0,3 °C	HygroPalm
instrument for temp. and humidity	ozonresistent und mit Digitalausgang	testo
compressor	Modell 50/15	ABN
catalyst	Ozon-Katalysator aus Mangan	Carulite 200

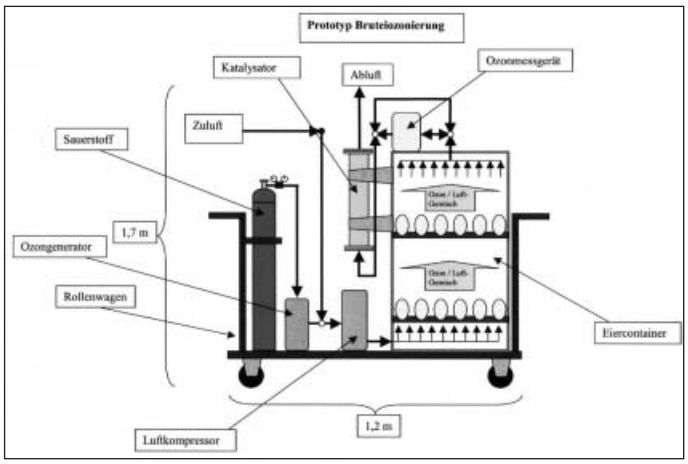

Fig.1: Sketch of the prototype

Fig.2: Inside view of the prototype

of S.E, (10^2-10^4, 10^5-10^6 CFU/shell), fumigated and afterwards micro-biologically and chemically examined together with control eggs. Based on these results a prototype with a capacity of approximately 500 hatching eggs was developed afterwards. After determination of the optimal ozone dose five tests were performed in total. All possible effects of the ozone treatment like the treatment day the hatching and the raising up to the end of the laying period on the hens were observed. In addition to that the following data were collected:

- pathological examinations at the end of the lifetime
- stable climate, clinical examinations, losses
- gain of weight, feed utilisation and laying performance

An inside view of the developed prototype is shown in *Figure 2*. The first project investigations on laboratory conditions showed that a reduction of salmonella on hatching eggs is possible by means of ozone gas. It could be even shown here that a 100 % reduction on the eggs can be achieved (*Table 2*), which other investigations concerning alternative procedures described in literature did not evince. A damage of the egg yolk or egg albumen (e.g. vitamin A) could be ascertained only with very high ozone concentrations, which are not relevant in practical use (starting from 2.5 % by weight). Fumigation parameters were found; with those on the one hand an optimal reduction of salmonella is attained and on the other hand the egg yolk or egg albumen is not damaged (1 to 2 hours impact time, 0.7-1.0 % ozone gas). Analyses showed no effects on the chemical composition or the embryonic history in this range of ozonation. In order to confirm the results beyond that point, ozonated eggs and eggs of comparative groups were hatched and raised under various husbandry conditions and examined up to the end of the laying period. Here it could be recognised that there was no measurable negative influence on the animals by the specific ozonation. After a test and adjustment phase the practical suitability of the prototype could also be confirmed. With the fumigation of the contaminated eggs the salmonellae could be inactivated to a not detectable degree in all five tests (*Table 2*). This corresponded to a reduction of up to 3.79 log10 after treatment with 0.7 % ozone and 120 minutes impact time. The accumulation showed that only sporadically isolated germs could be re-activated again (in two of five tests). Embryological damages or changes of the egg yolk or egg albumen could not be proven. The hatching of the eggs and the raising did not result in significant differences to the comparative groups.

Application recommendation

The investigations showed that the use of this technology in hatcheries is possible. In addition to that, the ozonation is generally classified to be more favourable than the use of formaldehyde regarding to the effect on humans and environment. In comparison with other treatments (e.g. egg washing and UV irradiation) the ozone fumigation performs better, since this procedure has lower costs than the egg washing, and/or works more effectively than the UV irradiation. For each user this procedure then has to be examined and/or adapted specifically. Nevertheless, this procedure is recommended for each hatchery. A combination of several measures for containing the salmonellae is recommended.

1. an optimal hygiene management
2. an immunization by vaccination campaigns
3. a disinfection of the hatching eggs

All three measures must be always carried out, whereby it is necessary to begin with the disinfection of the hatching eggs as early as possible. A rapid treatment of the hatching eggs after laying ensures that pathogens cannot be transferred and increased. The project investigations also showed that the disinfection with ozone is much more effective with small contamination degrees.

Table 2: Salmonella reduction (contaminatione dose 10^2 to 10^3 S.E. on the egg shell) after ozonation

Experim.	Ozone con. (% wt/wt)	EWZ (min)	Number of eggs	Amount of SE. Controlgroup (log_{10})[1] ± SE	Amount of SE a. ozonation (log_{10})[1] ± SE	Reduction (log_{10})*	neg. after enrichment (%)[2]
1	0,7	120	40	3,44 ± 0,00	0,00 ± 0,00	3,44	55
2	0,7	120	40	2,15 ± 0,03	0,00 ± 0,00	2,15	100
3	0,7	120	40	2,16 ± 0,04	0,00 ± 0,00	2,16	100
4	0,7	120	40	2,31 ± 0,04	0,00 ± 0,00	2,31	70
5	0,7	120	10	3,79 ± 0,06	0,00 ± 0,00	3,79	100

* sign. reduction ($p < 0,01$) in all experiments
[1] number per shell [2] eggs in %, being negativ e after enrichment

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