Keeping first class eggs flying high.

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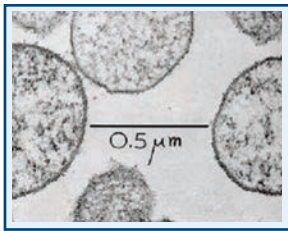
MSD Animal Health

Mycoplasma synoviae (MS) in chickens

MS IS A GLOBAL CHALLENGE AND OUTBREAKS HAVE BEEN SEEN AROUND THE WORLD SINCE THE 1960'S

- MS starts as an upper respiratory infection, which causes mild airsacculitis, with increasing risk of secondary respiratory pathogens (IB, ND, AI, E coli)[1].
- After initial infection MS may become systemic inducing infectious synovitis and/or drop in egg production and egg quality[1].



MYCOPLASMA'S

- The smallest self-replicating prokaryotes.
- No cell wall.
- Affinity for mucosal surfaces.
- Complex nutrient requirements from host.

WORLDWIDE PREVALENCE

- Prevalence is high and Mycoplasma is very persistent, infected birds are likely to carry the organisms for life.

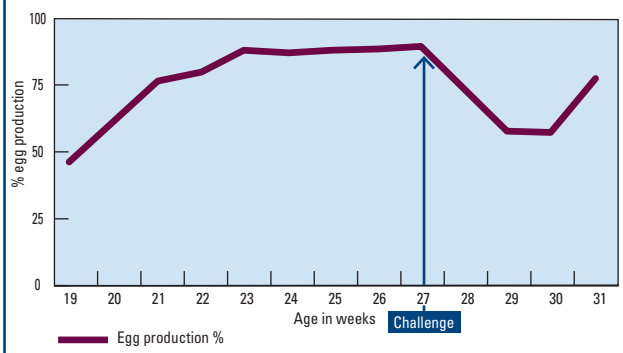
SURVEY NETHERLANDS GD DEVENTER VOLUNTARY MS MONITORING 2006[3]

Farm Type	Number of Farms	Number Sampled	% MS Positive
Meat grandparent	53	53	10
Rearing broiler parent	150	34	6
Broiler parent (production)	330	114	35
Broilers	800	185	6
Layer grandparent	13	13	0
Layer parent	50	40	25
Rearing layers	140	97	69
Layers	1250	173	73
Turkeys	75	50	16

ECONOMIC IMPACT[7]

After experimental infection of day-old broilers, a weight gain loss of 1.5 - 3.7 grams/day was shown over a 28 day period. Additionally, an increased condemnation rate (27-34%) and a 7 point increase of FCR was shown.

IMPACT OF MS FIELD CHALLENGE ON EGG PRODUCTION AFTER INFECTION WITH MS WVU 1853 FIELD STRAIN.



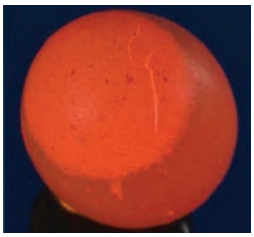
Challenge: MS field strain (WVU 1853) originated from the hock joint of a chicken. $10^{7.0}$ CFU/chicken inoculation intra airsac. *Data on file MSD Animal Health.

MS - THE HIDDEN DANGER

MS was once an endemic disease with few clinical consequences, however it's virulence is increasing.

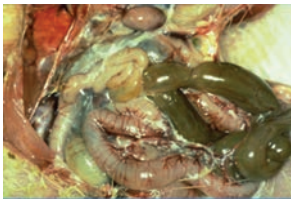
MS causes the following issues:

- Increased mortality (1-10%)
- Joint problems; synovitis, amyloid arthropathy[2], leg disorders
- Drop in weight gain and uniformity in broilers
- Increased feed conversion rate
- Drop in egg production and quality
- Eggshell apex abnormalities (EAA)[4,5]



Egg shell apex abnormalities (EAA)[5] brown and white layers*.

