MYCOPLASMA SYNOVIAE INFECTION

Slide study set #12

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This slide study set was originally created in 1983 by Dr. HARRY W. YODER, JR. and Dr. NORMAN O. OLSON and was updated in 2011.

***MYCOPLASMA SYNOVIAE* INFECTION**
By: Ziv Raviv and David H. Ley

Mycoplasma synoviae (MS) is a pathogen of chickens and turkeys, causing significant economic losses to poultry producers worldwide. Infection can be associated with upper respiratory disease, airsacculitis, synovitis, tenosynovitis, and bursitis. Disease severity has been influenced by other respiratory pathogens (e.g., Newcastle disease virus, infectious bronchitis virus), more virulent MS strains, and host species predilection (turkeys more susceptible than chickens).

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History, Distribution, and Incidence. Infectious synovitis was first described and associated with mycoplasma infection by Olson *et al.*, during the early 1950's. The causative organism was designated as Avian Mycoplasma Serotype S by Dierks et al., in 1967 and subsequently confirmed as a separate species, *Mycoplasma synoviae*, by Jordan *et al.*, in 1982. Soon after its identification MS appeared to have worldwide distribution. During the 1950's and 1960's the synovitis form of the disease was observed primarily in growing meat type chickens (broilers), while since the 1970's the respiratory form of the disease has been seen more frequently. Flocks of laying chickens are commonly infected with MS with mild or subclinical signs. The disease usually appears in turkey flocks 10 to 20 weeks of age, primarily in multiage farms and in endemically MS infected areas. The outcome of infection is significantly affected by management factors, other respiratory pathogens (e.g., Newcastle disease virus, infectious bronchitis virus), virulence of the involved MS strain, and the host species (turkeys more susceptible than chickens).

Etiology. MS is the classical cause of infectious synovitis of chickens and turkeys. MS more frequently produces a persistent infection of the upper respiratory tract which sometimes is involved with air sacculitis. MS is a very fastidious cell wall-less bacterium requiring a protein-rich medium with 10-15% swine serum, and specifically requires the addition of nicotinamide adenine dinucleotide (NAD). Updated specifications for MS culture follows:

MYCOPLASMA MODIFIED FREY'S BROTH MEDIUM

	Per Liter
Deionized distilled water	880.0 ml
Thallium acetate (10% sol.)	5.0 ml
Potassium penicillin G (aqueous)	500,000 units
Mycoplasma Frey's broth base	22.5 g
Swine serum (heated 56 C for 30 min)	120.0 ml
Dextrose	5.0 g
Phenol Red (1% sol.)	2.5 ml
NAD (1% sol.)	12.5 ml
Cysteine hydrochloride (l%-sol.)	12.5 ml

Adjust to pH 7.8 with 20% NaOH and filter Sterilize.

Add thallium acetate to the water first to avoid precipitation of proteins of media and serum. Horse serum is adequate for MG, but swine serum is best for MS. Cysteine hydrochloride is added to reduce the NAD (beta nicotinamide adenine dinucleotide) which is required for the growth of MS. For agar plates 1.5% agar is used. For potentially contaminated specimens, an extra 20 ml of 1% thallium acetate and 2,000,000 units of penicillin per liter may be added. Ampicillin (from 200-1000mg/l) may be substituted for penicillin.

Colony morphology. Colonies on solid media are best observed with a dissecting microscope at 30X magnification using indirect lighting. They appear as raised, round,

slightly latticed colonies with or without centers. Colonies with centers are commonly described as “fried egg” shaped. The “fried egg’ colony morphology is due to the lack of a cell wall and the tendency of the organisms to propagate into the agar matrix. It is important to note that walled bacteria in L-phase might demonstrate similar colony morphology, and needed to be differentiated from mycoplasmas. The colonies range from 0.1 to 0.5 mm in diameter, depending on the number of colonies present, suitability of media, and age of culture.

Cell morphology. In Giemsa-stained preparations MS cells appear as pleomorphic coccoid bodies or rods approximately 0.2 µm in diameter to 0.4 µm in length. In ultrastructural studies the MS cells appear round or pear shaped with granular ribosomes. The cells are 300-800 nm in diameter, lack a cell wall, and are bounded by triple-layered membrane.

Biochemical properties. MS ferments glucose and maltose with the production of acid, but not gas, in suitably enriched media. It does not ferment lactose, dulcitol, salicin, or trehalose. MS is phosphatase negative and produces film and spots. Most isolates of MS are capable of hemagglutinating chicken and turkey erythrocytes. Its ability to reduce tetrazolium salts is very limited.

