VITASSAY

Campylobacter

Rapid test for the qualitative detection of Campylobacter in human stool samples.

IUE-7355028 Ed02 August 2023

CE IVD EN

Vitassay

EN

For professional *in vitro* diagnostic use only.

INTENDED USE

Vitassay Campylobacter is a rapid one step immunochromatographic assay for the qualitative detection of *campylobacter* in human stool samples.

Simple, non-invasive and highly sensitive screening assay to make a presumptive diagnosis of *campylobacter* infection (campylobacteriosis).

INTRODUCTION

Campylobacter is a fastidious Gram-negative bacterium considered to be a common cause of acute, self-limiting gastroenteritis in the developed world.

Campylobacter jejuni and *Campylobacter coli* are the predominant causative agents of campylobacteriosis. The symptoms of gastroenteritis are sometimes severe in infants and at the elderly and bacterial culture is required for the diagnosis.

In developed countries, consumption of contaminated chicken, red meat, water, milk, and contact with pets and farm animals have been implicated as potential sources of Campylobacter infection.

Disease is associated with fever, bloody diarrhea, headache and severe abdominal pain. Campylobacteriosis is a self-limiting disease and antimicrobial therapy is not generally required. However, timely treatment can reduce the duration and severity of the infection. Most people who develop campylobacteriosis recover completely within 2-5 days, although sometimes recovery can take up to 10 days.

PRINCIPLE

Vitassay Campylobacter is a qualitative immunochromatographic assay for the detection of *Campylobacter* in human stool samples.

The test line zone of the nitrocellulose membrane is pre-coated with monoclonal antibodies against campylobacter.

During the process, the sample reacts with the antibodies against *Campylobacter*, forming conjugates. The mixture moves upward on the membrane by capillary action. If the sample is positive, antibodies present on the membrane (test line) capture the conjugate complex and a red line will be visible. Although the sample is positive or negative, the mixture continues to move across the membranes and the green control line always appears.

The presence of this green line (in the control zone (C)) indicates that sufficient volume is added; proper flow is obtained and serves as an internal control for the reagents.

PRECAUTIONS

- For professional *in vitro* use only.
- Do not use after expiration date.
- Do not use the test if the aluminium pouch is damaged.
- Read the instructions for use carefully before using the test.
- Do not use the kit if the label sealing the outer carton is torn or if the bags are open or damaged on arrival.
- Do not use the tests if the desiccant material is missing or broken inside the aluminium pouch.
- Specimens should be considered potentially hazardous and should be handled in the same manner as an infectious agent, following local/national regulations. A new test should be used for each sample to avoid contamination errors.
- Material exposed to the samples should also be considered potentially hazardous and should be handled in the same manner as an infectious agent, following local/national regulations.
- Do not reuse. This is a single-use device.
- Used material should be disposed of in an appropriate biohazard container after testing.
- Reagents contain preservatives. Avoid any contact with the skin or mucous membrane. Consult safety data sheet, available on request.
- All reagents included in the kit are approved for use with **Vitassay Campylobacter** only. Do not mix or use the components with other batches of Vitassay. Do not use with reagents from other kits or commercial assays.
- Follow Good Laboratory Practices. These practices should include, but are not limited to, personal protective equipment (PPE), such as lab coat, surgical or appropriate mask or face shield, disposable gloves and eye protection. Take the necessary precautions during sample collection, transport, storage, handling and disposal. Each sample must be correctly and unequivocally identified to ensure proper traceability of samples.
- In case of spillage, clean thoroughly with a suitable disinfectant.
- Do not eat, drink or smoke in the workplace.
- The presence of yellow lines in the result window (control line area and test line area), before using the test, is completely normal and does not imply a failure in the functionality of the test.
- The visual interpretation of the results is done by coloured lines, the interpretation of the results should be done by a professional user without problems of visualisation and colour interpretation.
- A certificate of analysis can be provided on request (not included).

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STORAGE AND STABILITY

The storage temperature of the kits should be 2-30ºC.

Do not freeze.

Under these conditions, they can be used until the expiry date indicated on the kit label.

All kit components are for single use only and must remain in their primary packaging until use. The test must remain in the sealed pouch until use.

