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For information Use only- Not to be used for performing the assay. Refer to the insert accompanying the kit

CE

Crypto-Strip , Crypto-CIT, Crypto Uni-Strip

***In vitro* Rapid Diagnostic Test for the detection of Cryptosporidium parvum oocysts in faecal specimens.**

FOR IN VITRO USE
FOR PROFESSIONAL USE ONLY
References: C-1005, 25 tests per kit.
C-1205, 20 tests individually packed
C-1505, 10 tests individually packed, sampling devices

I. INTRODUCTION

Cryptosporidiosis is one of the main causes of persistent diarrhoea in the developed world. It is caused by the presence of Cryptosporidium parvum oocysts in the gastro-intestinal tract. This parasite is known to be highly pathogenic and its infectious stage is transmitted by faecal-oral contract. It is also an opportunistic pathogen found in immunocompromised patients.
The symptoms of cryptosporidiosis are watery diarrhoea, stomach cramps, weight loss, nausea, and fever. In industrialized countries 2-2.5% of diarrhoeal hospitalized patients shed C. parvum oocysts. Ten percent of AIDS patients have chronic cryptosporidiosis and this figure can be as high as 40% in certain developing countries.
C. parvum is diagnosed by either Ziehl-Neelsen stain or immunofluorescence in smears of unconcentrated specimens. Several ELISA tests are also available for specific detection of oocyst antigens. New molecular biology methods, such as PCR, may be used to detect these parasites in the water supply or in specimens from asymptomatic carriers. All of these methods are extremely sensitive and must be used by experienced operators.
Coris BioConcept has developed an immunochromatographic test that can detect the oocysts of Cryptosporidium parvum in unconcentrated faecal samples within 10 minutes.

II. PRINCIPLE OF THE TEST

This test is ready to use and is based on the homogeneous membrane system technology with colloidal gold particles. The faecal sample must be diluted in the dilution buffer that is supplied with the test. A nitrocellulose membrane is sensitized with antibody to Cryptosporidium parvum oocysts.

The test's specificity is ensured by a monoclonal antibody specific to the Cryptosporidium parvum oocyst's membrane antigens that is conjugated to the colloidal gold. This conjugate is insolubilized on a polyester membrane.

When the strip is dipped into the faecal suspension, the solubilized conjugate migrates with the sample by passive diffusion and the conjugate and sample material come into contact with the anti-Cryptosporidium monoclonal antibody adsorbed onto the nitrocellulose. If the sample contains C. parvum oocysts the conjugate-oocyst complex remains bound to the anti-Cryptosporidium reagent. The result – in the form of a red line that forms on the strip – is visible within 10 minutes. The solution continues to migrate to encounter a second reagent (a control reagent) that binds the control conjugate, thereby producing a second dark red line and confirming that the test is working properly.

III. REAGENTS AND MATERIALS

Each kit contains crypto- strips, dilution buffer and optional components (for C-1505): :

1. **Crypto-Strip strips**

Each strip is sensitized with a mouse anti-Cryptosporidium monoclonal antibody that is specific to the Cryptosporidium parvum oocyst membrane antigens and a goat anti- chicken IgY antibody. These purified reagents are adsorbed to nitrocellulose. The anti-Cryptosporidium conjugate is produced using monoclonal antibody directed against the oocyst membrane antigens and coupled to colloidal gold particles after purification.

These strips come in a bottle or a pouch with a desiccant.

2. Dilution buffer (15 ml)

Salt solution buffered to pH 7.5 with Tris and containing EDTA, NaN3 (<0.1%), a detergent, and charged proteins.

3.Instruction for use (1)

4. Required materials supplied with C-1505):

- 3 or 5 ml test tubes
- inoculating loops for taking the faecal samples.
- carboard rack

Materials to be ordered separately :
-Cryptosporidium Control Test (Ref.: C-1085)

IV. SPECIAL PRECAUTIONS

- All operations linked to the use of the test must be performed in accordance with Good Laboratory Practices (GLP).
- The Crypto-Strip are for *in vitro* diagnostic only.
- Avoid touching the nitrocellulose with your fingers.
- Wear gloves when handling the samples.
- Dispose of gloves, swabs, test tubes, and sensitized strips in accordance with GLP.
- Never use reagents from another kit.
- If strips are stored in container, the container must be recapped resealed as soon as the necessary number of strips for the operation has been removed, since the strips are sensitive to humidity. Make sure that the desiccant is present.
- If strips are stored in individual pouches, pouch must be opened with care to avoid damaging the strip.Two green lines indicate the antibody adsorption sites. They disappear in the course of the test.
- Discard the buffer solution if it is contaminated with bacteria or mould.
- The reagents' quality cannot be guaranteed beyond their shelf-life dates or if the reagents are stored under inappropriate conditions.

To avoid diluting the colloidal gold conjugate in the solution, take care not to immerse the strip above the line placed under the orange arrow.

V. STORAGE

An unopened Crypto-Strip kit may be kept at between 4 and 30°C and used until the shelf-life date on the packaging.

The strips remain stable for 15 weeks (in the closed container) after the bottle is opened if they are kept at between 4 and 30°C and in a dry environment.
The Crypto-Strips and the buffer must not be frozen.

