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QIAstat-Dx[®] Gastrointestinal Panel 2 Instructions for Use



Version 2

IVD For In Vitro Diagnostic Use

Rx ONLY For prescription use only

For use with QIAstat-Dx Analyzer 1.0 and QIAstat-Dx Analyzer 2.0

REF 691421



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R2

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Intended Use

The QIAstat-Dx[®] Gastrointestinal Panel 2 is a multiplexed nucleic acid test intended for use with the QIAstat-Dx Analyzer 1.0 and QIAstat-Dx Rise for the simultaneous qualitative detection and identification of nucleic acids from multiple viruses, bacteria, and parasites directly from stool samples Cary-Blair or modified Cary-Blair transport media obtained from individuals with signs and/or symptoms of gastrointestinal infection. The following viruses, bacteria (including several diarrheagenic *E. coli*/*Shigella* pathotypes), and parasites are identified with the QIAstat-Dx Gastrointestinal Panel 2:

Pathogen	Para-Pak C&S	FecalSwab
Adenovirus F40/F41	✓	✓
Astrovirus	✓	✓
Norovirus GI/GII	✓	✓
Rotavirus A	✓	✓
<i>Campylobacter</i> (<i>C. jejuni</i> , <i>C. coli</i> and <i>C. upsaliensis</i>)	✓	✓
<i>Shigella</i> /Enteroinvasive <i>Escherichia coli</i> (EIEC)	✓	✓
Enteropathogenic <i>Escherichia coli</i> (EPEC)	✓	Not reported
Enterotoxigenic <i>Escherichia coli</i> (ETEC) lt/st	✓	✓
Shiga-like toxin-producing <i>Escherichia coli</i> (STEC) stx1/stx2 (including specific identification of <i>E. coli</i> O157 serogroup within STEC)	✓	Not reported
<i>Salmonella</i>	✓	✓
<i>Plesiomonas shigelloides</i>	✓	✓
<i>Yersinia enterocolitica</i>	✓	✓
<i>Cryptosporidium</i>	✓	✓
<i>Cyclospora cayatanensis</i>	✓	✓

Pathogen	Para-Pak C&S	FecalSwab
<i>Entamoeba histolytica</i>	✓	✓
<i>Giardia lamblia</i> *	✓	✓

*Also known as *Giardia intestinalis* and *Giardia duodenalis*

Concomitant culture is necessary for organism recovery and further typing of bacterial agents

The QIAstat-Dx Gastrointestinal Panel 2 is indicated as an aid in the diagnosis of specific agents of gastrointestinal illness, in conjunction with other clinical, laboratory, and epidemiological data. Positive results do not rule-out co-infection with organisms not detected by the QIAstat-Dx Gastrointestinal Panel 2. The organisms detected may not be the sole or definitive cause of the disease.

QIAstat-Dx Gastrointestinal Panel 2 is not intended to monitor or guide treatment for *C. difficile* infections.

Negative QIAstat-Dx Gastrointestinal Panel 2 results in the setting of clinical illness compatible with gastroenteritis may be due to infection by pathogens that are not detected by this assay test or non-infectious causes such as ulcerative colitis, irritable bowel syndrome, or Crohn's disease.

The QIAstat-Dx Gastrointestinal Panel 2 also aids in the detection and identification of acute gastroenteritis in the context of outbreaks. The QIAstat-Dx Gastrointestinal Panel 2 is intended for professional use only and is not intended for self-testing. The QIAstat-Dx Gastrointestinal Panel 2 is intended for *in vitro* diagnostic use.

Description and Principle

Pathogen information

Acute gastrointestinal infections can be caused by a variety of pathogens, including parasites, bacteria and viruses, and generally present with nearly indistinguishable clinical signs and symptoms (1). The rapid and accurate determination of the presence or absence of potential causative agent(s) help make timely decisions regarding treatment, hospital admission, infection control and return of the patient to work and family (2–4). It may also greatly support improved antimicrobial stewardship and other important public health initiatives (3,5).

