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Research Note—

Risk Factors Associated with Poult Enteritis Mortality Syndrome—Positive Turkey Flocks

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SUMMARY. Poult enteritis mortality syndrome (PEMS) has been an economically devastating disease in North Carolina since the early 1990s. Though much is known about the disease, many questions remain unanswered about the syndrome, including its cause, transmission of causative agent(s), and control methods. This study was designed to investigate the association between PEMS and farm management factors. A prospective longitudinal study was conducted by collecting farm data and monitoring weekly mortality in 54 commercial turkey flocks raised in PEMS-affected regions. Univariate and multivariate statistical analyses revealed that enhancing rodent control methods was negatively associated ($P = 0.0228$) with PEMS.

RESUMEN. *Nota de Investigación*—Factores de riesgo asociados con lotes de pavos positivos al síndrome de mortalidad y enteritis.

El síndrome de mortalidad y enteritis en pavos ha sido una enfermedad con consecuencias económicas devastadoras para la industria avícola de Carolina del Norte desde el inicio de la década de 1990. Aunque cada día se conoce más acerca de la enfermedad, aún quedan muchas preguntas sin contestar, incluyendo su causa, transmisión del agente causal y los métodos de control. Este estudio se diseñó para investigar la asociación entre el síndrome de mortalidad y enteritis en pavos y los factores de manejo en las granjas. Se realizó un estudio longitudinal tomando datos de las granjas y supervisando la mortalidad semanal en 54 lotes de pavos comerciales criados en regiones afectadas por el síndrome. Los análisis estadísticos uni y multivariados revelaron que mejorando los métodos de control de roedores estuvo negativamente asociado ($P = 0.0228$) con el síndrome.

Key words: poult enteritis mortality syndrome, risk factors, turkey, farm management

Abbreviations: EMT = excess mortality of turkeys; PEMS = poult enteritis mortality syndrome; SMT = spiking mortality of turkeys; TCV = turkey coronavirus

Poult enteritis mortality syndrome (PEMS) changed the nature of the turkey industry in North Carolina during the 1990s. Through corporate, university, and government efforts, research has generated much knowledge about this disease (1,2,3,4,5,6,7,8,9). Even so, to date, many questions remain unanswered about the syndrome, including its cause, transmission of causative agent(s), and control methods. Because no diagnostic test is available, PEMS has been defined as a clinical syndrome (1,2). The clinical syndrome associated with PEMS has

been characterized as excess mortality of turkeys (EMT) and spiking mortality of turkeys (SMT). EMT is defined as mortality greater than 2% for any 3-wk period during the brooding phase of production, and SMT is defined as mortality greater than 9% for any 3-wk period during the brooding phase of production. PEMS has been linked to turkey coronavirus (TCV), with areas having a higher prevalence of TCV also having an increased prevalence of PEMS. The relationship of TCV to the PEMS syndrome has been described (4), and TCV is

considered to be a separate disease but can complicate PEMS cases. Establishment and implementation of control efforts for PEMS have been based on observation and educated guesswork. This study was designed to investigate the possible associations among farm management factors and PEMS in North Carolina turkey flocks. Identification of statistically valid risk factors should allow the establishment of control measures based on scientific data rather than guesswork.

MATERIALS AND METHODS

Study design. A prospective longitudinal study was conducted with commercial turkey flocks reared in two areas of North Carolina. Both areas had historic evidence of PEMS. Historically, PEMS occurred in these areas starting in mid-May and continuing through October. Flocks were selected for the study on the basis of placement dates. Flocks placed between May 15 and July 15 were eligible for inclusion in the study so that they could be monitored closely for at least 6 wk with mortality data collection continuing until slaughter. Historic disease status with regard to PEMS for individual farms was not revealed to investigators prior to flock selection. Flocks from four commercial companies were monitored. Monitoring began before placement and continued through the life of the flocks.