MATERIALS

MATERIAL PROVIDED	MATERIAL REQUIRED BUT NOT PROVIDED
▪ **25** tests/kit **Vitassay Campylobacter.** ▪ Instructions for use. ▪ **25** vials with diluent for the sample dilution.	▪ PPE, such as disposable gloves ▪ Specimen collection container ▪ Timer ▪ Micropipette (in case of liquid stool)

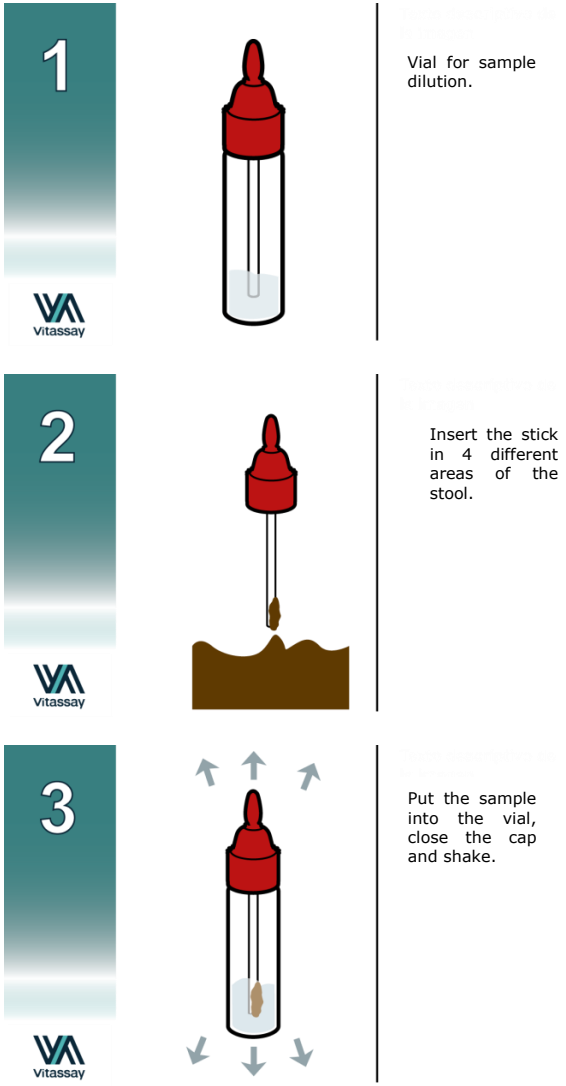
SPECIMEN COLLECTION

Collet sufficient quantity of feces: 1-2g or mL for liquid samples. Stool samples should be collected in clean and dry containers.

Samples can be stored in the refrigerator (2-8ºC) for 1-2 days prior to testing. For longer storage, maximum 1 year, the specimen must be kept frozen at -20ºC. Samples must be brought to room temperature before testing.

SPECIMEN PREPARATION

1. Remove the cap of the vial with diluent for the sample dilution (figure 1).
2. Use the stick to collect sufficient sample quantity. For solid stool, insert the stick in 4 different areas of the stool sample (figure 2), and add it into the vial with diluent for sample dilution. For liquid stool, take 125µL of the sample using a micropipette and transfer it into the vial with diluent for the sample dilution.
3. Close the vial with the diluent and stool sample. Shake vigorously the vial in order to assure good sample dispersion (figure 3).

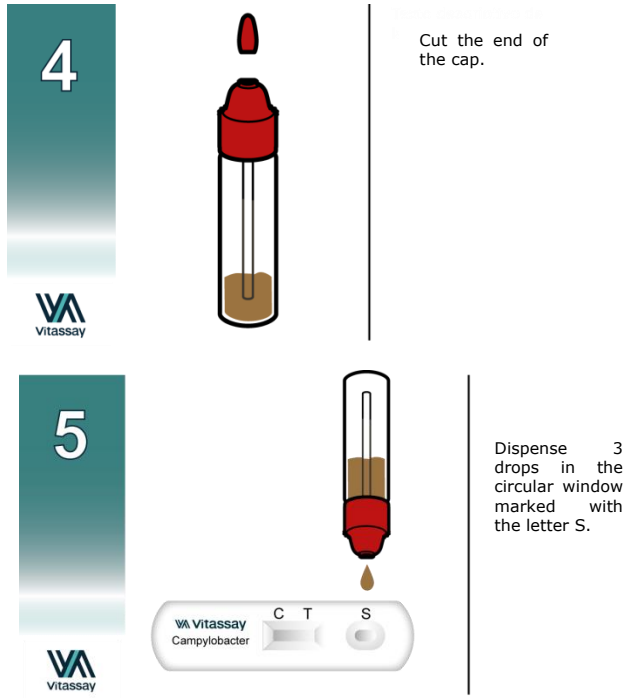


PROCEDURE

Allow the test, stool sample, controls and diluent to reach room temperature (15-30ºC) prior to testing. Do not open pouches until the performance of the assay.