Synovitis



Airsacculitis

*Image reprinted by permission of the publisher (Taylor & Francis Ltd, www.tandfonline.com) from the original article: A. Feberwee , J. J. de Wit & W. J. M. Landman (2009) *Induction of eggshell apex abnormalities by Mycoplasma synoviae: field and experimental studies, Avian Pathology, 38:1, 77-85.*

Diagnosis of MS

CLINICAL SIGNS MAY PRESENT AS PHYSICAL SYMPTOMS, BUT CONFIRMATION WILL BE ATTAINED THROUGH LABORATORY TESTING

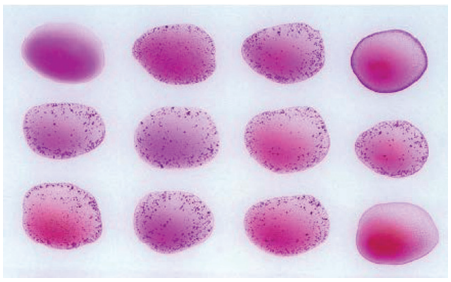
Tests include:

- Serology: SPA, Elisa, HI
- PCR plus typing (sequencing)

Standard monitoring for MS status control

- Serology: SPA, MS or Elisa MS
- PCR[6]
- Standard MS PCR based on vlhA primers for MS confirmation
- MSD Animal Health and GD Animal Health Service developed a distinguishing PCR to differentiate vaccinated birds from infected birds[8]
- Sequencing for epidemiological follow up

Nobilis® MS Live serology	SPA % positive		
Vaccine	1 wk PV	2 wk PV	3 wk PV
MG+MS	0	29	86
Control	0	0	0



SPA test

Prevention & control of MS

VACCINATION AS PART OF AN EFFECTIVE MS CONTROL PROGRAM REDUCES ANTIBIOTIC USE

MS Control Programme

- Biosecurity
- All in All out
- Monitoring
- Vaccination

MS Vaccination Programme

- Live attenuated vaccines
  - offer the most effective vaccination strategy
  - closely mimic a natural infection resulting in a strong local immunity
  - colonizes the upper respiratory tract for long periods
- Administration: Spray
- Vaccination reduces clinical signs and possible egg transmission. They do not block infection!

MS IS TRANSMITTED HORIZONTALLY & VERTICALLY!

Transmission of MS is important in relation to the control of MS.

Horizontal transmission occurs through direct and indirect contact - usually 100% of birds become infected.

Vertical transmission (parent→progeny) via hatching egg appears to be highest during the first 4-6 weeks after infection. Transmission thereafter may cease, but infected flocks may shed at any time.

MS Risk Factors:

- Egg transmission (vertical infection)
- Multi age production sites (horizontal infection)
- Dense poultry areas with high prevalence of MS infections
- Biosecurity (e.g. humans, equipment, spiking males)

Nobilis® MS live - for the prevention of MS

A NEW VACCINE FOR THE PREVENTION OF MS, THAT WILL HELP ENSURE PERFORMANCE AND QUALITY ARE MAINTAINED

Nobilis® MS live

- Nobilis® MS Live is a live attenuated MS vaccine based on the MS1 strain.
- MS1 strain is a spontaneous attenuation of the pathogenic field isolate WVU 1853.
- The original strain has been isolated from the hock joint of a chicken.
- Nobilis® MS Live is a freeze-dried vaccine containing at least ≥$10^{6.5}$ and ≤$10^{8.0}$ CFU per dose of live attenuated M. synoviae strain MS1.

Features and benefits

- Freeze dried vaccine
- Easy storage and preparation
- Convenient aerosol application
- Approved for combined use with Nobilis® MG 6/85. One application provides both MG and MS
- Reduces use of antibiotics in poultry
- Approved for use during lay

Safety of Nobilis® MS Live

SPREAD AFTER VACCINATION.
REVERSION TO VIRULENCE TEST.

Safety Experiment: Vaccine spread

- Nobilis® MS Live vaccine strain will spread to direct in-contact birds (flock mates) after vaccination.
- Vaccinated and non-vaccinated birds were mixed. No clinical response observed in either group.
- Trachea swabs and blood samples at set time points show excellent vaccine strain replication in vaccinates.
- Spread to non-vaccinated flock mates resulted in a positive SPA response in vaccinated and non-vaccinated flock mates.