Mortality. Mortality was monitored daily and was reported as number of birds dead each day, which included cull birds. Weekly mortality was used for the analysis and was calculated as the number of birds that died over a 7-day period. This number was then divided by the number of birds placed and multiplied by 100 to obtain weekly mortality percentage.

Health status. Two health status groups were identified: healthy and PEMS positive (PEMS+). Healthy flocks were defined as those that did not experience mortality patterns consistent with PEMS.

PEMS status. PEMS status was determined with the use of the clinical definitions previously defined by Barnes *et al.* (1,2). Mortality experienced the first week after placement was not included in the analysis because of possible effects of holding, shipment, and placement on mortality. Mortality rates from weeks 2 through 6 were analyzed, and those flocks experiencing unexplained 3-wk cumulative mortality greater than 2% in weeks 2–4, 3–5, or 4–6 were considered positive for PEMS.

Data collection. A questionnaire was designed in order to identify farm management risk factors that may be associated with PEMS. This survey was pre-tested and adjustments were made before implementation. Growers were interviewed in person (with the exception of five farms) and asked to answer 100

questions related to physical characteristics (e.g., topography, facility age, age, types of equipment) of their own farms, proximity to other farms, proximity of turkeys to other animals (including pets), biosecurity practices (both historic and current), mortality disposal, litter management, and farm worker profiles (including relationships with other poultry workers). Corporate employees were asked to provide information on flock size, hatchery source, breeder flock source, and disease history for each farm. Interviewers verified answers during weekly farm visits (except for five farms where data collectors were restricted from entering the production area).

Statistical analyses. Data gathered from the survey were entered into a Microsoft Excel (Microsoft Corporation, Redmond, WA) file. Descriptive analyses, including summaries and graphics, were generated with Excel. Data were transferred to Statistical Analysis System (SAS Institute, Inc., Cary, NC) for univariate and multivariate analyses. The outcome variables were categorical (PEMS+, healthy). Differences between means of PEMS+ and healthy flocks were compared by Student *t*-test for continuous variables and the chi-square test for categorical variables. A total of 478 potential risk factors (variables) were examined. A *P*-value ≤ 0.20 was used as the cutoff point for a variable to enter the multivariate analysis. Continuous independent variables were left unaltered, but dummy variables were created for each categorical variable for inclusion in the multivariate analysis. Variables were then entered in a stepwise fashion into a main effects model by the Logistic Procedure (SAS Institute, Inc.). Variables with a *P*-value ≤ 0.05 were considered significantly associated with the outcome (PEMS+).

RESULTS

Sample size. Because of missing data for some variables, the final logistic regression model had eight missing observations. Of the 47 remaining flocks, 18 were PEMS+ and 29 were healthy.

Management practices. Of the 478 management factors screened, 44 were associated with PEMS+ flocks at a *P*-value of 0.20 and related to factors in Table 1. Of the 44 variables, five were continuous and 39 were categorical.

Association between PEMS+ and management factors. None of the continuous variables was significantly associated with PEMS+ flocks in the multivariate analysis. Only one categorical management factor (enhanced rodent control practices because of location in a PEMS+ area) was significantly as-

Table 1. Management factors (general areas, $n \neq 44$) associated with PEMS+ flocks at P -value ≤ 0.20 (univariate analysis).

| Management factor | |
|--------------------------------------------------|--------------------------------------------------|
| Company raising birds | Ventilation type (natural <i>vs.</i> forced air) |
| Hatchery supplying birds | Visits by meter readers (yes/no) |
| Proximity to cattle (miles) | Load-out crews (biosecurity precautions) |
| Proximity to swine (miles) | Superchlorination of waterlines |
| Distance to nearest road (feet) | Drinker cleaning practices |
| Visitor restrictions (gates, locks, signs, etc.) | Mixing of disinfectants and insecticides |
| Rubber boots required for visitors | Interval between clean-up and new flock |
| Hand sanitation required for visitors | Downtime—time between flocks |
| Gloves required for visitors | Poult processing at hatchery |
| Coveralls required for visitors | Disinfection between flocks (when) |
| Footbaths used by workers | Disinfectant types used |
| Litter management practices | Mortality removal practices |
| Staff training in biosecurity | Rodent control practices |
| Pets in production area | Fly control practices |
| Proximity to trees (feet) | Number of birds in flock |

