

**VetLine**  
**Riemerella ELISA**  
**(RIEMVT0880)**

Performance Characteristics

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## 1 Introduction

The pathogen of infectious serositis (riemerellosis) is a 0.2-0.5 x 1.0-2.5 µm large, immobile, gram-negative rod-shaped bacterium of the genus *Riemerella*. *Riemerella* can be found worldwide and it is particularly important in the breeding of ducks and geese, since it leads to considerable economic losses. In addition to ducks (main host) also geese and turkeys, and more rarely chicken and wild birds are infected. Infection sources are latently infected adult animals. Here the pathogens persist in the mucous membranes of the upper respiratory tract. Infection occurs from animal to animal or indirectly by drinking water, interspersing etc. The acute disease usually affects young animals and is often fatal.

Symptoms of *riemerellosis* are:

- Catarrhal to fibrinous rhinitis, conjunctivitis and sinusitis
- Central nervous system disorders ("crooked")
- Lameness
- Heartbeat- and liver infections
- Catarrhal Enteritis
- Greenish diarrhea
- In the chronic course, brain inflammation and joint inflammation occur

The infection can be diagnosed by the following methods:

- Cultivation of *riemerella* in the embryonated egg or in cell culture
- Serologically by ELISA of *Riemerella* in the embryonated egg or in cell culture
- Serologically by ELISA

## 2 Intended Use

The NovaTec VetLine *Riemerella* ELISA is intended for the qualitative determination of antibodies against *Riemerella* in veterinary serum.

## 3 Principle of the Assay

The qualitative immunoenzymatic determination of specific antibodies is based on the ELISA (Enzyme-linked Immunosorbent Assay) technique.

Microplates are coated with specific antigens to bind corresponding antibodies of the sample. After washing the wells to remove all unbound sample material a horseradish peroxidase (HRP) labelled conjugate is added. This conjugate binds to the captured antibodies. In a second washing step unbound conjugate is removed. The immune complex formed by the bound conjugate is visualized by adding Tetramethylbenzidine (TMB) substrate which gives a blue reaction product.

The intensity of this product is proportional to the amount of specific antibodies in the sample. Sulphuric acid is added to stop the reaction. This produces a yellow endpoint colour. Absorbance at 450/620 nm is read using an ELISA microwell plate reader.

## 4 Performance Characteristics

### 4.1 Reproducibility (Precision)

#### Test Description

The reproducibility of the NovaTec VetLine Riemerella ELISA kit was determined by comparing 12-24 replicates of at least 2 different samples in one assay (within-run) and by comparing at least 2 different samples assayed in 12 different runs (between-run).

In accordance with the procedures prescribed in the instruction, the mean ( $\bar{X}$ ) and the standard deviation (s) of the absorbance values (E) were determined.

The CV value [%] was then calculated using the following formula:

$$CV = s/\bar{x} \times 100 \%$$

Acceptance Criterion: CV < 15 %

#### Results

Table 1: Within-Run Precision

Sample	n	Mean (E)	CV [%]
1	24	1,705	1,50
2	24	0,047	8,08
3	24	0,996	2,76

Table 2: Between-Run Precision

Sample	n	Mean (NTU)	CV [%]
1	12	41,56	3,96
2	12	2,07	4,09
3	12	2,05	3,39
4	12	15,58	2,45
5	12	15,47	2,55

#### Conclusion

The acceptance criterion was met for all samples.

## 4.2 Diagnostic Sensitivity and Specificity

### Introduction

To evaluate the diagnostic performance of the VetLine Riemerella ELISA, internal studies were conducted by NovaTec in comparison to pre-defined turkey samples.

### Materials

VetLine Riemerella ELISA  
Production date: 2017-03

Lot: RIEMVT-002  
Expiry date: 2018-03-31

10 positive turkey samples

16 negative turkey samples

### Results

Total number of turkey samples: 26

Table 3: Diagnostic Sensitivity and Specificity

	Demand		$\Sigma$
	positive	negative	
VetLine Riemerella ELISA	positive	10	10
	negative	0	16
$\Sigma$	10	16	26

Diagnostic Sensitivity turkey: 100 % (95% confidence interval: 69,15 % - 100,0 %)

Diagnostic Specificity turkey: 100 % (95% confidence interval: 79,41 % - 100,0 %)

Agreement: 100 % (16/16)

### Conclusion

The diagnostic sensitivity of turkey samples was 100 % and the diagnostic specificity of turkey samples was 100 % (agreement: 100 %).