The QIAstat-Dx Gastrointestinal Panel 2 Cartridge allows the detection and differentiation of 16 parasitic, viral and bacterial pathogens that cause gastrointestinal symptoms. Testing requires a small sample volume and minimal hands-on time, and the results are available in approximately 78 minutes.

The QIAstat-Dx Gastrointestinal Panel 2 Cartridge allows detection and differentiation of 22 parasitic, viral and bacterial pathogens that cause gastrointestinal symptoms, which includes specific identification of *E. coli* O157 serogroup within STEC, resulting in 23 targets in total. Testing requires a small sample volume and minimal hands-on time, and the results are available in approximately 78 minutes.

Pathogens that can be detected and identified with the QIAstat-Dx Gastrointestinal Panel 2 are listed in Table 1.

Table 1. Pathogens detected by the QIAstat-Dx Gastrointestinal Panel 2

Pathogen	Classification (genome type)
Adenovirus F40/F41	Adenovirus (DNA)
Astrovirus	Astrovirus (RNA)

Table 1. Pathogens detected by the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Classification (genome type)
Norovirus GI/GII	Calicivirus (RNA)
Rotavirus A	Reovirus (RNA)
Sapovirus (GI, GII, GIV, GV)	Calicivirus (RNA)
<i>Campylobacter</i> (<i>C. jejuni</i> , <i>C. upsaliensis</i> , <i>C. coli</i>)	Bacterium (DNA)
<i>Clostridium difficile</i> (toxin A/B)	Bacterium (DNA)
Enteroaggregative <i>E. coli</i> (EAEC)	Bacterium (DNA)
Enteroinvasive <i>E. coli</i> (EIEC)/ <i>Shigella</i>	Bacterium (DNA)
Enteropathogenic <i>E. coli</i> (EPEC)	Bacterium (DNA)
Enterotoxigenic <i>E. coli</i> (ETEC) <i>lt/st</i>	Bacterium (DNA)
<i>Plesiomonas shigelloides</i>	Bacterium (DNA)
<i>Salmonella</i> spp.	Bacterium (DNA)
Shiga-like toxin-producing <i>E. coli</i> (STEC) <i>stx1/stx2</i> (including specific identification of <i>E. coli</i> O157 serogroup within STEC)	Bacterium (DNA)
<i>Vibrio vulnificus</i>	Bacterium (DNA)
<i>Vibrio parahaemolyticus</i>	Bacterium (DNA)
<i>Vibrio cholerae</i>	Bacterium (DNA)
<i>Yersinia enterocolitica</i>	Bacterium (DNA)
<i>Cryptosporidium</i>	Parasite (DNA)
<i>Cyclospora cayentanensis</i>	Parasite (DNA)
<i>Entamoeba histolytica</i>	Parasite (DNA)
<i>Giardia lamblia</i>	Parasite (DNA)

Note: Shiga-like toxin-producing *E. coli* (STEC) *stx1* and *stx2* are grouped together and reported as Shiga-like toxin-producing *E. coli* (STEC) *stx1/stx2*

Table 1. Pathogens detected by the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Classification (genome type)
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Note: *Giardia lamblia* is also known as *Giardia intestinalis* and *Giardia duodenalis*

Summary of detected organisms

Bacteria

***Campylobacter* spp. (*C. jejuni*/*C. coli*/*C. upsaliensis*)** is a genus of gram-negative bacteria, which includes more than 30 species (6). *Campylobacter jejuni* and *Campylobacter coli* are the most common *Campylobacter* species associated with diarrheal illness, being *C. jejuni* responsible for 90% of cases (7,9). The consumption of undercooked poultry or raw milk are the most common sources of *Campylobacter* infections (10,11). *Campylobacter* are highly infectious, with an infectious dose as low as 500 bacteria (12), however, person-to-person spread is uncommon (10). Systemic disease, associated with significant morbidity and mortality, may occur in individuals who are immunocompromised (10,12). Infection can result in long-term consequences such as arthritis, irritable bowel syndrome, and Guillain-Barré syndrome (10,12).