sociated with PEMS+ flocks at a P -value ≤ 0.05 (Table 2). Enhanced rodent control practices because of location in a PEMS+ area was negatively associated with PEMS+ flocks, i.e., the risk of becoming PEMS+ decreased as enhanced rodent prevention increased. The odds ratio of 0.1 indicates a 10-fold reduction in risk of becoming PEMS+ with enhanced rodent control practices.

Though not significant at $P \leq 0.05$ and not included in the final model, two other factors were highly correlated with PEMS+ flocks. These were hatchery of origin ($P = 0.0709$) and use of an outside contractor to remove used litter ($P = 0.1852$).

DISCUSSION

The major focus of this study was to identify farm management risk factors associated with PEMS+ flocks. In the absence of a causative agent and diagnostic test for PEMS, growers and integrators requested help in identifying areas in which they could work to reduce their risk of becoming PEMS+. Because the risk of disease spread was thought to be high, limita-

tions were placed on the numbers of farms each company was willing to enroll in the study. Additional restrictions were placed on visiting more than one farm per day. In some instances, research personnel were not allowed on the farm but retrieved beetle, fly, and rodent data from mailboxes. In these instances, the questionnaire was not administered by interview but was self administered and answers were recorded by the growers. This procedure contributed to the missing observations in the final analysis.

This study showed a negative association between PEMS+ flocks and enhanced rodent control practices. Enhanced rodent control was defined as company-mandated changes in products and/or procedures geared toward reducing rodent populations. Company audits were in place in some instances to monitor the success of rodent control programs. Evidence of rodents was monitored by study personnel by visualization of presence/absence of rodent tunnels and inspection of bait stations. Those growers who had enhanced their rodent control initiatives as a precaution for PEMS had a 10-fold decrease in risk of contracting the disease. If rodents carry the PEMS agent(s) or if better

Table 2. Multivariate analysis to investigate association between PEMS+ and farm management factors by logistic regression.

| Management factor | Coefficient | P -value | Odds ratio (95% CI) |
|----------------------------------|-------------|------------|---------------------|
| Enhanced rodent control measures | -2.1914 | 0.0228 | 0.1 (0.013–0.966) |

rodent control is a proxy for other improved management practices is not clear, but clearly rodent control should be a high priority for farms in PEMS (+) areas.

Although the associations between PEMS+ and other factors such as hatchery ($P = 0.0709$) and litter removal by outside contractor ($P = 0.1852$) were not statistically significant, these factors deserve consideration by growers and integrators. First, the low sample size, imposed by logistics and reluctance to participate in the face of potential biosecurity risks, limits the power to detect statistical significance. Second, at a time birds are dying and decisions are being made on educated guesswork, waiting for a 95% confidence level to take action may be unrealistic. Depending on the costs associated with implementing a change, examination of practices that have a P -value = 0.1852 may be acceptable. This could mean conducting an additional study focusing on the practice in question or, if cost effective, implementing changes based on an 82% confidence level.

Concerning the variables examined that were not associated with PEMS, this information might be most valuable for the industry. Negative results provide much needed information on which factors are not important. When prioritizing areas for immediate attention in a disease outbreak, negative results provide a much shorter list of areas of true concern. This method allows more focus on areas that are associated with the disease.

This study provides much needed information on which factors are, but also which factors are not, associated with PEMS+ flocks. In addition to statistically valid risk factors, this study provided growers and integrators information on where to target their limited resources in preventing PEMS.

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