Thermal Disinfection of Poultry Grow-Out Facilities in Central-Northern Italy

Peruzzi A., Fontanelli M., Sorelli F., Frasconi C., Raffaelli M.
University of Pisa. MAMA – DAGA.
Via San Michele degli Scalzi, 2, 56124, Pisa, Italy
Tel 0039 0502218942, Fax 0039 0502218966 aperuzzi@agr.unipi.it

Abstract
Chemical treatments are commonly adopted for poultry houses sanitation. In fact, an ordinary floor disinfection is needed to achieve the depletion of pathogenic population (i.e. some species of bacteria and fungi) and reduce the risk of meat contamination. Recently, the increasing of popular attention to the consumers and operators health, and to the quality of food, brought the farmers to consider environmental friendly alternative methods.
A specific two-year experimental trial (2005-2007) was carried out in the Central-Northern part of Italy in order to set up new machines and strategies for floor disinfection of poultry houses by means of an open flame. Trials were run in the machine shop of the University of Pisa (indoor, under controlled conditions) and in two different private farms (a broiler house and an outdoor pheasant farm).
The first experiment consisted in a series of test bench trials, carried out in order to test the efficacy and the adjustment of LPG fed open flame burner on pre-inoculated steel plates. During the second experiment, the ordinary chemical disinfection strategy was compared, in the broiler house, to the innovative technique, carried out by means of an on purpose made 1.5 m wide mounted flaming machine. The last experiment consisted in the evaluation of the efficacy and operative performances of a 2 m wide flaming machine prototype on purpose built for open air pheasant farms.
The obtained results are very promising as, thermal disinfection strategy seems to be very effective on floor pathogens and cheaper than the ordinary chemical one.

Keywords: poultry house sanitation, flame disinfection, flaming machine

Introduction
Reducing bacterial and fungal populations is a major issue in poultry houses (Payne *et al*., 2002 and 2005).The presence of a high population of pathogenic bacteria in broiler grow-out houses can contribute in declining the wellness of the flock and lead to a sensitive production loss (Payne *et al*., 2002 and 2005).
Moreover there are further problems concerning industrial processing, food quality and consumers health as carcasses could be seriously contaminated by dangerous micro organisms like *Salmonella* and *Campylobacter* (Mead & Scott, 1994; Payne *et al.*, 2002 and 2005).
For these reasons, a major issue of poultry industry is to prevent product contamination through bacterial population reduction programs during the growing phase of animals (Payne *et al*., 2002 and 2005).
In this respect, specific researches have been carried out in Europe and United States aiming to find effective solutions to decrease dangerous micro organisms presence or significantly low their growth rate (Payne *et al.*, 2002 and 2005; Gradel *et al.*, 2004 and 2005).
In this regard, recent studies showed that considerable reductions of bacterial population can be achieved by removing the old litter followed by cleaning and disinfecting of facilities. It

usually occurs between the end of a growing cycle and the beginning of the following one, as broiler houses generally are not cleaned during the growing cycle. This disinfection treatment is generally performed by means of specific chemical sanitizers (Marin *et al.*, 2009). Moreover there is a common concern in Europe about food health and the application of potentially dangerous chemical products.

The aim of this research is to develop effective and environmental-friendly solutions in order to eliminate or reduce dangerous bacterial population in the period between flocks in broiler houses (indoor-reared broilers) and open air pheasant houses.

An on purpose made flaming machine was build and set up in order to carried out specific trials and evaluate the possibility to substitute chemical disinfectants with thermal treatments.

Materials and methods

<u>*Controlled conditions experiment*</u>

The first experiment was carried out in the laboratory of agricultural machinery and farm mechanization of Dipartimento di Agronomia e Gestione dell'Agroecosistema (DAGA) of the University of Pisa (Central Italy).

A specific test bench was used in order to evaluate the effectiveness of an on purpose made open flame burner for house floor thermal disinfection. This bench was used in the past to evaluate the effect of different adjustments of specific burners for flame weeding.

Artificially pre-inoculated steel plates were treated with an open flame cylinder shaped burner, the same one used for on-farm trials (fig. 1).

