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Product Information (11.12.07)

Name of Kit: **ImmunoComb[®] Canine Brucella Antibody Test Kit**

Catalog No: 50CBR101/ 50CBR110

No of Tests: 12 (Standard Kit)/ 120 (Lab Kit)

Intended Use: The ImmunoComb[®] Canine Brucella Antibody Test Kit is intended to screen for previous exposure to *Brucella canis*.

Diagnostic Method: The ImmunoComb[®] test is based on solid phase "dot"-ELISA technology. Antigen is applied to test 'spots' on the solid phase, which is a comb-shaped plastic card. (The Comb has 12 teeth-sufficient for 12 test samples.)

The samples to be tested are mixed with diluent in the first row of wells of a multi-compartment developing plate. The test spots on the Comb are then incubated with the samples in the developing plate. Specific IgG antibodies from the samples, if present, bind to the antigen at the test spots.

The Comb is then transferred to a well, where unbound antibodies are washed from the antigen spots. In the next step, the Comb is allowed to react with an anti-dog IgG Alkaline Phosphates conjugate, which will bind to antigen-antibody complexes at the test spots. After 2 more washes, the Comb is moved to the last well, where a color result develops via an enzymatic reaction. Significant levels of IgG anti-*B. canis* antibodies are produced by dogs following infection. The intensity of the color result of test spots corresponds directly to the antibody level in the test sample.

Specificity: 93%

Sensitivity: 98%

Pathophysiology

Transmission: *B. canis* is sexually transmitted by the mating of infected males and females. *Brucella canis* in the female dog will live in the vaginal and uterine tissue and secretions for years, usually for life. In males, the Brucella bacteria live in the testicles and seminal fluids. An infected male is just as contagious as the female since he can spread the Brucella bacteria via urine or semen.

Transmission between dogs occurs via mucous membranes, so the bacteria may enter the body through the nose, mouth, conjunctiva of the eye and vagina. The majority of bacteria in infected dogs are secreted in semen and vaginal secretions. Bacteria may also be present in milk, urine and saliva, thus any body fluids can infect another dog.

When an infected bitch aborts, spread throughout a kennel can be very rapid. Infected bitches may deliver both living and dead puppies. These surviving puppies are infected and will shed bacteria in their secretions.

Clinical signs: Usually clinical signs are absent in a non-gravid bitch, and are hard to be noticed during a physical examination. *Brucella canis* affects the reproductive system both in female and male dogs, and is characterized by reproductive disturbances.

Litters are commonly aborted, usually in the last two weeks of gestation, or the puppies may die shortly after birth. A bitch that aborts after 45 days of gestation should be highly suspected of Brucellosis. Usually the fetuses are partially decayed and accompanied by a gray to green vaginal discharge. This discharge may contain very high numbers of the bacteria. If embryos die early, they may be reabsorbed and the female may never appear to be pregnant at all. In males, there are often no signs, except for advanced cases when the testicles may be uneven in size. Please refer to table 1.

Table 1. Major Clinical signs

Gender	Clinical signs
Both	Mild or no signs. Usually no fever.
Female	Lymphadenopathy or no signs. Fertility problem (conceiving or keeping).
Pregnant female	Abortion of dead litter in late gestation stage. Brown or greenish-gray vaginal discharge.
Male	Lymphadenopathy, epididymitis, testicular abnormalities, infertility, enlarged scrotum.
Surviving pups	General lymphadenopathy, persistent globulinemia, transient fever, leukocytosis, seizures.

Since many infected dogs have no clinical signs, and since *B.canis* bacteria are very persistent and rapidly transmitted, it is highly recommended to screen all dogs as a routine procedure. This is mostly important in breeding kennels and wherever dogs are kept together.

Interpretation: The level of antibodies (i.e., antibody titer) is determined according to the intensity of the test color result. Thus, no color or a light

gray color indicates no (negative) or low level of antibodies. Higher levels of antibodies are indicated by darker color results. For the ImmunoComb[®] Canine Brucella Antibody Test Kit a positive reference spot on each Comb tooth (top spot) has been calibrated to develop a distinct gray color. This is the same color that is generated by a significant positive result.

A color result that is equal to or darker than the positive reference spot is considered positive. A colorless (white) or faint color result, which is lighter than the positive reference, is considered negative. In some cases, a faint color result may be interpreted as "suspicious". Please refer to Table 2.

Table 2. Interpretation of Results

IC* Score	Result	Clinical Situation	Interpretation	Recommendations
0	Neg.	Well Dog	Undetectable levels of IgG antibodies to <i>B. canis</i> .	Recheck before next breeding
		Suspected Dog		Retest in 7-10 days.
1-2	Low Pos.	Well Dog	Insignificant levels of IgG antibodies to <i>B. canis</i> . Results may be non-specific.	Recheck before next breeding
		Suspected Dog		Retest in 7-10 days.
3	Med. Pos.	Well Dog	Positive IgG titer. Confirms exposure to <i>B. canis</i> .	Perform confirmatory <i>B. canis</i> isolation.
		Suspected Dog		Isolate dog, until a decision regarding the dog fate is done.
≥4	High Pos.	Well Dog	Significant IgG titer. Confirms exposure to <i>B. canis</i> .	Perform confirmatory <i>B. canis</i> isolation. Isolate dog, until a decision is accepted.
		Suspected Dog		

* IC = ImmunoComb[®]

Main Application: Screening for previous exposure to *Brucella canis*. Negative results confirm the absence of antibodies.

Preferred Method of Diagnosis: The diagnosis of canine Brucellosis is largely based on serology. As most serological methods had been proven to have false positive results, it is recommended to confirm infection by either *B. canis* culture or PCR. Until a confirmation is done, and decision is accepted with the aid of the vet, it is recommended to keep the dog isolated, and take precautions while handling it.

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For more information:

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