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MYCOPLASMA BOVIS ELISA KIT

BIO K 302/2 - BIO K 302/5

For serum or milk

Mycoplasma bovis is associated with many cattle diseases, including arthritis, pneumonia in calves and young stock, mastitis, and genital infections. The infectious pneumonias that affect intensively-raised calves are responsible for sizable economic losses due to the mortality, treatment costs, and growth delays that they cause. These respiratory infections often involve multiple factors and are caused by interactions among viruses, mycoplasmas, and bacteria. Several species of Mycoplasma have been isolated from the respiratory tracts of calves. Some of them are most probably simple commensals or opportunistic species that merely worsen the lung damage caused by other agents. Mycoplasma bovis has been isolated from the lungs of calves with pneumonia. It is probably the most pathogenic species affecting the Bovidae after Mycoplasma mycoides mycoides. *Mycoplasma bovis* can induce the development of pneumonia in gnotobiotic calves. *Mycoplasma bovis* is frequently found in association with *Mannheimia haemolytica* in pneumonia in calves.

Use of the kit

The kit is designed to follow on paired sera or milks

Reliable Results

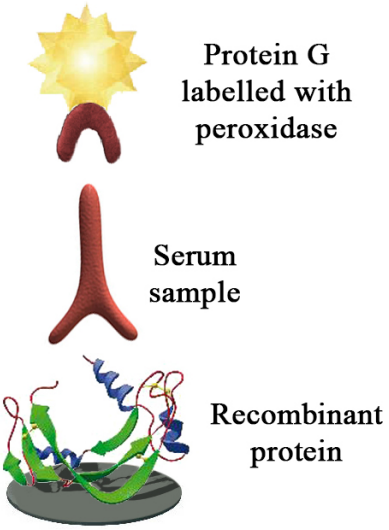
The use of monoclonal antibody as conjugate ensures excellent specificity and very reliable results.
The use of recombinant protein on the plate also makes it possible to obtain an excellent specificity

Ease-of-Use

Minimal hands-on-time
Room temperature incubation
Results available in 140 minutes for single or batch testing

EIA Procedure

1- Microplate coated with recombinant protein.
2- Add samples and positive control.
Incubate 1 hour at 21°C+/-3°C.
Wash
3- Add conjugate.
Incubate 1 hour at 21°C+/-3°C.
Wash
4- Add chromogen (TMB).
Wait 10 minutes
Add stop solution. Read at 450 nm



Bio-X Diagnostics - 38, Rue de la Calestienne (PAE) - 5580 Rochefort - Belgique
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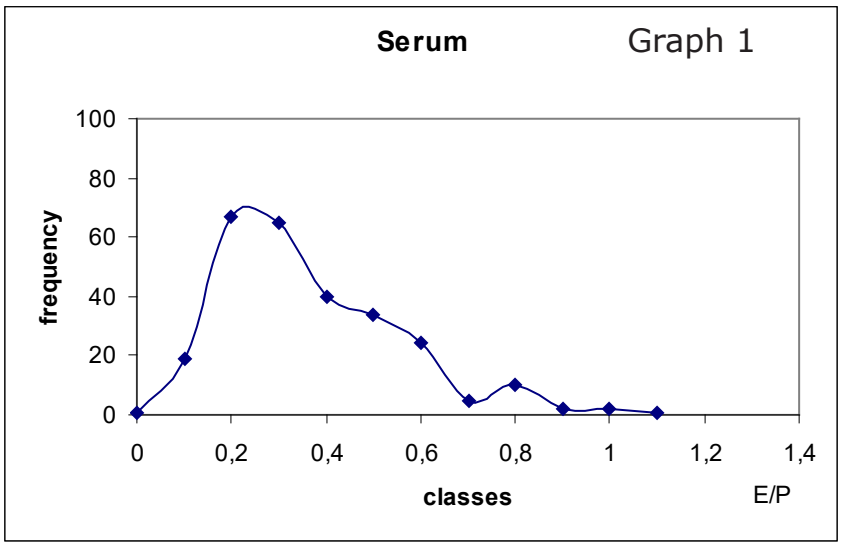
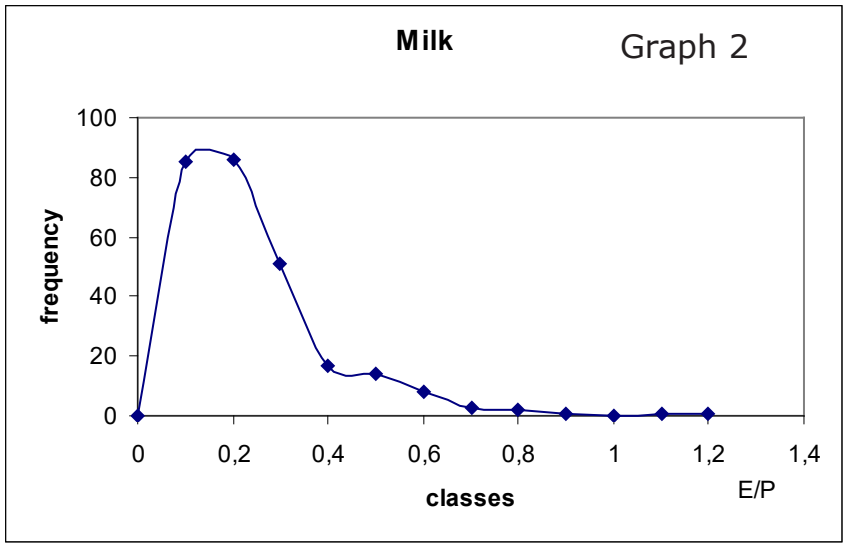
Example of results