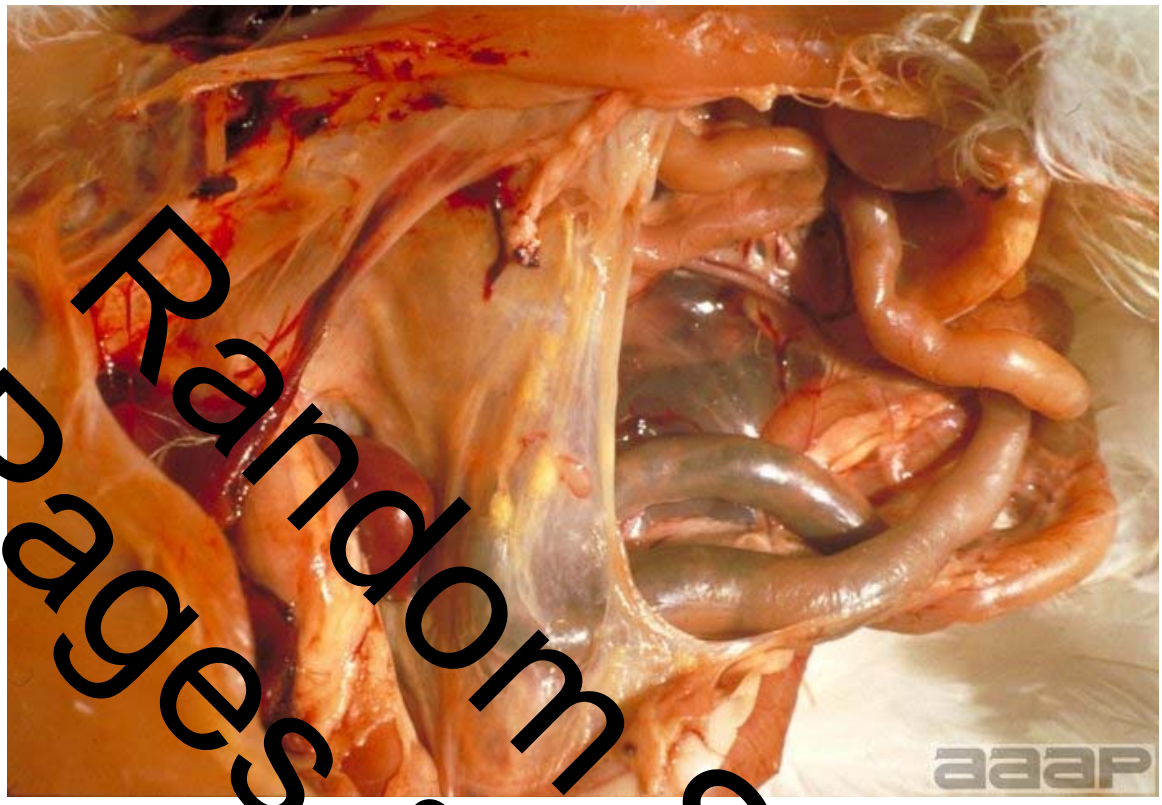
Resistance to chemical and physical agents. Resistance to disinfectants has not been determined, but MS is probably sensitive to most disinfectants as are other mycoplasmas. MS is not stable at pH 6.8 or lower. It is sensitive to temperatures above 39°C. It will withstand freezing, but the titer is reduced. Broth cultures stored at -70°C and lyophilized cultures stored at 4°C usually survive storage for several years. Survival occurred up to 3 days at room temperature on feathers and up to 12 hours in the nasal cavity of a volunteer, while survival was less than 1 day on most other materials. Viable MS organisms were detected in the environment of an isolator unit 2.5 days after the depopulation of MS-infected chickens.

Antigenic structure. MS has two immunodominant surface proteins, MSPA and MSPB, which are size and phase variable and associated with haemadsoption. These proteins are coded by a single variable lipoprotein and haemagglutinin A (*vlhA*) gene. In most MS strains VlhA is post-translationally cleaved into the N-terminal lipoprotein MSPB and the C-terminal part MSPA, which is involved in binding to erythrocytes (haemagglutinin). The mechanism of MSPA and MSPB antigenic variation is controlled by homologous recombination events between the expressed *vlhA* gene and a cluster of tens of promoter-less *vlhA* pseudogenes, located immediately upstream to the expressed gene.

