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Low-Pathogenicity Avian Influenza Virus in Live Bird Markets—What About the Livestock Area?

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SUMMARY. Low-pathogenic avian influenza virus (AIV) continues to be isolated from the live bird markets (LBMs) in the Northeastern United States. Recent years have seen increasing numbers of these markets opening and an expansion of the type of animals they sell in conjunction with traditional live poultry. Specific-pathogen-free chickens were released into the livestock area of 13 New York City LBMs and then tested for evidence of AIV. We were able to recover virus or demonstrate seroconversion among the chickens introduced to four of the markets.

RESUMEN. Virus de influenza aviar de baja patogenicidad en centros de mercadeo de aves vivas—que se conoce acerca de las zonas de produccio´n?

Los virus de influenza aviar de baja patogenicidad continúan siendo aislados a partir de centros de mercadeo de aves vivas en el Noreste de los Estados Unidos. En años recientes se ha venido incrementando el número de centros de mercadeo de aves vivas además de una expansión en el tipo de animales que se venden junto con las aves domésticas tradicionales. Se colocaron aves libres de pato´genos especı´ficos en las zonas de produccio´n de 13 centros de mercadeo de aves vivas en la ciudad de Nueva York y fueron posteriormente evaluadas para determinar la presencia del virus de influenza aviar. Se aisló el virus de influenza o se demostró seroconversio´n en las aves introducidas en 4 de los 13 centros de mercadeo.

Key words: avian influenza, live bird markets, livestock

Abbreviations: AGID = agar gel immunodiffusion; AIV = avian influenza virus; LBMs = live bird markets; NVSL = National Veterinary Services Laboratories; SPF = specific pathogen free

Since the fall of 1994, low-pathogenic avian influenza virus (AIV) has been isolated from poultry from live bird markets (LBMs) in New York and New Jersey (D. A. Senne, pers. obs., 1994). In 1998, the New York Department of Agriculture and Markets promulgated regulations in an effort to control AIV in the LBMs (2). The regulations addressed the flock status of the arriving birds and sanitation levels in the poultry area of the markets. In recent years we have found that an increasing number of New York markets include the sale of red meat such as sheep, goats, and calves or small steers. Poultry workers in the LBMs often move back and forth freely between the red meat and the poultry areas as work demands. We attempted to determine what, if any, role the presence of livestock play as a possible source of infection for poultry in the markets.

Since 1994, the number of LBMs in New York has nearly doubled from 44 to more than 80 markets. Whereas earlier the population served by these markets was primarily Asian, currently many of the markets serve a population demanding Halal slaughter. Over 20 of these markets now offer livestock as well as poultry to their customers.

Although the New York City LBMs must keep separate the area in which livestock are housed, the poultry area, and the slaughter area, the separation usually consists of a wall to divide poultry from
livestock, with a door for access. Often the livestock area will be used to store feed and some of the empty coops used for the poultry. In most markets the workers routinely move between the two areas.

Prolonged viability of AIV was demonstrated during the Pennsylvania outbreak in 1983–84, in which virus was isolated from wet manure up to 105 days after the depopulation of an infected flock (1). Although we could find no evidence in the literature of cattle, sheep, or goats maintaining the viruses of concern (H5 or H7 AIV), we postulated that the virus potentially could be maintained in the manure pack associated with the livestock. If so, this could serve as a reservoir for virus reintroduction to birds entering the market.

MATERIALS AND METHODS

New York City LBMs selling livestock were asked to voluntarily participate by allowing specific-pathogen-free (SPF) chickens to be placed in the livestock area. All markets selected for the study had at least one isolation of AIV during the previous 9 mo. On February 22, 2001, five to seven SPF chickens (approximately 1 yr old) were placed in each of 13 markets. The birds were identified and tested for AIV and AIV-specific antibodies before being released into the livestock area.

Per state regulations, live poultry must be separated from the live red meat area (3). This also applies to the processing area. The SPF birds were released into the livestock area but were confined to that area as long as the door remained closed between the poultry area and the livestock area. Market owners were instructed to provide feed and water access to the SPF birds and to keep them confined in the livestock area. They were also instructed that should the bird(s) escape to the poultry area, they were not to be returned to the livestock area but were to be captured and contained in the poultry area. Inspectors visited the markets weekly to collect samples and to determine the housing status of the SPF chickens.