270 serum and 270 milk samples taken from the same animals were tested using the BIO K 302 kit. These samples came from twenty-seven Belgian farms. Their optical density readings were divided by the optimal density reading for the kit's reference serum (E/P). Frequency histograms were then plotted for the blood sera (Graph 1) and milk samples (Graph 2).





These two graphs show that Belgian cows have a non-negligible residual level of specific antibody against Mycoplasma bovis. This is not at all surprising, given that the bacterium is a normal commensal in cattle's nasal passages. This can explain the presence of a population of animals for which the E/P ratio ranges from 0 to 0.7 Cows with an E/P ratio above 0.7 could doubtless be considered to belong to a population of animals that had mycoplasmal pneumonia. It is worthwhile noticing that the two histograms' profiles are fairly similar, even though they refer to two different groups of samples, i.e., blood sera and milk.

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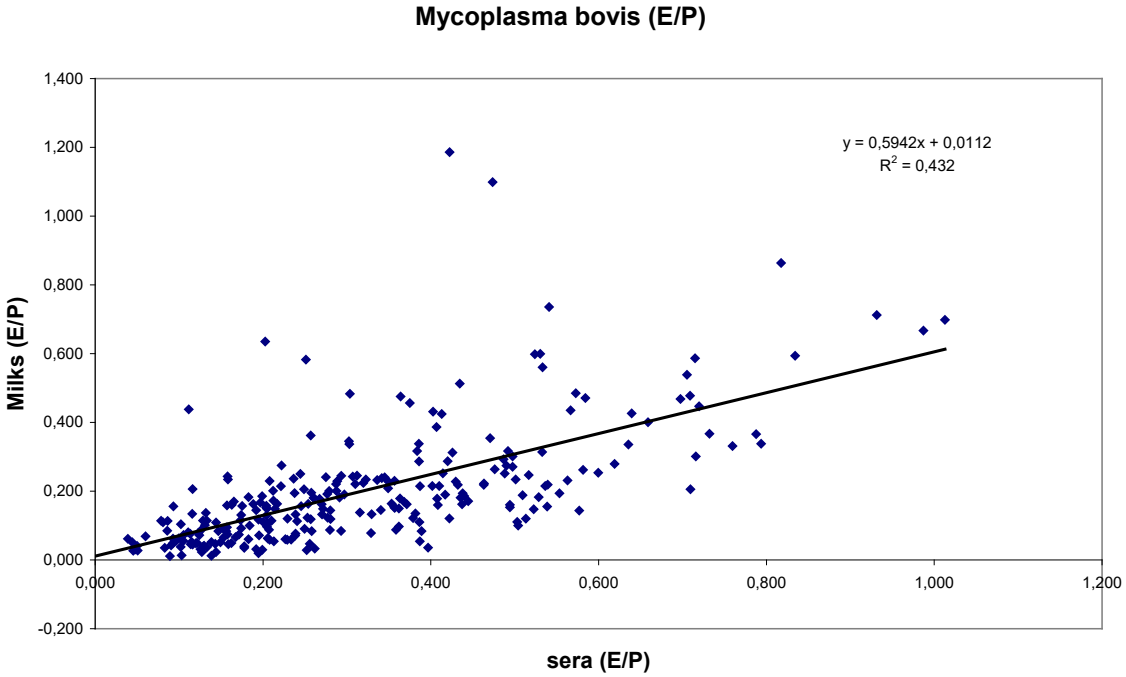
Correlation between sera and milks



Example of results

Five cows were inoculated experimentally with a *Mycoplasma bovis* culture. Serum samples were then taken from these animals at regular intervals and tested using the BIO K 302 kit. At the end of the trial the animals were sacrificed and their lungs removed to be tested for the bacterium's presence.

