

Seroprevalence of *Mycoplasma synoviae* and *Mycoplasma gallisepticum* in one day old broiler chickens in Libya

F. S. Elgnay^{1*} and S. M. Azwai²

¹Technical Center of Animal Health of Libya

²Faculty of Veterinary Medicine, University of Tripoli, Libya

Abstract

The aim of this study was to carry out a serological survey of antibodies against *Mycoplasma gallisepticum* (MG) and *Mycoplasma synoviae* (MS) in 1500 one day old broiler chickens sera were examined by serum plate agglutination test. Antibodies against MG and MS were detected 3.4% and 6.4%, respectively. The seroprevalence of MG was 5.2% in chickens from imported fertile eggs and 0% in chickens from local fertile eggs, whereas for MS was 9.3% in chickens from imported fertile eggs and 0.8% in chicks from local fertile eggs.

Key words: *Mycoplasma gallisepticum*, *Mycoplasma synoviae*, Serum plate agglutination test, Chickens, Libya

*Corresponding author: Tel: +218913815623

E-mail address: fawzia_salem@yahoo.com

Introduction

Mycoplasmas are widespread in nature and infect a wide range of hosts. Species from the genus *Mycoplasma* (over 110 species) have been isolated from mammals, birds, reptiles and fish. Pathogenic *Mycoplasma* species cause diseases in domesticated animals and lead to high economic losses in farm animal production. *Mycoplasma* infections are frequent, or even enzootic in pigs, poultry, and ruminants on numerous farms all over the world (Bencina et al., 2005). Some of them are economic importance to the poultry industry through their causation of disease (Jordan and Pattison, 1999). For veterinary medicine, the most important *Mycoplasma* which has been isolated from domestic avian species is *M. gallisepticum*, *M. synoviae*, *M. meleagridis* and *M. lowae* (Stipkovits and Kempf, 1996).

Avian Mycoplasmosis transmitted vertically through the eggs or horizontally often by direct contact. Indirect transmission via people, wild birds, drinking water, litters or breeding materials may play major role in the initiation of outbreak because of possible persistence of mycoplasma in environment (Marois et al., 2000). The *MG* and *MS* are responsible for respiratory disease and synovitis (Silveira et al., 1994). Losses are due to poor feed conversion, decreased egg production, increased embryo death and culling of hatched progeny and carcass condemnation at processing (El- Shater et al., 1995). There are few published papers on avian mycoplasma infection in Libya (Elgany and Azwai, 2009) and this study focuses on seroprevalence of *MG* and *MS* antibodies in chickens from imported fertile eggs and local chickens in Libya.

Materials and Methods

Chickens

One day old broiler chickens were randomly collected from 5 different hatcheries in Tripoli, Aljafarah (35 km west of Tripoli) and Tarhowna area (100 km south east of Tripoli) 1500 samples of one day old chicken were from 5 different hatcheries of A (30%), B (10%), C (6.6%), D (20%) and E (33.4%) as shown in figure 1. The imported one day old chickens were from Spain (13.4%), Belgium (6.6%), Holland (33.3%) and USA (13.4%), while (33.3%) were from Libya as shown in figure 2.

Blood Samples

1500 one day old broiler chickens were bled from the jugular vein. The collected blood samples were allowed to clot in sterile tubes at room temperature. After clotting, sera were separated by centrifugation. Fresh sera were tested against *MS* and *MG* by serum plate agglutination test (SPA).

SPA

Antigens for *MS* and *MG* were provided by Charles River Laboratories, Inc USA. The SPA test conducted by contacting and mixing of test serum with serum plate antigen on glass at room temperature (Alan et al, 1988)

Results

Samples were positive for specific antibodies against *MS* and *MG* as shown in figure 3. The seroprevalence of *MS* was higher in D and A hatcheries (12%) in comparison with C (3%), E (0.8%), and B (0%) hatcheries. The highest seroprevalence of *MS* antibodies were recorded in the chickens of imported eggs from Spain (16%), Holland (11%), and Belgium (6%); while it was 0.8% and 0% in sera from Libya and USA chickens consequently. *MG* seroprevalence was similar to *MS* as the higher seroprevalences were recorded in D (9.3%) and A (5.3%) hatcheries while it was 0% in C, E and B hatcheries (figure 4). The seroprevalence of *MG* antibodies in chickens imported from Spain was 12.5%, while it was 4.8% and 3% in chickens from Holland and Belgium consequently. Blood samples originated from Libya (local) and USA showed no prevalence (0%) of *MG* antibodies (Figure 5). As figure 6 shows, the seroprevalence of *MS* and *MG* antibodies in imported chickens were higher than local chickens.