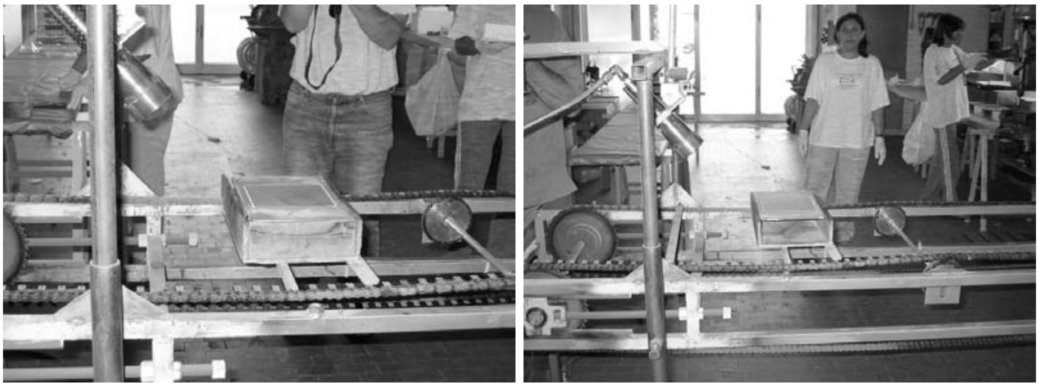


Figure 1. Flaming treatment of a pre-inoculated steel plate on test bench.

Three thermal treatments were compared, as generated by different combinations of LPG pressure and working speed: 0.12 MPa and 1 km h^{-1}; 0.12 MPa and 3 km h^{-1}; 0.16 MPa and 3 km h^{-1}. LPG consumption per hour and flame temperature were assessed.

Flame temperature was registered by means of a specific "R" type bifilar thermocouple (range 600-1700 °C). It was made by a platinum-rhodium alloy. The diameter of each filament was equal to 0.5 mm. LPG tanks were weighed before and after 15 minutes of continuous work in order to evaluate LPG consumption. Replication was 6-fold.

Each plate (containing an inoculated marked surface of 12 × 12 cm) was identified, sterilized and packed with a paper sheet. *Escherichia coli* AT 25922 and *Lactobacillus plantarum* ATCC 8014 were used as contaminating bacteria. Bacterial suspension was prepared with horse serum and a concentration of 10^9 cfu/mL.

Bacterial suspensions were successively titrated through decimal progressive dilutions, inoculating, by means of micro method (20 μL per dilution), blood agar for *E. coli* numeration

(24 h of growth at 37 °C) and MRS agar for *L. plantarum* numeration (48 h of growth at 37 °C - microaerophilic, 10% CO_2). After removing the package, each plate was inoculated with 100 μL of *E. coli* and 100 μL of *L. plantarum* suspension. Two drops were placed within the marked surface and uniformly distributed with a "L shaped" sterile spatula. Treatments were performed just after leaving plates drying out. Replication was 6-fold. Six plates were inoculated and used as untreated control. Bacteria were recovered after treatment in order to evaluate its effectiveness. Plates recover was carried out just by one operator in order to make this process as homogeneous as possible. Test tubes containing bacterial suspension were maintained at refrigeration temperature before laboratory analysis. Surviving microorganisms numeration was carried out following the same procedures described above. Moreover, *E. coli* numeration was carried out on both on blood agar and Violet Red Bile agar.

On-farm trials

Trials were carried out in two different farms: "Pratomagno" for house reared broilers and "La Viola" for open air reared pheasants.

Experiments in broilers house were carried out in September 2006 in two different barns of 1000 m^2 each. Ordinary chemical disinfection technique was compared to the innovative flaming disinfection.

Ordinary technique consisted in litter removal, equipments removal, equipments and ceiling washing, floor washing by means of water jet and/or submersion plus detergents based on sodium and potassium, washing water draining and dry chemical disinfection and finally litter re-establishment.

Innovative-low environmental impact method was characterized by the use of an on purpose made prototype equipped with LPG fed open flame burners with and a rear hood. Thermal treatments were performed as an alternative to floor washing and dry disinfection. Operative (working width, working speed, actual and operative working time, operative efficiency and fuel and LPG consumptions) and economic parameters were assessed and estimated referring to 1000 m^2.

"Biological" effectiveness of the two techniques was evaluated by means of steel plates placed on the floor after treatments. Plates were the same described above. After flock removal plates were recovered and analyzed. Plates recover was carried out just by one operator in order to make this process as homogeneous as possible. Microorganism numeration was carried out on mesophilic aerobic bacteria (Plate Count Agar, 48 hours at 37 °C), Enterobacteriaceae (Violet Red Bile Glucose agar, 24 hours at 37 °C), anaerobic sulphite reducers clostridia (TSC agar, 48 hours at 37 °C). On each plate (on a 30 cm^2 surface), *Salmonella* spp. was detected.

Trials carried out on pheasant open-air aviary were performed in April 2007 aiming to assess operative performances and biological effectiveness of an innovative on purpose made flaming machine.

In this case, ordinary strategy was already characterized by the use of thermal treatments. It consisted in dry *sorgum-chenopodium* meadow mulching (sowed for hosting game), thermal treatment, fertilization and deep moldboard plowing followed by secondary tillage. Thus this strategy was compared to a conventional one characterized by all the above described passes but thermal intervention. Aviaries dimension was ranging from 3000 up to 10000 m^2. Operative and biological assessments were carried out as described for broiler house.