The SPF chickens were sampled on day 4, day 15, and then weekly through 8 wk postplacement. Tracheal and cloacal swabs were collected, as were blood samples. Virus isolation was performed in embryonating SPF chicken eggs at the National Veterinary Services Laboratories (NVSL) (4). Serum samples were tested by the agar gel immunodiffusion (AGID) test at Cornell University (5). Sera positive on the AGID test were forwarded to the NVSL for subtyping (4). In addition, environmental swabs were collected from the poultry area of the market to compare to the findings in the SPF study chickens. In all instances, samples were first collected from the SPF chickens to avoid contamination being carried by inspectors from the live bird sales area back to the SPF birds.

RESULTS

Baseline sampling of the SPF chickens was negative for evidence of avian influenza of any type. After being in the markets for varying periods, the SPF chickens in four markets demonstrated evidence of avian influenza.

Market 1 had H7N2 AIV isolated from the SPF chickens 4 days after placement. This virus was also isolated from the poultry environment of the market. At 15 days postplacement, virus was again detected from the SPF chickens and the poultry sales environment. Antibodies to AIV were detected in the SPF chickens on day 4.

Market 2 was negative until week 4 of the study. At that time, the market poultry environment was positive for AIV subtypes H5 and H7, but the SPF birds were negative for AIV and antibody. At week 5, the SPF birds were positive for H7N2 AIV, and the concurrent environmental sampling of the bird market was negative for virus. At week 7, the SPF chickens showed evidence of seroconversion to AIV subtype H7N2, whereas the market samples remained negative for AIV.

Market 3 was negative for evidence of influenza until day 15. At that time, the SPF chickens were positive for antibodies to AIV subtype H7N2. Sampling in the poultry environment and among the SPF chickens did not yield AIV.

Market 4 had positive H7N2 AIV SPF birds from swabs collected on day 4. At that time, specific antibody to H7N2 AIV was also detected among these SPF chickens. No virus was recovered from environmental swabs of the poultry area in the market. On day 15, H7N2 AIV was isolated and seroconversion detected among the SPF chickens, but no virus was recovered from the environment of the poultry area.

DISCUSSION

There were several limitations of this study. Although according to New York law, the area holding the livestock must be separated from the poultry area, very often the separation is effected simply by closing a door between the two areas. The study birds were released to move freely in the livestock area. It is possible that the study birds could have entered the LBM poultry area (although
market owners denied this), become infected with influenza virus, and then returned to the livestock area. Although possible, we found that the birds established a 'comfort zone' within the livestock area, and when offered the opportunity to exit the market or move to the poultry area, they did not do so. A second limitation was loss of study birds to follow up. In some instances the market owners became concerned that the study birds were being overly crowded by the livestock, and they moved the birds to the poultry area. Five market owners removed the SPF chickens from the livestock area and were dropped from the study for this reason.

One market each removed the birds after only 1 wk, 2 wk, and 4 wk. Two others removed the birds after 3 wk. All birds were negative on the sampling prior to removal.

While considering the potential problems with introducing SPF chickens to the livestock area, we chose this option rather than collecting environmental samples from the livestock area. We felt that chickens allowed to freely roam the manure pack and entire area associated with, but not part of, the LBM would provide a better determination as to the maintence of influenza virus in the livestock area. Had we opted to collect environmental samples from the livestock area and found them negative for virus, we would still be uncertain as to whether or not influenza virus was present in this setting. The proposed regional LBM 3-day closure did not address animals other than poultry in these markets, and we wished to determine if the livestock area should be included in the closure.

Despite these limitations, we were able to demonstrate that chickens allowed to roam freely among the livestock areas associated with LBMs could be used to detect the presence of AIV. In two of the markets, sampling the poultry area failed to detect AIV subtype H7N2, while SPF chickens placed in the nearby livestock area were positive for the H7N2 AIV. This evidence indicates that the livestock area associated with poultry markets may serve as a potential source of reintroduction of influenza virus to the poultry area of the market. For this reason, New York and New Jersey included depopulation of livestock, as well as cleaning and disinfecting of this area, during the regional closure efforts in April 2002.

REFERENCES


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