Plesiomonas shigelloides is a facultatively anaerobic gram-negative bacterium that can cause enteric disease in humans. The prevalence of *P. shigelloides* enteritis varies considerably, with higher rates reported from Southeast Asia and Africa and lower numbers from North America and Europe. It is unknown how many people suffer from disease caused by *P. shigelloides* each year, but mortality is rare. Infection especially occurs following the consumption of raw seafood or contaminated water (13).

Salmonella is a gram-negative bacterium comprising more than 2600 serovars, including the distinct typhoidal serotypes, Typhi and Paratyphi A–C (14,15). Enteric (typhoid) fever is an invasive, life-threatening, systemic infection with predominantly non-gastrointestinal symptoms

(14,16). Non-typhoidal salmonellosis is an acute, usually self-limiting gastroenteritis that is characterized by symptoms such as watery diarrhea, fever, abdominal pain, and sometimes vomiting (14,16,17). Less commonly, non-typhoidal *Salmonella* serovars cause invasive disease due to bloodstream infections that are not usually associated with diarrhea (14,16). There are 100–200 million cases of non-typhoidal salmonellosis each year, resulting in approximately 85,000–155,000 deaths (16,18). The incidence of non-typhoidal *Salmonella* gastroenteritis is highest in the developing world but is also of considerable importance in developed countries (14).

Yersinia enterocolitica is a gram-negative bacterium that has more than 70 serotypes (19); serotypes most commonly associated with infection are O:3, O:9, O:8, and O:5,27 (20). *Y. enterocolitica* infection have been reported frequently in northern Europe, particularly in Belgium, Norway, and the Netherlands; it is rarely observed in tropical countries (21). *Y. enterocolitica* is usually transmitted through the consumption of raw meats, unpasteurized dairy products, contaminated water, or via the fecal-oral route (22). Symptoms range from self-limiting enteritis with diarrhea, low-grade fever, and abdominal pain to severe disease such as terminal ileitis and mesenteric lymphadenitis, which also mimics appendicitis (23,24,25).

Diarrheagenic *Escherichia coli*/*Shigella*

E. coli* (EIEC)/*Shigella.are gram-negative facultative anaerobic bacteria belonging to the Enterobacteriaceae family. In addition to being part of the normal intestinal microflora of mammals, *E. coli*/*Shigella* contains several pathotypes that cause a variety of diseases (26,27). There are four major pathotypes of diarrhoeagenic *E. coli*/*Shigella*, which each have unique features in their interaction with eukaryotic cells: Enteropathogenic *E. coli* (EPEC), Enterohaemorrhagic *E. coli*/Shiga-like toxin-producing *E. coli* (EHEC/STEC), Enterotoxigenic *E. coli* (ETEC) and Enteroinvasive *E. coli* (EIEC)/*Shigella* (26,27). *E. coli*/*Shigella* have a conserved core genome and a flexible gene pool containing virulence and fitness genes, which are carried on mobile genetic elements (26,27). Gene gain, via horizontal transfer,

and gene loss afford the pathogenic traits to *E. coli/Shigella* that give rise to the different pathotypes (27).

Enteroinvasive *E. coli* (EIEC) and *Shigella*. EIEC is an invasive strain of *E. coli* that is very closely related in virulence and other pathogenic properties to *Shigella* (28,29). Sequencing indicates that EIEC is more related to *Shigella* than to non-invasive *E. coli*; however, they are currently classified as distinct species (26,28,30). The virulence of this pathogen is primarily due a plasmid-encoding virulence factors that allows the adhesion and invasion to the epithelial cells (31). EIEC is under-represented in epidemiological studies due its less severe manifestation and potential misclassification as *Shigella* (27). EIEC infection often leads only to self-limiting, mild watery diarrhea; in rare situations it can cause symptoms of shigellosis, but complications are uncommon (27). *Shigella* is the second leading cause of diarrhea mortality, causing approximately 13% diarrhea deaths (32). Numbers of deaths are greatest in young children and the elderly (32). It is recommended that individuals with shigellosis should not take anti-diarrheal medications such as loperamide, as these can make symptoms worse (33).