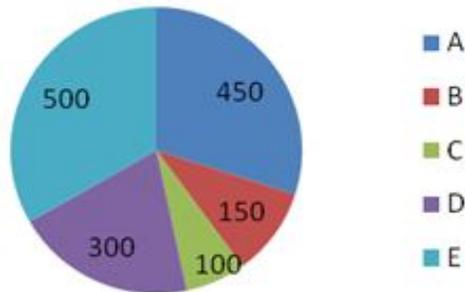


Fig 1. Number of collected samples from different hatcheries

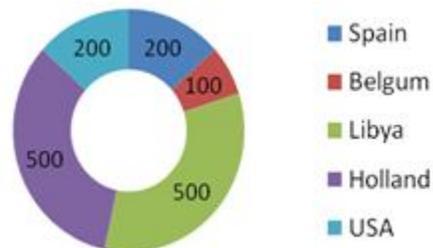


Fig 2. Origin of chickens

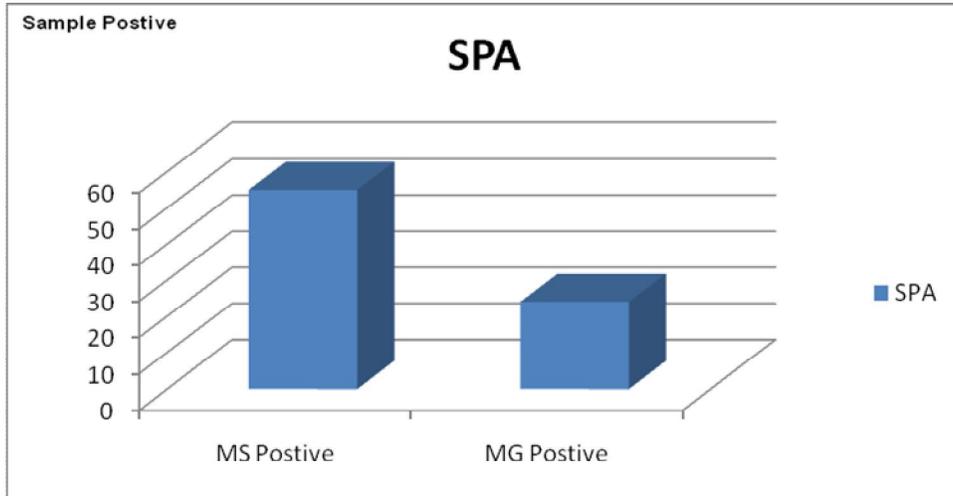


Fig 3. Percentages of *M. Synoviae* (MS) and *M. Gallisepticum* (MG) seropositivity by SPA test.

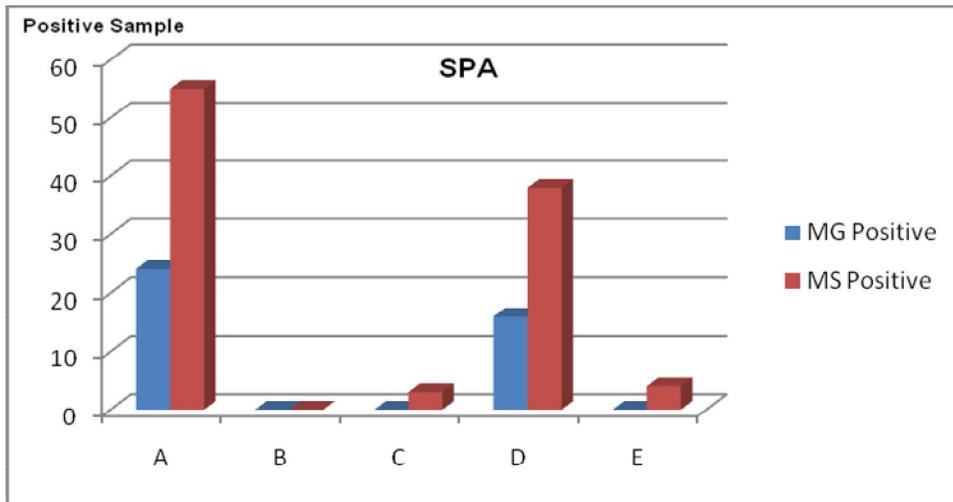


Fig 4. Seropositivity of *MS* and *MG* (%) in different hatcheries (A, B, C, D and E)