Statistical analysis

Concerning bacterial data, Kruskall-Wallis and multiple rank comparisons tests were utilized for under controlled conditions experiment. On-farm data were processed with median test. Analysis were performed with R software (version 2..5.1).

Operative and economic data were not statistically processed.

Description of innovative machines for flame disinfection of poultry houses

Two different semi-mounted operative machines were tested. They were built in cooperation with Officine Mingozzi (Ferrara, Northen Italy).

Figure 2. Flaming treatment in broiler house.

They were similar concerning operative principle, but working width and the numbers of burners and LPG tanks were different.

The first prototype, PTE1600, was on purpose made for indoor treatments in broiler houses (fig. 2).The machine is small sized (1.60 m wide, 1.45 m long and 280 kg in weigh) and equipped with just one common commercial LPG tank 25 kg in weigh. These features allow this machine to be coupled with low power tractors (10-12 kW) and work on the upper floor of broiler houses without any problem of exceeding weigh and manoeuvrability. This operative machine is equipped with 20 cylinder shaped open flame burners (16 fixed under the hood and 4 placed on the side of the machine in order to treat the base of walls) and a manual lance connected with a 10 m long pipe (fig. 2). Moreover the machine is also equipped with an heat exchange system (that utilizes part of LPG contained in the tank) and an electronic panel placed on the hood of the tractor, reporting any anomalies in functioning, as burners turning off, pressure decrease.

The other prototype, PTR2000 (fig. 3), was developed to work outdoor on the soil, in pheasant aviary, thus it was characterized by a larger size. The machine was 2 m wide, 2 m long, 900 kg in weigh and it was equipped with two LPG tanks (25 kg in weigh each). In this case, the operative machine was coupled with a 55 kW 4WD tractor. All the other features are similar to those described for PTE1600.

Figure 3. Flaming treatment in pheasant aviary.

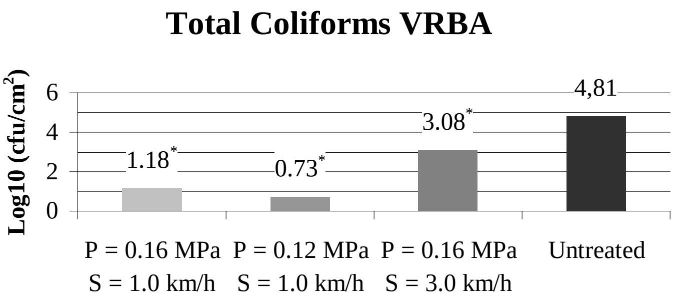
Results

<u>*Controlled conditions experiment*</u>

In table 1 it is shown that temperature of flame measured at the working level was the same for the two pressures tested but LPG consumption per hour was 20% higher for the higher pressure.

Table 1. Flame temperature and LPG consumption per hour of burner.

Pressure	Temperature	Consumption (kg h^{-1})
0.12	1220	2.5
0.16	1220	3.0



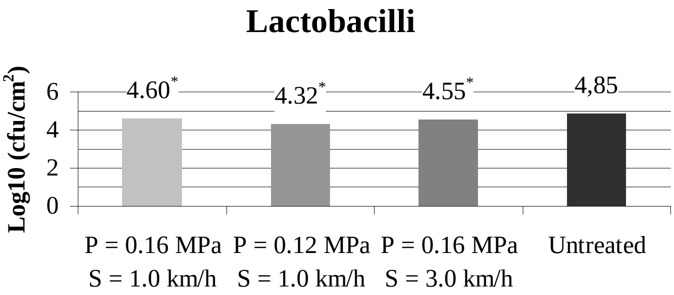


Figure 4. Microbial response to flaming treatments. Star means (*) significant differences *vs* untreated control.

In Figure 4 the microbiological results are shown. Recovery of Coliforms was considerably lower for treated plates, as flaming achieved relevant reductions of bacterial population. This difference was significant for Kruskall-Wallis test ($p<0.01$) and also for multiple comparison test.

Lactobacilli, as expected, showed an higher recovery, as it is known their superior tolerance to heat. However, statistical tests showed again a significant decrease in bacterial population. Furthermore, lower pressure treatment achieved better results with respect to higher pressure intervention.

On-farm trials

Operative performances for indoor treatments are shown in table 2.

Working speed ranged from 1.2 up to 1.5 km h^{-1}, and LPG pressure was 0.12 MPa as resulted the best adjustment during the previous series of trials. This technique required 30 minutes and 15 kg of LPG to treat 1000 m^2. Operative efficiency was equal to 85%. Fuel consumption was really very low (about 0.3 kg/1000 m^2).

Table 2. Operative performances of innovative flaming machine registered during on-farm trials carried out in broiler houses.