Enteropathogenic *E. coli* (EPEC) is primarily a disease of infants <2 years (27,34,35), and is commonly present in co-infections with other gastrointestinal pathogens (36). EPEC are classified into typical (tEPEC) and atypical (aEPEC) strains based on the presence of the *E. coli* adherence factor plasmid (pEAF). tEPEC is considered an important cause of infantile diarrhea in developing countries (37). Infections in adults, including travelers to developing countries, are rarely reported (27,35). aEPEC is frequently detected in both developing countries and industrialized countries and is suggested to be more prevalent than tEPEC (34). aEPEC is an important cause of both endemic diarrhea and outbreaks (34).

Enterotoxigenic *E. coli* (ETEC) is characterized by the production of heat-labile enterotoxins (LT) and heat-stable enterotoxins (ST) (38,39). ETEC is the most common diarrhea-associated *E. coli*, and although infections are usually self-limiting (39), is the eighth leading cause of diarrhea globally and accounts for >50,000 deaths every year (32). It also remains a major

cause of diarrhea in travelers to low resource countries (39). ETEC is a frequent antimicrobial resistant (39).

Shiga-like toxin-producing *E. coli* (STEC) *stx1*/*stx2*, including *E. coli* O157 is defined by the production of Shiga toxin 1 (*stx1*) or 2 (*stx2*), which show homology to *stx* toxins from *Shigella dysenteriae* (27). There are >400 serotypes of STEC, of which O157:H7 is the most common (27). Symptoms of STEC infection range from mild intestinal disease to hemorrhagic diarrhea and can lead to hemolytic uremic syndrome (HUS), end-stage renal disease and death (27,40). Approximately 5–10% of individuals diagnosed with STEC infections develop HUS, which can be a life-threatening complication (41). The impacts of STEC are often greater in infants and children, compared to other ages (40). Antibiotics should not be used to treat STEC infections as there is currently no evidence they aid recovery and have instead been associated with a worsening of symptoms and the development of HUS (41).

Parasites

***Cryptosporidium* spp.** are protozoan parasites that can infect humans and other animals, being *C. hominis* and *C. parvum* the causative strains of the majority of humans infections (42). *Cryptosporidium* spp. are found globally, but those in developing countries, particularly in sub-Saharan Africa, are at greater risk of infection due to poorer water treatment and food sanitation (32,43). It is also one of the leading causes of diarrheal mortality in children <5 years of age (32,44).

Cyclospora cayentanensis is a single-celled protozoa parasite, and the only known species of the genus *Cyclospora* to infect humans (45,46), It is endemic in tropical/subtropical areas, and in non-endemic regions, cases and outbreaks of cyclosporiasis are usually linked to international travel and consumption of contaminated produce imported from endemic regions (45,46,47). Direct fecal-oral transmission cannot occur, the unsporulated oocysts sporulate in water and food environments, enabling them to infect another host (45,46,48).

Entamoeba histolytica is an anaerobic, protozoan parasite (49). *E. histolytica* is common in developing countries, particularly those in the tropics and sub-tropics with poor sanitation (49,50,51). Only 10–20% of individuals infected with *E. histolytica* are symptomatic [1,2]. Through destruction of the intestinal walls, trophozoites can also spread systemically to the liver, lungs, and central nervous system (49,10,50,51). The liver is the most common extra-intestinal site of infection (49,50,51).