VI. SAMPLES

The stool specimens must be tested as soon after they are collected as possible. If necessary, they may be stored at 2-8°C for 1 week or -20°C for longer periods of time.
Make sure that the specimens are not treated with solutions containing formaldehyde or its derivatives.

VII. PROCEDURE

Preparations: If the Crypto-Strip was kept at 4°C, let all the reagents warm up to room temperature before proceeding with the test.
Write the patient's name or specimen number on the test tube (foresee one test tube per sample).
Place the marked test tubes in a rack.

Procedure:

1. Add 0.5 ml or 15 drops of the dilution buffer solution to each tube.
2. Dip the inoculating loop containing the stool sample into the tube. The dilution ratio must be at most 4% w/v.For liquid samples, take 2 loops of 10 µL, for solid samples, take 1 loop.
3. Stir to homogenize the solution and let stand for 1-2 minutes.
4. Discard the inoculating loop and immerse the sensitized strip in the direction indicated by the orange arrow.
5. Let react for 10 minutes, Results must be read on wet strips after 10 minutes incubation.

VIII. INTERPRETING THE RESULTS

The results are to be interpreted as follows:

1 line = negative
2 lines = positive
0 line = invalid*
* The absence of the control line, whic

The intensity of the test line may vary according to the quantity of antigens found in the sample. Any signal, even weak, on the test line must be regarded as a positive result. Nevertheless, the test is qualitative and cannot predict the quantity of antigens present in the sample. The clinical presentation and other test results must be taken into consideration to establish diagnosis.
During the drying process, a very faint shadow may appear at the test line. It should not be regarded as a positive result.

To store the results, let the strip dry after removing the absorbent material at its base.

IX. QUALITY CONTROL

In accordance with Good Laboratory Practices, we recommend checking the test's performance regularly in line with the laboratory's requirements. To do this, the Cryptosporidium Control Test (C-1085), in which the strip is immersed, may be used. Refer to the C-1085 package insert.

X. PERFORMANCES

A. Sensitivity – Specificity (Correlation):

The test was validated in Coris BioConcept's laboratories by comparing the Crypto-Strip's results with those of a rapid immuno enzymatic Test. The Crypto-Strip kit's sensitivity was tested on 100 stool samples. The results of these tests are shown below:

Immuno enzymatic Test Crypto-Strip	Positive	Negative	Total
Positive	45	0	45
Negative	2	53	55
Total	47	53	100

Sensitivity = 95.7% (45/47)
Specificity = 100% (53/53)

Positive Predictive Value = 100% (45/45)
Negative predictive Value = 96.4% (53/55)

Reliability = 98.0% (98/100)
(N = 100)

B. Accuracy:

To check the intra-lot accuracy, same positive sample and a buffer solution have been processed 15 times on sticks of the same production lot in the same experimental conditions. All observed results were correct as expected.

To check the inter-lot accuracy, some samples (positive and buffer) were processed on three different production lots. All results were correct as expected.

C. Interference:
Tests for cross-reactivity to the following pathogens were conducted and found to be negative:

- Adenovirus – groups A-F (n = 25)
- Adenovirus 40/41 (n = 15)
- *Campylobacter jejuni* (n = 10)
- *Giardia lamblia* (n = 10)
- Rotavirus (n = 25)
- *E. coli* 0157: H7 (n = 2)
- *S. typhimurium* (n = 1)
- *S. enteritidis* (n = 1)
- *Yersinia enterocolitica* (n = 3)
- *Helicobacter pylori* (n = 1)
- *Aeromonas hydrophila* (n = 1)

XI. LIMITS OF THE KIT
Crypto-Strip kit results must be compared with all other available clinical and laboratory information.
A positive test does not rule out the possibility that other pathogens may be present.
The Crypto-Strip is an acute-phase screening test. Stool specimens that are collected after this phase may contain antigen titres below the reagent's sensitivity threshold.

XII. TECHNICAL PROBLEMS / COMPLAINTS

If you encounter a technical problem, or if performances do not correspond with those indicated in this package insert:

1. Record the lot No of the kit in question
2. If necessary, store the problematic sample in the freezer as soon as possible
3. Contact Coris BioConcept or your local distributor

XI. BIBLIOGRAPHIC REFERENCES

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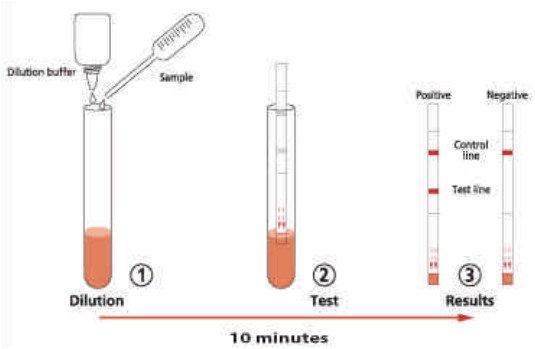
REF	Catalogue number		Manufactured by
IVD	In vitro diagnostic medical device		Temperature limitation
Σ	Contains sufficient for <n> tests	DIL SPE	Diluent specimen
	Consult instructions for use		Do not reuse
	Keep dry		Use by
DIL AS	Diluent assay	CONT NaN3	Contains Sodium azide