Operative performances		Results
Engine power	kW	12
Working width	m	1.5
Working speed	km/h	1.2-1.5
Actual working time	h/1000 m^2	0.45-0.54
Operative working time	h/1000 m^2	0.54-0.63
Operative efficiency	%	83-86
Working capacity	m^2/h	1590-1850
Fuel consumption	kg/1000 m^2	0.27-0.31
LPG pressure	MPa	0.12
LPG consumption	kg/1000 m^2	14.5-16.9

Operative cost evaluation for thermal treatment was about 30 €/1000 m^2. This value is very encouraging as estimated costs for ordinary chemical disinfection is over 200 €/1000 m^2 just for labour salary (two persons for the entire working day). Moreover thermal treatments do not require floor washing water and disposal.
Operative performances for indoor treatments are shown in table 3.

Table 3. Operative performances of innovative flaming machine registered during on-farm trials carried out in pheasant aviary.

Operative performances		Results
Engine power	kW	55
Working width	m	2.0
Working speed	km/h	1.5
Actual working time	h/ha	3.33
Operative working time	h/ha	3.84
Operative efficiency	%	86
Working capacity	ha/h	0.26
Fuel consumption	kg/ha	10.7
LPG pressure	MPa	0.12
LPG consumption	kg/ha	166.5

Tractor power, in this case, was 55 kW and working width was 2 m. LPG consumption was 166 kg ha^{-1} while fuel consumption was about 10 kg ha^{-1}. Thus, these values were definitively similar to the ones observed in broiler house.

Table 4. Biological results of on-farm trials.

Farm	Assessment	Flaming (cfu/cm^2)			Ordinary (cfu/cm^2)		
		min	median	max	min	mediana	max
broiler (1)	mesophilic aerobic bacteria	1,050,000	1,912,500	33,000,000	23,250	1,672,500	20,250,000
	Enterobacteriaceae	<150	125	375	<150	<150	225
broiler (2)	mesophilic aerobic bacteria	65,000	450,000	22,000,000	975	61,250	12,500,000
	anaerobic sulphite reducers clostridia	<50	<50	3,000	<50	<50	225
Pheasant aviary	mesophilic aerobic bacteria	1,150	22,500	107,500	17,500	65,000	170,000
	Enterobatteriaceefila	20	166	775	98	500	950

In table 4 bacterial populations results for on-farm trials are shown.
Concerning broiler houses, no significant differences were found between ordinary chemical disinfection strategy and innovative thermal technique. Thus the two methods appeared equivalent from a biological point of view.
Concerning open air pheasant aviary trials, median test showed significant differences, thus the use of flaming allowed a relevant decrease of bacterial population.

Conclusion
The results of this research showed that flaming disinfection of poultry grow-out facilities can be seriously taken into account as an alternative to chemical disinfection.
Moreover it is an environmental friendly technique and does not require floor washing and water disposal.
Under controlled conditions trials gave precious advice about burners adjustment and effectiveness, in order to set up on-farm trials properly.
The results of the on-farm trials showed that flaming can reach a very good bacterial control level, comparable to chemical dry treatment in broiler houses. Moreover flaming allowed to reach a significant reduction of micro organisms population when applied in open air aviary for pheasants grow-out.
Innovative equipments allow to carry out effective treatments with good operative performances in difficult contexts. Furthermore, for broiler houses, flaming estimated costs were relevantly lower than ordinary chemical dry disinfection. Finally, as future perspective, it could be possible to improve operative performances by means of a specific experiment, aiming to further reduce LPG consumption and costs.

References
Gradel K.O., Sayers A.R. & Davies R.H. 2004. Surface disinfection tests with Salmonella and a putative indicator bacterium, mimicking worst-case scenarios in poultry houses. Poult Sci, 83, 1636-1643.
Gradel K.O., Randall L., Sayers A.R. & Davies R.H. 2005. Possible associations between Salmonella persistence in poultry houses and resistance to commonly used disinfectants and a putative role of mar. Veterinary Microbiology, 107, 127-138.
Marin C., Hernandiz A. & Lainez M. 2009. Biofilm development capacity of Salmonella strains isolated in poultry risk factors and their resistance against disinfectants. Poult Sci,

88, 424-431.
Mead G.C. & Scott M.J. 1994. Coagulase-negative staphylococci and coliform bacteria associated with mechanical defeathering of poultry carcasses. Applied Microbiology, 18, 62-64.
Payne J.B., Kroger E.C. & Watkins S.E. 2002.Evaluation of Litter Treatments on Salmonella Recovery from Poultry Litter. J APPL POULT RES, 11, 239-243.
Payne J.B., Kroger E.C. & Watkins S.E. 2005. Evaluation of Disinfectant Efficacy When Applied to the Floor of Poultry Grow-Out Facilities. J APPL POULT RES, 14, 322-329.