RECOVERY OF MS LIVE STRAIN AFTER VACCINATION FROM TRACHEAL SWABS

Group	Strain	Dosage	Group size	Days after administration				
				14	28	42	56	70
1	MS1	$10^{7.0}$	7	3/6 50%	5/6 83.3%	5/5 100%	6/6 100%	4/7 57.1%
2	Commingled with group 1	-	7	2/7 28.6%	7/7 100%	5/5 100%	4/6 66.7%	4/5 80%
3	non-vaccinated	-	7	nt	nt	nt	nt	nt

* Data on file, MSD Animal Health

1. Number of MS recovery / number examined.

Safety Conclusions

Spread

- In case it can spread to in-contact birds without inducing clinical signs
- Recovery of vaccine strain only from infra orbital gland, trachea and airsac after vaccination.
- Field isolate WVU 1853 is also recovered from abdominal cavity, hock joint, and carpal joint.

Reversion to virulence test

Shows no increase of virulence up through 10 passages.

Efficacy of Nobilis® MS Live

AIR SAC LESION AND OVARY LESION SCORES AND THEIR EFFECT ON EGG PRODUCTION

AIRSAC AND OVARY LESION SCORES*

Vaccine	Dosage CFU/chicken	MS challenge 10^7 CFU/chicken intra airsac	Examined organ	
			Air sac[1] score	Ovary[1] lesion score
MS Live	10^7	WVU 1853	0.2 ± 0.42[2]	0.1 ± 0.32[2]
Non vac. challenge control	-	WVU 1853	1.4 ± 1.26	0.9 ± 1.20

*Data on file, MSD Animal Health (1). Mean ± SD (2). Mann-Whitney U test P< 0.05

DEMONSTRATION OF NOBILIS® MS LIVE'S EFFICACY AGAINST THE MS CHALLENGE

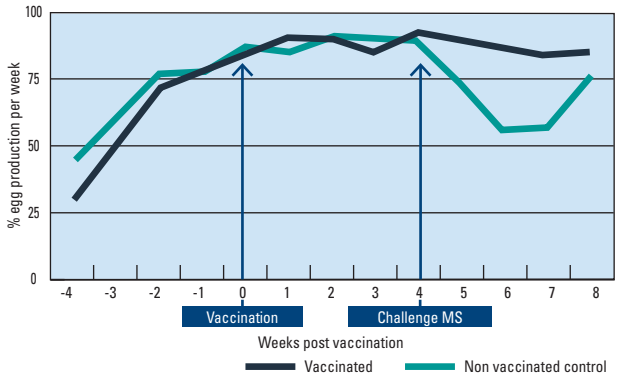
Challenge experiment

- MS challenge 4 weeks post-vaccination
- Birds vaccinated at 23 weeks of age
- Birds challenged at 27 weeks of age with pathogenic MS WVU 1853 strain

Conclusion

- Significant difference shown between vaccinates and non-vaccinates
- Non-vaccinated, challenged group egg production dropped from ±90% to ±60%

EGG PRODUCTION PER WEEK



*Data on file, MSD Animal Health

***KEY TO SCORING SYSTEM**

Air sac scoring system:

Macroscopic evaluation score 0-4 according method. Yoder et al: *Avian diseases 1984 28, 224-234.*

- Score 0: normal air sacs, sparkling clear and thin.
- Score 1: only cloudiness or gray areas with slight thickening or flecks of yellowish exudate, involving a limited area of one or two air sacs.
- Score 2: readily visible grayish to yellow exudate, sometimes foamy with thickening of the air sac, involving one or portions of two air sacs.
- Score 3: somewhat more severe exudative, thickened airsacculitis, but mainly more extensive, involving essentially three air sac regions.
- Score 4: severe airsacculitis with considerable exudate and thickening of almost all air sacs.

Ovary lesion score system:

Macroscopic evaluation score 0-4.

- Score 0: normal ovary with equal to or more than 6 mature follicles.
- Score 1: the number of mature follicles is slightly less than of a normal ovary. 4-5 mature follicles are observed.
- Score 2: the number of mature follicles is less than that of a normal ovary. 2-3 mature follicles are observed.
- Score 3: the number of mature follicles is apparently less than that of a normal ovary. 1-2 mature follicles are observed.
- Score 4: Ovarian atrophy is observed while mature follicles are not observed.