1. Shake the stool collection tube to obtain a good sample dilution.
2. Remove the **Vitassay Campylobacter** from its sealed bag just before using it.
3. Take the stool collection tube containing the diluted sample, cut the end of the cap (figure 4) and dispense 3 drops in the circular window marked with the letter S (figure 5).
4. Read the results at **10 minutes**. Do not read the results later than 10 minutes.

If the test does not run due to solid particles, stir the sample added in the sample window with the stick. If it does not work, dispense a drop of diluent until seeing the liquid running through the reaction zone.



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INTERPRETATION OF THE RESULTS

	**NEGATIVE** Only one green line in the control zone **(C).**	There is no *Campylobacter* presence. No infection caused by *Campylobacter*.
	**POSITIVE** In addition to the green line (**control line C**), a red line appears (**test line T**).	There is *Campylobacter* presence. Possible *i*nfection caused by *Campylobacter*.
ANY OTHER RESULTS		Invalid result, we recommend repeating the assay using the sample with another test. **Note**: Wrong procedural techniques, insufficient sample volume or deterioration of the reagents are mostly the main reasons for control line failure. If the symptoms or situation persist, discontinue using the test kit and contact your local distributor.

Notes: The intensity of the red colored test line in the result line zone (T) will vary depending on the concentration of antigens in the specimen.

QUALITY CONTROL

Internal procedural control is included in **Vitassay Campylobacter**. Green line appearing in the results window is an internal control, which confirms sufficient specimen volume and correct procedural technique.

LIMITATIONS

- An excess of stool sample could cause wrong results (brown bands appear). Dilute the sample with the diluent and repeat the test.
- The intensity of test line may vary depending on the concentration of antigens.
- Freeze-thaw cycles of samples are not recommended as they may affect the results.
- The use of other samples different from human samples has not been established.
- The quality of **Vitassay Campylobacter** depends on the quality of the sample; Proper fecal specimens must be obtained.
- Positive results determine the presence of *Campylobacter* in faecal samples. A positive result should be followed up with additional techniques (PCR) to confirm the results. A confirmed infection should only be made by a physician after all clinical and laboratory findings have been evaluated and must be based in the correlation of the results with further clinical observations.
- Negative results should not be considered as conclusive; it is possible that the concentration of antigens is lower than the detection limit. If symptoms or situation still persist, a *Campylobacter* determination should be carried out, on a sample from an enrichment culture or another more sensitive technique.
- **Bloody stool** samples and/or **mucous stool** samples can cause non-specific reactions in the test. Such positive samples should be followed up with other diagnostic techniques to confirm the result.

EXPECTED VALUES

A strong association between *Campylobacter* infection and diarrhoea has also been described in the developed world. In developing country settings, *Campylobacter* infection has been most clearly implicated as a cause of diarrhoea only in the first 6 months of life. *Campylobacter* is often shed for extended periods following such episodes, and asymptomatic excretion is common.

Campylobacter gastroenteritis is especially common in children during the first 5 years of life with reported isolation rates of up to 46%.

In developed and devloping countries, *Campylobacter* cause more cases of diarrhoea than food borne *Salmonella*.

Campylobacteriosis occurs much more frequently in the summer months than in the winter.

PERFORMANCE CHARACTERISTICS

Analytical sensitivity (detection limit)

Detection limit values for the different species are:

For *Campylobacter jejuni* and *Campylobacter coli* detection:

The typical detection limit value is: 0.78 ng/mL of *Campylobacter jejuni* recombinant protein and 0.78 ng/mL of *Campylobacter coli* recombinant protein.

Clinical sensitivity and specificity

An evaluation with fecal samples was performed using **Vitassay Campylobacter** and qPCR technique (VIASURE Campylobacter Real Time PCR Detection kit, CerTest). The results were as follows:

		qPCR: VIASURE *Campylobacter* Real Time PCR Detection Kit		
		Positive	**Negative**	**Total**
Vitassay Campylobacter	**Positive**	59	1	60
	Negative	4	49	53
	Total	63	50	113

Table 1. Results of **Vitassay Campylobacter** compared to a commercial kit.

Vitassay Campylobacter vs VIASURE *Campylobacter* Real Time PCR Detection Kit		
	Mean Value	**95% confidence interval**
Sensitivity	93.7%	84.5-98.2%
Specificity	98.0%	89.4-99.9%
PPV	98.3%	91.1-100%
NPV	92.5%	81.8-97.9%

Table 2. Sensitivity, specificity, positive predictive values and negative predictive values of the **Vitassay Campylobacter** kit compared to a commercial kit (CI=95%).

The results showed that **Vitassay Campylobacter** has a high sensitivity and specificity to detect *Campylobacter*.

