Keeping first class eggs flying high.
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).<sup>1</sup>
- After initial infection MS may become systemic inducing infectious synovitis and/or drop in egg production and egg quality.<sup>1</sup>

**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<sup>2</sup>**

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

**MS - THE HIDDEN DANGER**

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

**MS causes the following issues:**

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

**ECONOMIC IMPACT**<sup>7</sup>

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.**

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
- PCR⁶
- 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 birds⁸
- Sequencing for epidemiological follow up

### Mock Table

<table>
<thead>
<tr>
<th></th>
<th>1 wk PV</th>
<th>2 wk PV</th>
<th>3 wk PV</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Vaccine</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MG+MS</td>
<td>0</td>
<td>29</td>
<td>86</td>
</tr>
<tr>
<td><strong>Control</strong></td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

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 $\geq 10^{6.5}$ and $\leq 10^{8.0}$ 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**

<table>
<thead>
<tr>
<th>Group</th>
<th>Strain</th>
<th>Dosage</th>
<th>Group size</th>
<th>Days after administration</th>
<th>14</th>
<th>28</th>
<th>42</th>
<th>56</th>
<th>70</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>MS1</td>
<td>$10^{7.2}$</td>
<td>7</td>
<td>3/6 50% 5/6 83.3% 5/5 100% 5/6 100% 4/7 57.1%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Commingled with group 1</td>
<td>-</td>
<td>7</td>
<td>2/7 28.6% 7/7 100% 5/5 100% 4/6 66.7% 4/5 80%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>non-vaccinated</td>
<td>-</td>
<td>7</td>
<td>nt nt nt nt nt nt</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Data on file, MSD Animal Health

**Safety Conclusions**

**Spread**
- In case it can spread to in-contact birds without inducing clinical signs
- Recovery of vaccine strain only from infraorbital 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

<table>
<thead>
<tr>
<th>Examined organ</th>
<th>CFU/chicken</th>
<th>MS challenge</th>
<th>CFU/chicken intra air sac</th>
<th>Air sac&lt;sup&gt;1&lt;/sup&gt; score</th>
<th>Ovary&lt;sup&gt;2&lt;/sup&gt; lesion score</th>
</tr>
</thead>
<tbody>
<tr>
<td>MS Live</td>
<td>10&lt;sup&gt;7&lt;/sup&gt;</td>
<td>WVU 1853</td>
<td>0.2 ± 0.42</td>
<td>0.1 ± 0.32</td>
<td></td>
</tr>
<tr>
<td>Non vac. challenge control</td>
<td>-</td>
<td>WVU 1853</td>
<td>1.4 ± 1.26</td>
<td>0.9 ± 1.20</td>
<td></td>
</tr>
</tbody>
</table>

*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%

*Data on file, MSD Animal Health

Air sac scoring system:
- 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.
- Score 2: the number of mature follicles is less than that of a normal ovary.
- Score 3: the number of mature follicles is apparently less than that of a normal ovary.
- 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

### Conclusion

Significant difference between vaccinated and non-vaccinated (X² test P<0.05). Non-vaccinated challenged group dropped from ±90% → ±70%.

## 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.

### Conclusion

Significant difference between vaccinated and non-vaccinated (X² test P<0.05). Non-vaccinated challenged group dropped from ±95% → ±29%.
SUMMARY OF PRODUCT CHARACTERISTICS

1. NAME OF THE VETERINARY MEDICINAL PRODUCT
Nobis® MS Live, lyophilisate for suspension for chickens. E5: Nobis® MS Live.

2. QUALITATIVE AND QUANTITATIVE COMPOSITION
Per dose of reconstituted vaccine.
Active substance:
Live attenuated M. synoviae strain MS/105.6 and MS/108.0 C/FU.
SafetyHandling 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 24 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 ingestion, 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 vaccination.

4.6 Adverse reactions (frequency and seriousness)
None.

4.7 Use during pregnancy, lactation or by 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 or the solvent recommended for use with the veterinary medicinal product. M. synoviae 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 ingestion, 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 vaccination.

4.9. Amounts to be administered and administration route

4.9.1 Dose of vaccine

4.9.1.2 For a full list of excipients, see section 6.1.

4.9.1.3 Use 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.

4.9.1.4 Shut off all fans and close air inlets while fine-spray vaccinating.

4.9.4.1.5 Clean the fine-spraying device thoroughly after use according to the manufacturer’s recommendation.

4.9.1.6 Reconstitute according to directions. Follow the instructions of the fine-spraying device.

4.10 Overdose (symptoms, emergency procedures, antidotes), if necessary
• 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.

4.10.1 Following symptoms are possible:
• Respiratory tract infection in vaccinated chickens.
• Signs of respiratory tract irritation, such as coughing, sneezing, or nose-bleeding.

4.11 Withdrawal period(s)
Zero days.

5. PHARMACEUTICAL PROPERTIES
Pharmacotherapeutic group: live bacterial vaccines for poultry
ATC vet code: QI01AE03To stimulate active immunity in chicken against M. synoviae.

6. PHARMACEUTICAL PARTICULARS
6.1 List of excipients
Sodium hydrogen phosphate dihydrate
Disodium hydrogen phosphate dihydrate
Mannitol
Sodium chloride
Sucrose
Pancaristic digest of casein
Lactobacillus hydrogenarum
Sodium bicarbonate
6.2 Incompatibilities
Do not mix with any other veterinary medicinal product, except Nobilis MS Live or the solvent recommended for use with the veterinary medicinal product.

6.3 Shelf life
Shelf life of the veterinary medicinal product as packed for sale is 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).

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:

1. Glass vial of hydrolytical class type I containing 500 doses of lyophilisate.
2. Glass vial of hydrolytical class type I containing 1000 doses of lyophilisate.
4. Glass vial of hydrolytical class type I containing 5000 doses of lyophilisate.
5. Glass vial of hydrolytical class type I containing 10000 doses of lyophilisate.

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.

6.7.1 Marketing Authorisation holder
Intervet International BV
Wim de Körverstraat 35
5035 AR Uden
The Netherlands

References
2. Landman et al. Avian Pathology; AP 30 2001 629-638: Field studies on the association between amyloid arthropathy and Mycoplasma synoviae infection and experimental reproduction of the condition in brown layers.
5. Dijkman et al. Avian pathol; AP 43 2014 465-472: Variable lipoprotein haemagglutinin (vliA) gene sequence typing of mainly Dutch Mycoplasma synoviae isolates; comparison with vliA sequences from Genbank and with amplified fragment length polymorphism analysis.
7. Remco Dijkman. GD report 8074074: Validation of Mycoplasma synoviae PCR developed by MSD Animal Health to differentiate between Mycoplasma synoviae vaccine and field strains.

For more information contact your local MSD Animal Health representative.