Efficacy of Nobilis® MS Live & Nobilis® MG 6/85 combination

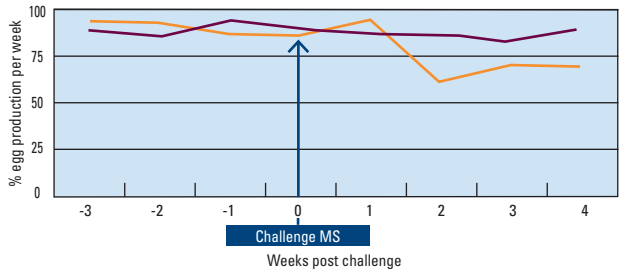
COMBINE NOBILIS® MS LIVE AND NOBILIS® MG 6/85 FOR COMPLETE, COMPREHENSIVE MYCOPLASMA PROTECTION

- Combined Nobilis® MS Live and Nobilis® MG 6/85 at 6 weeks of age
- Challenge at 50 weeks of age
- Challenge strains: MS WVU 1853 or Pathogenic MG-R

EFFICACY NOBILIS® MS LIVE & NOBILIS® MG 6/85 COMBINATION

Group	Vaccination: 6 weeks of age	Challenge: 50 weeks of age
		Route
Vaccine combination	MS Live & MG 6/85	Intra air sac
Non-vac - Challenge	-	Intra air sac

MS CHALLENGE WVU 1853

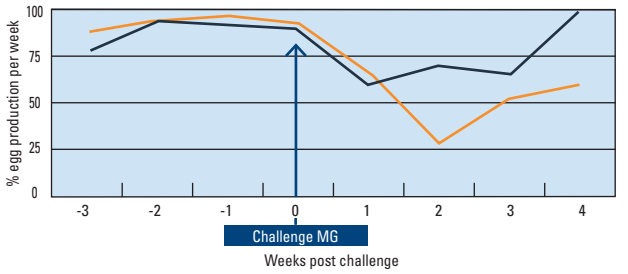


*Data on file, MSD Animal Health

Conclusion

Significant difference between vaccinated and non-vaccinated (X^2 test $P<0.05$). Non-vaccinated challenged group dropped from ±90% → ±70%.

MG CHALLENGE MG-R



*Data on file, MSD Animal Health

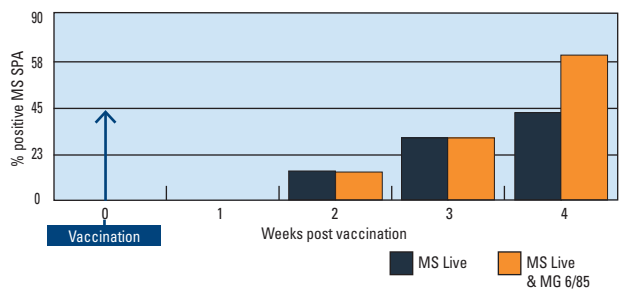
Conclusion

Significant difference between vaccinated and non-vaccinated (X^2 test $P<0.05$). Non-vaccinated challenged group dropped from ±95% → ±29%.

SEROLOGIC MONITORING POST VACCINATION

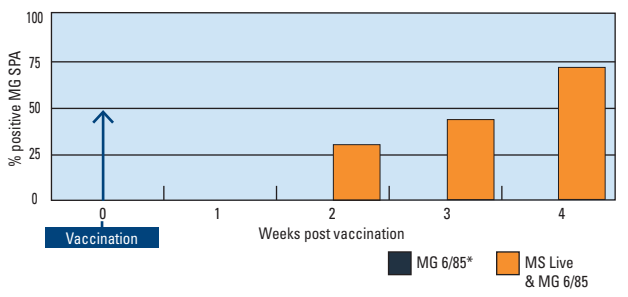
SPA MS and SPA MG monitoring in Nobilis® MS Live and Nobilis® MS Live & Nobilis® MG 6/85 vaccinated birds that are vaccinated at 6 weeks of age.

