Progress In Poultry
"THROUGH RESEARCH"

A TEST OF TURKEY HATCHING EGG SANITATION PROCEDURES

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OBJECTIVES: To determine the comparative effects of three sanitation procedures on 1) the incidence of naturally occurring Arizona infection and on 2) fertility and hatchability. The three sanitation procedures were: 1) quaternary ammonium wash only; 2) quat wash followed by formaldehyde fumigation; and3) quat wash followed by ozone treatment.

MATERIAL AND PROCEDURES:

1. Source of Eggs - Eggs used were laid by a commercial flock of Nicholas-strain large white turkeys on April 15 and 16, 1975. These hens were two-thirds of the way through the breeder season, having laid about 75 eggs per bird. Hens were maintained in several outdoor pens and were inseminated on a biweekly schedule with semen diluted with Minnesota Turkey Semen Extender containing no antibiotic (pink). Test eggs were separated by the date of last insemination, which varied with the pen--April 1, 4, and 8.

Earlier in the breeder season, naturally occurring Arizona hinshawii (7:1, 7, 8) was isolated in eggs from this flock: in 10-day candle-outs, in dead-in-shell, and in pips.

2. Egg Handling - Relatively clean eggs were collected in plastic filler flats 4 times a day from rice-hull nests. After each collection they were washed for 3 minutes at 110 to 114°F in a tub-type Potter egg sanitizer (Roser Co., Salt Lake City), 5 flats atatime strapped together. Wash water, which then had a pH of 7.7, was changed after each. collection and contained Duo-Quat 40 percent (Poultry Antigen Laboratories,Ontario,California) mixed to contain approximately 250 ppm quaternary ammonium and 12 ppm EDTA.

3. Treatments - All eggs were washed as described above. Dividing the eggs by last insemination date, approximately one-third received no further treatment, approximately one-third were fumigated with formaldehyde gas on the ranch soon after washing, and the last third were treated with ozone on the ranch soon after washing.

Formalin was used at the rate of 1.2 ml per cubic foot and added to potassium permanganate. The fumigation extended for 20 minutes in a small room with an air temperature of 70 to 75°F, water to raise humidity, and a circulating fan.

In a room with a temperature of about 60°F, ozone was pumped into a. cabinet with a circulating fan and maintained at 100 ppm for 1 hour. Ozone was produced by a unit consisting of an air compressor and a small 100-tube ozone generator (Ionaerator) manufactured by Scientific Industries of California, Garden Grove, CA. Ozone concentration was determined about every 25 seconds with a model 1003 Dasibi Corporation Monitor (Glendale, CA.) and ozone levels were controlled by turning power on and 'off to the ozone generator.

4. Incubation - Eggs were not dipped prior to setting on April 22, 1975 in a single-stage Jamesway incubator, model 1080C. Each setter tray held about 150 eggs and 24 trays were followed through incubation. Eggs in 9 trays were treated with just quat wash, eggs in 6 trays with

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quat wash and formaldehyde fumigation, and eggs in 9 trays with quat wash and ozone. Eggs were candled on May 2 at 10 days of incubation and hatched May 21. Data on each tray of eggs consisted of a 10-day candling report ("fertility report"), percentage hatch of 10-day candling report, and hatch of total eggs set. Eggs at the hatchery were fumigated with formaldehyde on the first day of incubation and after transfer to the hatcher.

5. Bacteriology - "Hatchery infertiles" after 10 days incubation were cultured at Dr. Marion Hammarlund's laboratory in Riverside using selective media for coliform organisms. Contents of each egg were sampled by swab after puncturing the large end of the hatching egg with the tip of a sterilized metal punch. Two swabs were placed in a single tube containing enrichment broth. After 24 hours of incubation, sterile swabs were dipped in the broth and streaked on agar plates--usually swabs from 6 tubes to an agar plate marked off in sections. A total of 297 tubes were examined, representing approximately twice that number of eggs. Agar plates were observed for growth after 36 hours incubation. After further incubation, samples from the same plates were'sent to Abbott Laboratories for possible identification of A. hinshawii.

RESULTS AND DISCUSSION:

1. Bacteriology - Results are shown in Table 1, both for a coliform reading after 36 hours of incubation and subsequent identification of A. hinshawii. On plating, coliforms were found in 22 tubes out of a total of 297. After further incubation, the same agar plates produced 17 samples identified as A. hinshawii.

Relating the various sanitation treatments on the farm to the bacteria isolated from hatchery infertiles, we find no statistical difference between treatments. Small amounts of formaldehyde and ozone may well have penetrated the egg shell after washing, but these two treatments show no reduction in the number of isolates made. Also supporting the idea of penetration of the washed egg by formaldehyde and ozone during farm treatment are small decreases in candling reports and hatch of total eggs, as reported below. The results suggest that Arizona organisms in this test may have been deep within the eggs, perhaps on the vitelline membrane, rather than on the shell or between the shell and shell membranes at the time of treatment.

