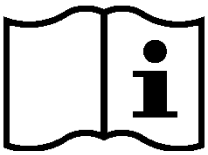
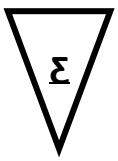


Useŕs Manual

Canine Rheumatoid Factor One-Step

For the detection of all types of rheumatoid factors (IgG, IgA, IgM) in serum or plasma samples of dogs

REF D1010-AB01



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January 2022

Please use only the valid version of the package insert provided with the kit.

1. Table of Contents

2. Introduction

Rheumatoid factor (RF) antibodies are macroglobulines which appear in the serum of dogs suffering from Rheumatoid Arthritis (RA). This is frequently characterised by chronic and progressive multi-site lameness, joint swelling and joint destruction. These macroglobulines can be oft the IgM, IgA or IgG subclass (mainly the IgM class). The majority of these RF antibodies are directed against the Fc part of IgG (3) raised levels of antibodies can be found in most patients with RA (70%), but also in the other connective tissue diseases, malignancy, chronic infection and even small percentage (<2%) in the normal population.
It is not known why patients with Rheumatoid Arthritis produce increased amount of Rheumatoid factor, but Rheumatoid factor complexes are thought to have a role in the propagation of the Rheumatoid arthritis by intra articular activation of various inflammatory factors mechanism. This can lead to inflammation with serious destructive changes of different joints caused by lysomal destruction.
IgA Rheumatoid factor was found, to be significantly associated with later development of erosive bone disease; IgA and IgG Rheumatoid factor levels increased precede clinical manifestations. The IgA and IgG Rheumatoid factor levels also correlate better with the erythrocyte sedimentation rate (ESR) and elevated levels of C-reactive protein. IgG Rheumatoid factor is important due to the property of self-association leading to production of complexes without bacterial/viral antigenic stimulus.

Diagnostic criteria for Rheumatoid arthritis according to the American Rheumatoid Association:

- Morning stiffness
- Pain on motion (at least one point)
- Swelling (fluid/bone)
- Symmetric joint swelling
- Subcutaneous nodules over bony prominences
- X-ray changes typical for Rheumatoid arthritis
- Positive RF test, by a method which has been positive (<5%) of normal controls
- Pour mucin precipitate from synovial fluid
- Characteristic histologic changes in synovial membrane
- Characteristic histologic changes in nodules showing granulomatous foci

3. Intended use of the test kit

This One-Step test is intended to use as practical/routine screening test that can be done in a few minutes. This test kit is designed to detect antibodies against Rheumatoid factors (IgA, IgM and IgG) by use of a Rapid Immunochromatic Assay.

4. Principle of the test kit

This RF One-Step test is based on a chromatographic principle in which a purified dog specific immunoglobulin reacts with epitopes of different subclasses. The purified dog specific immunoglobulin are conjugated to colloidal gold particles and the dog specific immunoglobulin is immobilized on the strip in the test zone “T”.
Rheumatoid factor in a sample that is applied to the strip at the sample zone “S” will bind to the gold particles which the migrate to zone “T”. A colour change in zone “T” indicates a positive test. Labelled colloidal gold particles are also immobilized on the test strip in the control zone “C”, to indicate that the test is working properly.

5. Contents

- 6 x Pouches, each containing 1 test strip and 1 pipette
- 6 x Buffer vial
- 1 x Protocol

6. Handling and storage of specimens

The One-Step should be stored at room temperature (±21°C). An unopened package can be used until the expiry date. An opened package must be used immediately. If the conditions are no longer fulfilled the test can no longer be used. Avoid freezing and heating as this will contribute to destruction of the test. Samples may be used fresh or may be kept frozen below -20°C before use.

7. Sample material

It is advised to test fresh serum or plasma samples. Do not use haemolytic or lipemic serum.

8. Precautions

- Handle all biological materials as though capable of transmitting infectious diseases.
- Do not pipette by mouth
- Do not eat, drink, smoke, prepare foods or apply cosmetics within the designated work area.
- Do not use components which passed the expiry date and do not mix components from different serials lots together.
- Optimal results will be obtained by strict adherence to this protocol. Careful pipetting and sampling throughout this procedure are necessary to maintain precision and accuracy.
- Each test strip is ultimately used as an optical reference. Therefore, do not touch the surface of the test strip and protect it from damage and dirt.

9. Test protocol

1. Unpack the test strip and pipette. Only open the amount of pouches to be used. An opened package should be used immediately.
2. Add **1 drop** of the sample to the buffer vial using the pipette (Figure 1).
3. Mix well using the pipette(Figure 2).
4. Add **4 drops** of the buffer vial containing the sample, with the included pipette ***slowly*** to the sample zone "S" (Figure 3).
5. Read the result after 5-10 minutes (for the interpretation of the test result see chapter 10 and chapter 11).