MS SPA RESPONSE



*Data on file, MSD Animal Health

MG SPA RESPONSE



*MG 6/85 did not induce a positive SPA up till 4 weeks post vaccination

*Data on file, MSD Animal Health

SUMMARY OF PRODUCT CHARACTERISTICS

1. NAME OF THE VETERINARY MEDICINAL PRODUCT

Nobilis® MS Live, lyophilisate for suspension for chickens.
ES: Nobilis® MS Viva.

2. QUALITATIVE AND QUANTITATIVE COMPOSITION

Per dose of reconstituted vaccine:
Active substance:
Live attenuated *M. synoviae* strain MS1: 106.5 and 108.0 CFU1.
[1]Colony Forming Units.
For a full list of excipients, see section 6.1.

3. PHARMACEUTICAL FORM

Lyophilisate for suspension.
Off-white to yellowish-coloured pellet.

4. CLINICAL PARTICULARS

4.1 Target species
Chickens (layers).
4.2 Indications for use
For the active immunisation of chickens (layers) from 6 weeks of age to reduce air sac lesions, ovary lesions, and a drop in egg production due to infection caused by *M. synoviae*. Onset of immunity: 4 weeks Duration of immunity: 44 weeks.
4.3 Contraindications
None.
4.4 Special warnings
Do not use antibiotics or other substances with any antimicrobial activity known to inhibit *M. synoviae*.
4.5 Special precautions for use
Special precautions for use in animals
Vaccinate healthy chickens only. It is not recommended to vaccinate in the presence of (sub-) clinical infection with *M. synoviae*.
The vaccine strain has been detected in the respiratory tract of vaccinated chickens by PCR at 34 weeks after vaccination. Taking into account the potential spread of the vaccine strain by direct or indirect transmission, all chickens in the chicken house should be vaccinated. Adequate biosecurity measures should be in place, such as change of clothing and boots and the use of properly disinfected equipment.
After vaccination interference with serological screening methods for *Mycoplasma* infections may occur, but the vaccine strain can be differentiated from wildtype *M. synoviae* by PCR or by culture in *Mycoplasma* growth medium containing nicotinamide instead of NAD.
Special precautions to be taken by the person administering the veterinary medicinal product to animals
To avoid skin and eye injuries as well as inhalation or digestion, personal protective equipment consisting of a mask, gloves and eye protection should be worn when handling the veterinary medicinal product. Wash and disinfect hands after vaccinating.
4.6 Adverse reactions (frequency and seriousness)
None.
4.7 Use during pregnancy, lactation or lay
Can be used during lay.
4.8. Interaction with other medicinal products and other forms of interaction
Safety and efficacy data are available which demonstrate that this vaccine can be mixed and administered with Nobilis MG 6/85 (in member states where this product is authorized). The product literature of Nobilis MG 6/85 should be consulted before administration of the mixed product. The mixed product is not to be used within four weeks of onset of egg production or during lay. The Nobilis MS Live vaccine strain may spread from vaccinated to unvaccinated chickens in case it is used mixed with Nobilis MG 6/85. The adverse effects observed after administration of one dose or an overdose of the mixed vaccines are not different from those described for Nobilis MS Live alone. When mixed with Nobilis MG 6/85, the demonstrated efficacy claims are not different from those described for Nobilis MS Live alone.
No information is available on the safety and efficacy of this vaccine when used with any other veterinary medicinal product except the product mentioned above. A decision to use this vaccine before or after any other veterinary medicinal product therefore needs to be made on a case by case basis.
4.9. Amounts to be administered and administration route
After reconstitution, administer 1 dose of vaccine by nebulization (fine-spray) to chickens (layers) from 6 weeks of age.
Use the entire contents when first opened.
Preparation of vaccine

- Use only clean, cool, non-chlorinated, preferably distilled water of 25 °C. The volume of water for reconstitution should be sufficient to ensure an even distribution when sprayed onto the birds. This will vary according to the size of the birds being vaccinated and the management system, but 250 to 400 ml of water per 1000 doses is recommended. Follow the instructions of the fine-spraying device.
- Open the vial submerged under water.
- Remove the seal and stopper from the vial.
- In case of mixed-use, repeat steps 2 and 3 in the same water using a vial of Nobilis MG 6/85 containing the same number of doses.