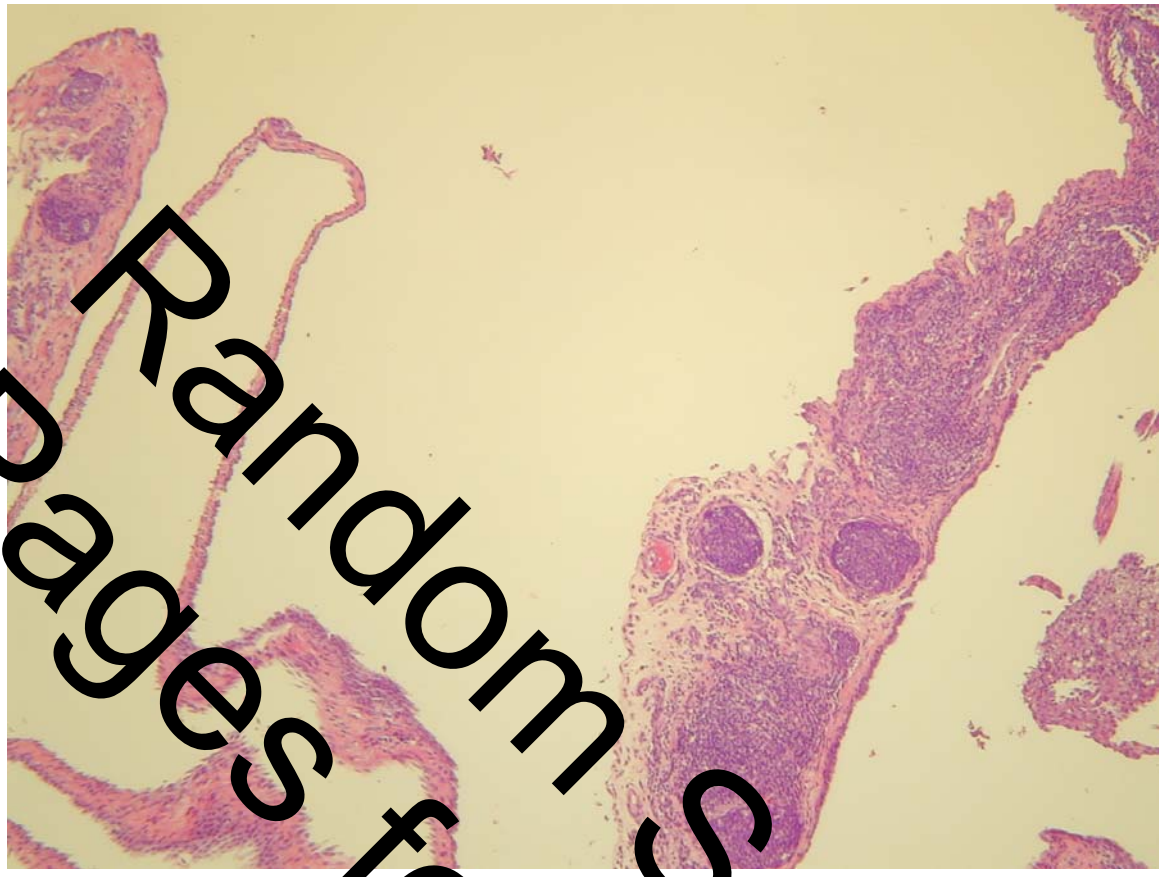
Transmission. Horizontal transmission occurs readily by direct contact between MS susceptible and infected birds. The organism can also spread by airborne dust or aerosol droplets. Spread by contact with contaminated equipment is generally assumed, but has not been well documented. However, in a recent study day-old chicks became infected after being placed in contaminated isolators. MS was demonstrated to survive on turkey feathers for 3 days, on cotton cloth for 2 days, and on poultry barn materials and human hair, ear, and nose for only few hours. Transmission occurs through the respiratory tract

SLIDE 5. Chicken with its tongue pulled aside to position the larynx for insertion of a swab to obtain tracheal exudate for cultivation of most avian mycoplasma including MS. (Slide courtesy of Dr. Stanley H. Kleven).

SLIDE 6. Normal air-sac membranes are so thin and clear that they are almost invisible. The air sacs are primarily paired extensions (thoracic, abdominal, etc.) of the air passages from the bronchioles on out beyond the lungs into various body cavity spaces. Some are within hollow bones. This slide shows a moderately inflamed air sac as noted by slight thickening of the membrane, some bloody exudate, and increasing flecks of yellowish caseous exudate as the process continues.



SLIDE 7. Extensive airsacculitis denoted by severe thickening of the air sac membrane with large masses of caseous exudate containing cellular debris and increased vascularization as the lesion is starting to regress. It is rarely possible to isolate mycoplasma organisms from such chronic lesions.



SLIDE 8. Chronic lymphoplasmacytic airsacculitis in response to MS infection in an adult layer chicken. Some sections of the air sac are in normal width (loop on the left) while other sections are greatly thickened and contain air sac's stroma fibroplasia, stromal infiltration of lymphocytes, plasma cells, and macrophages, and moderate air sac's epithelium hyperplasia. Multifocal lymphoid aggregates, some with germinal centers, can be observed. The early stages of infection are characterized by exudation of fibrin and infiltration of heterophils and lymphocytes. H&E, 50X. (Slide courtesy of Dr. Ziv Raviv).