In addition, **Vitassay Campylobacter** was evaluated in a tertiary hospital, where 200 samples from patients with suspected *Campylobacter* gastrointestinal infection were used. To determine the presence of *Campylobacter*, samples were tested with Vitassay *Campylobacter* and a commercial rapid test. Subsequently, discrepant samples were tested with a commercial PCR.

The results were as follows:		**Commercial kit**		
		Positive	**Negative**	**Total**
Vitassay Campylobacter	**Positive**	71	0	71
	Negative	2	127	129
	Total	73	127	200

Table 3. Results of **Vitassay Campylobacter** compared to the reference method.

Vitassay Campylobacter vs Commercial kit			
Sensitivity	**Specificity**	**PPV**	**NPV**
99% (90.5-99.7%)	100% (97.1-100%)	100% (94.9-100%)	98.4% (94.5-99.8%)

Table 4. Sensitivity, specificity, positive predictive values, and negative predictive values of the **Vitassay Campylobacter** compared to the reference method.

These two evaluations show that the **Vitassay Campylobacter** kit has satisfactory clinical sensitivity and specificity values for the detection of *Campylobacter* in human stool samples.

Cross reactivity

No cross reactivity was detected against gastrointestinal pathogens that are occasionally present in feces:

Adenovirus	*Helicobacter pylori*	*Shigella boydii*
Astrovirus	*Legionella*	*Shigella dysenteriae*
Clostridium difficile antigen GDH	*Listeria monocytogenes*	*Shigella flexneri*
Clostridium perfringens	Norovirus GI	*Shigella sonnei*
Cryptosporidium	Norovirus GII	*Staphylococcus aureus*

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Entamoeba dispar	Rotavirus	*Streptococcus pneumoniae*
Entamoeba histolitica	*Salmonella enteritidis*	*Streptococcus pyogenes*
Escherichia coli O111	*Salmonella paratyphi A*	*Yersinia enterocolitica O:3*
Escherichia coli O149	*Salmonella paratyphi B*	*Yersinia enterocolitica O:9*
Escherichia coli O157:H7	*Salmonella typhi*	
Giardia	*Salmonella typhimurium*	

REFERENCES

1. JAMES A. PLATTS-MILLS; JIE LIU; JEAN GRATZ; ESTO MDUMA; CAROLINE AMOUR; NDEALILIA SWAI, MAMI TANIUCHI; SHARMIN BEGUM; PABLO PEÑARATO YORI; DRAKE H. TILLEY; GWENYTH LEE; ZELI SHEN; MARK T. WHARY; JAMES G. FOX; MONICA MCGRATH; MARGARET KOSEK; RASHIDUL HAQUE; ERIC R. HOUPT. "Detection of Campylobacter in stool and determination of significance by culture, Enzyme Immunoassay, and PCR in developing countries". Journal of Clinical Microbiology, April 2014, Volume 52, number 4, p. 1074-1080.

2. HIROSHI USHIJIMA; SHUICHI NISHIMURA; AKSARA THONGPRACHUM; YUKO SHIMIZU-ONDA; DINH NGUYEN TRAN; NGAN THI KIM PHAM; SAYAKA TAKANASHI; SHUVRA KANTI DEY; SHOKO OKITSU; WATARU YAMAZAKI, MASASHI MIZUGUCHI; SATOSHI HAYAKAWA. "Sensitive and rapid detection of Campylobacter species from stools of children with diarrhea in Japan by the Loop-Mediated Isothermal Amplification Method". Jpn. J. Infect. Dis., 67, 374-378, 2014.

3. P. RAJENDRAN; S. BABJI; AT GEORGE, DP RAJAN; G. KANG; SS. AJJAMPUR. "Detection and species identification of Campylobacter in stool samples of children and from Vellore, south India". Indian J. Med Microbiol. 2012; 30(1): 85-88.

4. SM SALIM; J MANDAL; SC PARIJA: "Isolation of Campylobacter from human stool samples". Indian Journal of Medical Microbiology, 2014, volume 32, Issue 1, pp. 35-38.

SYMBOLS FOR IVD COMPONENTS AND REAGENTS

IVD	*in vitro* diagnostic device		Keep dry
	Consult instructions for use		Temperature limitation
	Use by		Manufacturer
LOT	Batch code	Σ n	Contains sufficient for <n> test
DIL	Sample diluent	REF	Catalogue number
CE	CE Marking		

Changes control		
Nº Version	**changes**	**date**
IUE-7355028 Ed02 August 2023	Addition of a new evaluation. Format has been updated. Limitations sections has been updated. Transcription error in interpretation section has been corrected. Wording and grammatical changes have been implemented in Precautions, Limitations, Specimen collection, Storage and Stability. Material required but not included updated with minor changes.	24/08/2023

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