Administration

- Vaccinate with a fine-spraying device suitable for nebulization application of vaccines (particle size: < 100 µm). The vaccine suspension should be spread evenly over the correct number of birds, at a distance of approximately 40 cm.
- Do not use any disinfectants, skimmed milk or other agents impairing the performance of the vaccine in the fine-spraying device.
- Shut off all fans and close air-inlets while fine-spray vaccinating.
- Clean the fine-spraying device thoroughly after use according to the manufacturer's recommendation.

4.10 Overdose (symptoms, emergency procedures, antidotes), if necessary
None.
4.11 Withdrawal period(s)
Zero days.

5. IMMUNOLOGICAL PROPERTIES

Pharmacotherapeutic group: live bacterial vaccines for poultry
ATC vet code: QI01AE03
To stimulate active immunity in chicken against *M. synoviae*.

6. PHARMACEUTICAL PARTICULARS

6.1 List of excipients
Sodium dihydrogen phosphate dihydrate
Disodium hydrogen phosphate dihydrate
Glutamine
Sodium chloride
Sucrose
Pancreatic digest of casein
Lactalbumin hydrolysate
Gelatin
6.2 Incompatibilities
Do not mix with any other veterinary medicinal product, except Nobilis MG 6/85 or the solvent recommended for use with the veterinary medicinal product.
6.3 Shelf life
Shelf-life of the veterinary medicinal product as packaged for sale: 2 years.
Shelf-life after dilution or reconstitution according to directions: 2 hours.
6.4. Special precautions for storage
Store in a refrigerator (2 °C – 8 °C).
Do not freeze.
Protect from light.
6.5 Nature and composition of immediate packaging
Glass vial of hydrolytical class type I containing 500, 1000 or 2000 doses of lyophilisate. The vial is closed with a halogenobutyl rubber stopper and sealed with an aluminium cap.
Package sizes:
Cardboard box with 1 vial of 500 doses of lyophilisate.
Cardboard box with 1 vial of 1000 doses of lyophilisate.
Cardboard box with 1 vial of 2000 doses of lyophilisate.
Cardboard box with 10 vials of 500 doses of lyophilisate.
Cardboard box with 10 vials of 1000 doses of lyophilisate.
Cardboard box with 10 vials of 2000 doses of lyophilisate.
Not all pack sizes may be marketed.
6.6 Special precautions for the disposal of unused veterinary medicinal product or waste materials derived from the use of such products
Any unused veterinary medicinal product or waste materials derived from such veterinary medicinal products should be disposed of in accordance with local requirements.

7. MARKETING AUTHORISATION HOLDER

Intervet International BV
Wim de Körverstraat 35
5831 AN Boxmeer
The Netherlands
As represented by the national companies in the member states.

References

1. Stanly H Kleven & Naola Ferguson-Noel. *Diseases of Poultry 12th edition: Chapter 21 Mycoplasmosis. Mycoplasma synoviae infection.*
2. Landman et al. Avian Pathology; *AP 30 2001 629-639: Field studies on the association between amyloid arthropathy and Mycoplamsa synoviae infection and experimental reproduction of the condition in brown layers.*
3 Feberwee et al. Avian Pathology; *AP 37 2008 629-633: Seroprevalence of Mycoplasma synoviae in Dutch commercial poultry farms.*
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5. Feberwee et al. *Avian Pathology; AP 39 2010: 133-137: Induction of eggshell apex abnormalities in broiler breeder hens.*
6. Dijkman et al. *Avian pathology: AP 43 2014: 465-472: Variable lipoprotein haemagglutinin (vlhA) gene sequence typing of mainly Dutch Mycoplasma synoviae isolates: comparison with vlhA sequences from Genbank and with amplified fragment length polymorphism analysis.*
7. Vardaman et al. *Poultry Science 52 : 1973 Effect of Mycoplasma synoviae on Broiler Performance.*
8. Remco Dijkman. *GD report 8074074: Validation of Mycoplasma synoviae PCR developed by MSD Animal Health to differentiate between Mycoplasma synoviae vaccine and field strains.*

Data on file from registration dossier.

For more information contact your local MSD Animal Health representative.

MSD Animal Health - Optimizing vaccine performance through partnership & support.

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