Figure 1

Figure 2

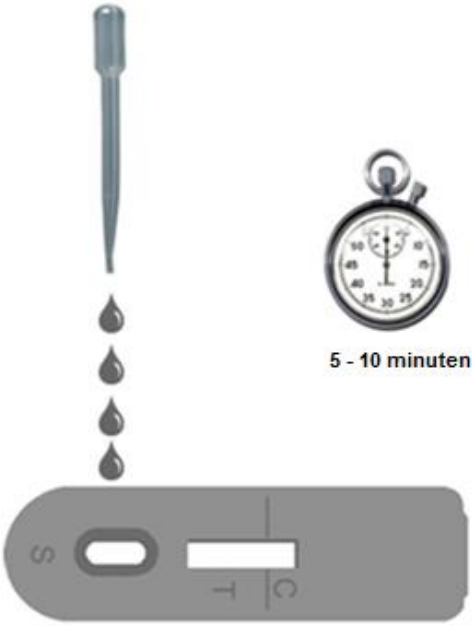

Figure 3

10. Validation of the test

To validate an EVL One-Step a control line should always be visible at control zone "C". If no control line is visible the test should be considered invalid.
Results should be read in the given time. Results read after the given time should be considered invalid. Invalid tests should be repeated with a new test.

11. Interpretation of the test results

Positive:
Two lines are visible in zone "T" and in zone "C" (Figure A). The sample contains Rheumatoid factor antibodies.
Positive results may vary in optical density due to variations in viral concentrations in the sample.

Weak positive:
Two lines are visible, a weak line in zone "T" and a line in zone "C" (Figure B). The sample contains low concentrations of Rheumatoid factor antibodies.

Negative:
Only one line is visible in zone "C" (Figure C). The sample does not contain Rheumatoid factor antibodies.

Not valid:
No line is visible in zone "C" (fig. D). Repeat the test procedure with a new test cassette.

Note:
Diseased, but negative tested patients should be re-tested within 2-3 weeks.

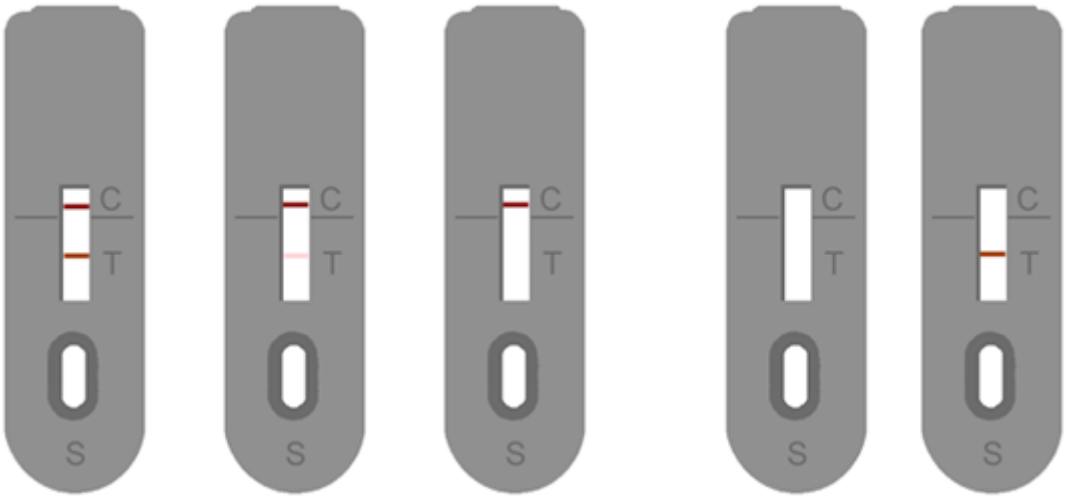

Figure A: Positive **Figure B: Weak positive** **Figure C: Negative** **Figure D: Not Valid**

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12. Symbols used with EVL ASSAYS

Symbol	English
	Consult instructions for use
	European Conformity
IVD	In vitro diagnostic device
RUO	For research use only
REF	Catalogue number
LOT	Lot/ No. / Batch code
	Contains sufficient for <n> tests
	Storage Temperature
	Expiration Date
	Legal Manufacturer
Distributed by	Distributor
Content	Content
Volume/No.	Volume / No.

The entire risk as to the performance of these products is assumed by the purchaser. EVL shall not be liable for indirect, special or consequential damages of any kind resulting from use of the products. In case of problems or questions contact EVL.