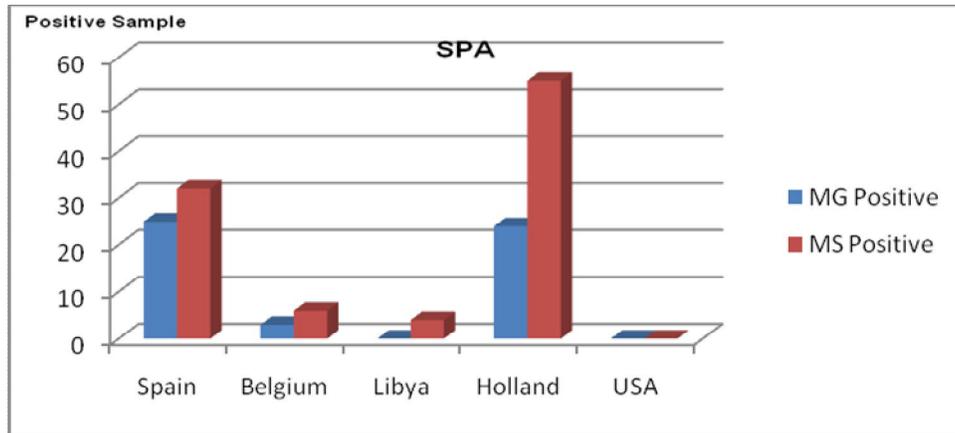


Fig 5. Seropositivity of *MS* and *MG* (%) based on chicken origins

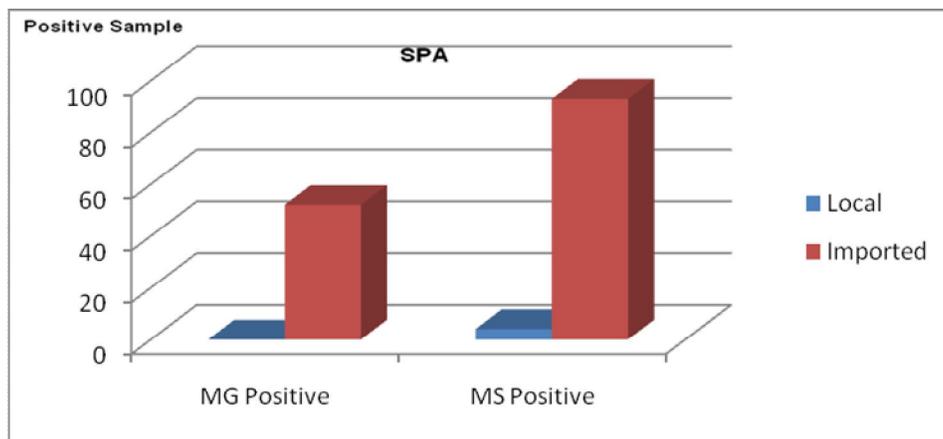


Fig 6. Seropositivity of *MS* and *MG* (%) in local and imported origin chickens.

Discussion

Avian mycoplasmosis transmitted vertically through the eggs or horizontally often by direct contact, ill birds, unaffected carriers and sensible animals. Indirect transmission via people, wild birds, drinking water, litters or breeding materials may play a major role in the initiation of outbreak because of possible persistence of mycoplasma in environment (Marois et al., 2000).

Mycoplasma synoviae responsible of infectious synovitis and the economic consequences may be important because of decreased egg production, growth and hatchability rates. *Mycoplasma gallisepticum* is a pathogen that commonly affects intensively reared chickens and turkeys and economically impacts the poultry industry through increased mortality. Uncomplicated cases of *MG* infection do not always cause overt clinical signs or mortality but can result in sub-optimum production and downgrading of carcasses (Charleston et al., 1998). Rapid detection of *MG* infection and differentiation of *MG* strains is essential in order to effectively monitor outbreaks, trace the point source of contamination and design relevant controlling strategies (Levisohn and Kleven, 2000). The sensitivity of SPA was superior to that of ELISA and HI in the ability to detect antibodies formed in early response to *MG* infection (acute stage); however, both ELISA and HI tests had higher degree of specificity.