Eggs from hens last inseminated April 1 (36-hour data) showed significantly fewer coliform isolates than eggs from hens inseminated April 4, for no apparent reason.

2. Fertility and Hatch - Fertility (10-day candling report) is shown in Table 2. Eggs treated on the farm with ozone show a. small but significant drop in candling report when compared to eggs washed in quat solution only. Differences in candling reports for date of last insemination were not statistically significant.

Data for hatch of 10-day candling report, Table 3, showthat eggs washed and treated on the farm with formaldehyde hatched poorer than those only washed. This result indicates that poorer hatches can be expected if washed eggs are also given the recommended formaldehyde farm treatment. It also makes one wonder if washed eggs are more susceptible to damage by strong formaldehyde fumigation during incubation. Date of last insemination had no effect on the hatching percentage.

Data for hatch of total eggs set appear in Table 4. While washed eggs treated on the farm with ozone hatched slightly better than those treated with formaldehyde, these two treatments were not statistically different. Eggs washed in quat solution only gave significantly better hatches than the other two treatments. Date of last insemination had no effect. Washing followed by either formaldehyde or ozone depressed the hatch slightly without showing any decrease in the number of coliform isolates.

CONCLUSIONS:

1. The pre-incubation use of formaldehyde gas for 20 minutes on washed eggs resulted in a lower percentage hatch of 10-day candle and a slightly lower hatch of total eggs set.

2. The pre-incubation use of 100 ppm ozone for one hour on washed eggs was too high a concentration: it resulted in a slightly lower candling report and a slightly lower hatch of total eggs set.

3. The added sanitation procedures were ineffective in reducing the number of coliform and Arizona isolates made in this test.

Note: The ozone generator and the ozone monitor used in this test were provided through the courtesy of Scientific Industries of California, Garden Grove, California.

Table 1. Bacteriology on 10-day candled-out eggs for coliforms. (Observations on agar plates with selective media after 36 hours incubation and after further incubation samples identified as Arizona hinshawii (A.h., paracolon))

Sanitation treatment	Last A.I. date						36-hour totals	
	April 1		April 4		April 8			
		A.h.		A.h.		A.h.		A.h.
Quat Wash	1/30[1/]	0	0/30	0	2/24	2	3/84	2/84
Quat + Form.	1/26	1	6/23	3	4/38	2	11/87	6/87
Quat + Ozone	1/42	3	6/36	5	1/48	1	8/126	9/126
36-hour totals	3/98	4	12/89	8	7/110	5	22/297	17/297

1/ Numerator is number of tubes showing growth on agar plates; denominator is total number of tubes tested.

Table 2. Ten-day candling report percentages

Sanitation treatment	Last A.I. date			Average[1/]
	April 1	April 4	April 8	
		percent		
Quat Wash	88.9[2/]	84.0	84.0	
	82.7	84.7	80.1	84.7 a
	88.7	83.9	85.2	
Quat + Form. gas	83.1	84.0	79.1	
	81.5	84.7	86.7	83.2 a b
Quat + Ozone	76.7	86.6	82.4	
	83.1	82.3	78.7	80.2 b
	74.8	81.8	75.8	
Average	82.4	84.0	81.5	82.6

1/ Percentages with the same letter are not statistically different at the 5 percent level of significance.
2/ Percentages for individual incubator trays.

Table 3. Percentage hatch of 10-day candle - #1 poults

Sanitation treatment	Last A.I. date			Average 1/
	April 1	April 4	April 8	
	percent			
Quat Wash	67.2"	84.1	84.1	
	71.0	81.9	61.5	75.0 a
	79.7	69.2	76.4	
Quat + Form.	65.0	64.3	68.4	
	68.3	63.9	62.3	65.4 b
Quat + Ozone 3	63.4	62.8	69.2	
	68.3	73.6	74.6	70.8 a b
	73.8	81.8	69.9	
Average	69.6	72.7	70.8	71.0

Table 4. Percentage hatch of total eggs set - #1 poults

Sanitation treatment	Last A.I. date			Average 1/
	April 1	April 4	April 8	
	percent			
Quat Wash	59.72'	70.7	70.7	
	58.7	69.3	49.3	63.6 a
	70.7	58.0	65.1	
Quat + Form.	54.1	54.0	54.1	
	55.6	54.2	54.0	54.3 b
Quat + Ozone 3	48.6	54.4	57.0	
	56.8	60.5	58.7	56.8 b
	55.2	66.9	53.0	
Average	57.4	61.0	57.7	58.7

1/ Percentages with the same letter are not statistically different at the 5 percent level of significance.
2/ Percentages for individual incubator trays.

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