	Day of experimental infection										
		Infect									
Days	-3	0	3	5	7	10	14	17	21	28	35
Animal 1	+	+	+	0	0	+	+	+	+++		
Animal 2	0	0	0	0	0	0	0	0	0		
Animal 3	+	+	0	+	+	++	++				
Animal 4	+	+	+	+	+	+++	++	+++	+++	+++	+++
Animal 5	0	0	0	+	+	+	++	+++	+++	+++	+++

Mycoplasma bovis was isolated from the lungs of four of the five artificially infected animals. The bacterium was not isolated from the lungs of Subject 2. It is worthwhile noting that this subject was the only one that did not show seroconversion following the infection. The antibody's presence, at low levels, in Subjects 1, 3, and 4 prior to infection was doubtless due to the presence of M. bovis in their nasal cavities. However, this preliminary detection test was not performed.

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Echantillon	OD450	S-P/P-N*100	Interpreta-tion
1	0,297	9,38	-
2	0,488	19,28	-
3	0,106	-0,52	-
4	0,186	3,63	-
5	0,283	8,66	-
6	0,120	0,21	-
7	0,347	11,98	-
8	0,285	8,76	-
9	0,158	2,18	-
10	0,262	7,57	-
11	0,236	6,22	-
12	0,299	9,49	-
13	0,331	11,15	-
14	0,383	13,84	-
15	0,239	6,38	-
16	0,293	9,18	-
17	2,847	141,58	+
18	2,871	142,82	+
19	2,217	108,92	+
20	2,498	123,48	+
21	2,029	99,17	+
22	2,000	97,67	+
23	0,843	37,69	+
24	2,556	126,49	+
25	2,030	99,22	+
26	1,465	69,93	+
27	2,447	120,84	+
28	2,665	132,14	+
29	2,502	123,69	+
30	2,542	125,76	+

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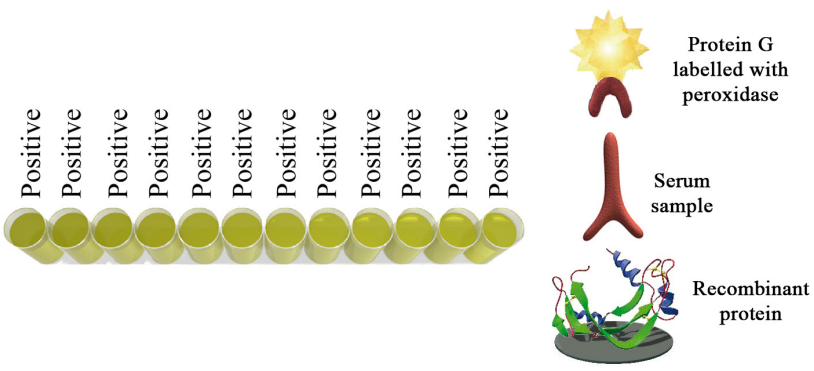
BIO K 302	Experimentally infected calves: +	Negative samples: -
+	14	0
-	0	16

National Contagious Bovine Pleuropneumonia Designated Detection Laboratory, State Key Laboratory of Veterinary Biotechnology, Division of Bacterial Diseases, Harbin Veterinary Research Institute, Chinese Academy of Agricultural Sciences, Harbin 150001, China

PS2 international standardpositive serum of contagious bovine pleu-ro-pneumonia: Negative

Composition of the kit

M. bovis ELISA KIT	BIO K 302/2	BIO K 302/5
Microplate	2 (192 tests)	5 (480 tests)
Washing solution	1 X 100 ml (20 X)	1 X 250 ml (20 X)
Dilution buffer	1 X 50 ml (5 X)	2 X 100 ml (5 X)
Conjugate	1 X 0,5 ml (50 X)	1 X 1,4 ml (50 X)
Positive serum	1 X 0.5 ml (1 X)	1 X 0.5 ml (1 X)
Negative serum	1 X 0.5 ml (1 X)	1 X 0.5 ml (1 X)
Single component TMB	1 X 25 ml (1 X)	1 X 55 ml (1 X)
Stopping solution	1 X 15 ml (1 X)	1 X 30 ml (1 X)



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