It is extremely important for poultry producer to be able to detect *MG*-infected birds early in the course of the infection (Kempf et al., 1994). Prevention and control programs based on strict biosecurity, surveillance (serology, culture, and molecular identification) and eradication of infected breeder flocks are preferable (Raviv et al., 2007). The most common method of diagnosis of either *M. gallisepticum* or *M. synoviae* infection in poultry farms is determination of antibody status. The Rapid serum agglutination test (RAT) is commonly used to screen for Mycoplasma infections (Brown et al., 1996).

In this study, rate of 3.4% positivity using SPA test were recorded for *MG* antibodies, whereas seroprevalence of *MS* antibodies was 6.4% and slightly higher than *MS* antibodies. Ewing et al. (1996a) reported a slightly higher rate (11.2%) of *MG* using SPA on commercial broilers in Florida (USA). However, Feberwee et al. (2008) in Holland reported much higher rate of 73% positivity in commercial layers, while it was only 6% in both broiler parents and broiler frames, this rate is similar to our report for *MS* antibodies. Ewing et al. (1996b) tested a flock showing clinical signs of the disease in Florida (USA), and reported a high rate of (73%) positivity for *MS* antibodies. From Bangladesh, Sarkar et al. (2005) reported that sero-prevalence of *MG* infection was 58.9% in the study area. In an experiment conducted by Pakpinyo (2003) in Thailand, they showed that birds had detectable antibodies of 20% seropositivitiy by SPA after 3 weeks of inoculation with *MG* live vaccine, it was slightly higher (30%) when they experimentally infected the group.

In another experiment carried out by Fiorentin et al. (2003) in Brazil, they housed a flock of broiler known to be free of *MS* and *MG* (using serology, culture and PCR techniques with farms known to be endemic for *MS*), detectable antibodies against *MS* using SPA test were 5% at 22 week of age. Silveira et al. (1994) in Brazil showed that when 12 week-old layers were experimentally infected by *MG* and *MS* had a seropositivity rate of 100% for *MG* and 55.5% for *MS* after 32 day of infection. Kempf and Gesbert (1998) carried out an experiment in France on bird's 57-weeks-old free of *MG*, where they infected them with *MG*; signs of disease started to show after 3 days of the challenge. While it has been taken 10 days for antibodies in 100% of bird to appear when SPA was used.

This is the first study of seroprevalence of *MS* and *MG* in Libya. Results indicated that *MG* and *MS* have a great impact on the Libya poultry farms.

Acknowledgements

In particular, deepest thankfulness to Dr. Masoud abu soai, Secretary of the Management Committee of the Agricultural Bank who is currently nominated for the study to obtain master degree. In addition, deepest thankfulness to Dr. Abubaker Almansouri, Secretary of the Committee of the General People's Agriculture, Livestock and Marine for encouragement and moral support of this work.

REFERENCES

- Alan, P., S. Avakian, H. Kleven, and J. R. Glisson. 1988. Evaluation of the Specificity and Sensitivity of Two Commercial Enzyme-Linked Immunosorbent Assay Kits, the Serum Plate Agglutination Test, and the Hemagglutination-Inhibition Test for Antibodies Formed in Response to *Mycoplasma gallisepticum*. *Avian diseases*, 32(2): 262-272.
- Bencina, D., J. M. Bradbury, L. Stipkovits, Z. Varga, A. Razpet, A. Bidovec, and P. Dovc. 2005. Isolation of *Mycoplasma capricolum*- like strains from chickens. *Veterinary Microbiology*, 112(1): 23-31.
- Brown, M. B., J. M. Bradbury, and J. K. Davis. 1996. Pages: 289-290 in: *Molecular and Diagnostic Procedures in Mycoplasma*. J. G. Tully, and S. Razin, ed. Volum 2, Second ed, Academic Press. London, UK.
- Charleston, B., J. J. Gate, I. A. Aitken, and L. Reeve- Johnson. 1998. Assessment of the efficacy of tilmicosin as a treatment for *Mycoplasma gallisepticum* infections in chickens. *Avian Pathology*, 27:190-195.
- Elgany, F.S., and S. M. Azwai. 2009. Detection and identification of avian *Mycoplasma* in one day old chick (broiler) in Libya. pp264 in: *XVIth World Veterinary Poultry Association Congress*. Marrakakesh, Morocco.
- El- Shater, S.A.A., E. A. Ebiary, S.A. Essia, and R. E. K. Nahla. 1995. Direct detection of *Mycoplasma Gallisepticum* (s6 strain) by polymerase chain reaction (PCR). *Assiut Veterinary Medical Journal*, 32(64): 102-109.
- Ewing, M. L., S. H. Kleven, and M. B. Brown. 1996a. Comparison of Enzyme-Linked Immunosorbent Assay and Hemagglutination –Inhibition for Detection of Antibody to *Mycoplasma gallisepticum* in Commercial Broiler , Fair and Exhibition, and Experimentally Infected Birds. *Avian Diseases*, 40:13-22.
- Ewing, M. L., L. H. Lauerman, H. Kleven, and M. B. Brown. 1996b. Evaluation of Diagnostic Procedures to Detect *Mycoplasma synoviae* in Commercial Multiplier-Breeder Farms and Commercial Hatcheries in Florida. *Avian Diseases*, 40:798-806.
- Feberwee, A., T. S. A. Vries, and W. M. Landman. 2008. Seroprevalence of *Mycoplasma synoviae* in Dutch commercial poultry farms. *Avian pathology*, 37(6):629-633.
- Fiorentin, L., M. A. Z. Mores, I. M. Trevisol, S. C. Antunes, J. L. A. Costa, R. A. Soncini, and N. D. Vieira. 2002. Test Profiles of Broiler Breeder Flocks Housed in Farms with Endemic *Mycoplasma synoviae* infection. *Brazilian Journal of Poultry Science*, 5(1):37-43.
- Jordan, F. T. W., and M. Pattison. 1999. Pages 81-93 in: *Poultry Diseases*. 4th ed. Saunders- London.

- Kempf, I., F. Gesbret, M. Guittet, G. Bennejean, and L. Stipkovits. 1994. Evaluation of two commercial enzyme-linked immunosorbent assay kits for detection of *Mycoplasma gallisepticum* antibodies. *Avian Pathology*, 23:329-338.
- Kempf, I., and F. Gesbert. 1998. Comparison of serological tests for detection *Mycoplasma gallisepticum* antibodies in eggs and chicks hatched from experimentally infected hens. *Veterinary Microbiology*, 28(60): 207-213.
- Levisohn, S., and S. H. Kleven. 2000. Avian mycoplasmosis (*Mycoplasma gallisepticum*). *Revue scientifique et technique*, 19: 425-442.
- Marois, C., F. Oufour-Gesbert, and I. Kempf. 2000. Detection of *Mycoplasma synoviae* in poultry environment samples by culture and polymerase chain reaction. *Veterinary Microbiology*, 73: 311-318.
- Pakpinyo, S., P. Pitayachamrat, S. Saccavadit, T. Sntaswang, A. Tawatsin, and J. Sasipreeyajan. 2003. Laboratory Diagnosis of *Mycoplasma gallisepticum* infection in Experimental Layer Chicken Receiving MG Vaccines and MG Organisms. *Thai Journal of Veterinary Medicine*, 36(2): 29-38.
- Raviv, Z., S. A. Callison., N. Ferguson-Noel, and S. H. Kleven. 2007. Strain differentiating real-time PCR for *Mycoplasma gallisepticum* live vaccine evaluation studies. *Veterinary Microbiology*, 129: 179-187.
- Sarkar, S. K., M. B. Rahman, K. M. Amin, M. F. R. Khan, and M. M. Rahman. 2005. Sero-Prevalence of *Mycoplasma gallisepticum* infection of Chickens in Model Breeder Poultry Farms of Bangladesh. *International Journal of Poultry Science*, 4 (1): 32-35.
- Silveira, R., L. Fiorntin, and K. Marques. 1994. Polymerase Chain Reaction Optimization for *Mycoplasma gallisepticum* and *Mycoplasma synoviae* Diagnosis. *Avian Diseases*, 40: 218-222.
- Stipkovits, L., and I. Kempf. 1996. Mycoplasmoses in Poultry. *Revue scientifique et technique*, 15(4): 1495-1525.