Giardia lamblia is a unicellular, protozoan parasite that can cause disease in humans and other mammals (52,53). *G. lamblia* has a global distribution and is common in both children and adults (54,55). Prevalence of infection is higher in developing regions of the world and in children (52,54,55). The majority (50–75%) of *G. lamblia* infections are asymptomatic (56). In immunocompetent individuals, infections are usually self-limiting, although some may become chronic (52).

Viruses

Adenovirus F40/41 is a double-stranded DNA, non-enveloped virus (57,58), with many distinct serotypes described and classified into 7 species (A–G) (57). Serotypes F40/41 are the most common cause of acute gastroenteritis in young children, causing 5–20% cases. More than 80% of diagnosed infections occur in children aged <4 years (58). Adenoviruses have a worldwide distribution and infections occur throughout the year without significant seasonal variability (57). Infections are usually mild and self-limiting in immunocompetent individuals but can be fatal in individuals who are immunocompromised (57,59,60).

Astroviruses are non-enveloped, positive-sense, single-stranded RNA viruses (61). Human astroviruses are distributed all over the world and are associated with 2–9% of cases of acute, nonbacterial diarrhea in children (61,62). It is estimated that 90% of the global population aged ≥9 years have antibodies against astrovirus type 1 (61). Many infections in healthy children and adults are asymptomatic, although they can cause severe diarrhea in children, older adults and those who are immunocompromised or have comorbidities (61,62).

Noroviruses GI/GII are small, non-enveloped, positive-stranded RNA viruses from the family Caliciviridae (63). They are responsible for >90% of viral gastroenteritis and around 50% of all-cause gastroenteritis outbreaks globally (64), causing approximately 685 million cases every year (65). Approximately 200 million cases are in children aged <5 years, leading to 50,000 child deaths (65). Norovirus is commonly known as the "winter-vomiting bug"; outbreaks are more common during the winter months, but infection can occur at any time of year (65). Norovirus is infectious at very low doses and is transmitted via aerosolized droplets and touching of contaminated surfaces (65). Individuals infected with norovirus usually recover within 1–3 days, but infections in infants, older adults and immunocompromised individuals can be severe and sometimes fatal (65). In some individuals, viral shedding can occur for many weeks/months after they have stopped experiencing symptoms, and this is a large contributing factor for outbreaks (66).

Rotavirus A is a non-enveloped, double-stranded RNA viruses of the Reoviridae family, with 10 species that cause infection in humans (A–J). However, rotavirus A is the most common species and causes >90% of all rotavirus infections (67,68). Rotavirus is a leading cause of diarrhea in children <5 years (67), with a seasonal infection pattern that differs across the world, particularly in middle–high income countries (69). Severe infection is most common in young children and infants; in adults, infections are often associated with milder symptoms (70). Two oral rotavirus vaccines are approved in the United States (71) and have been available in >100 countries since 2006 (71). These vaccines have substantially reduced the burden of rotavirus-associated illness (70).

Summary and explanation

QIAstat-Dx Gastrointestinal Panel 2 Cartridge description

The QIAstat-Dx Gastrointestinal Panel 2 Cartridge (Figure 1) is a disposable plastic device that allows performance of fully automated molecular assays for the detection of gastrointestinal pathogens. Main features of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge include compatibility with a liquid sample type, hermetical containment of the pre-loaded reagents necessary for testing, and true walk-away operation. All sample preparation and assay testing steps are performed within the cartridge.

All reagents required for the complete execution of a test run are pre-loaded and self-contained in the QIAstat-Dx Gastrointestinal Panel 2 Cartridge. The user does not need to come in contact with and/or manipulate any reagents. The QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0 houses air filters for both incoming and outgoing air, further safeguarding the environment. After testing, the cartridge stays hermetically closed at all times, greatly enhancing its safe disposal.

Within the cartridge, multiple steps are automatically performed in sequence using pneumatic pressure to transfer samples and fluids via the transfer chamber to their intended destinations.

Description of the process

After sample is manually loaded, the diagnostic tests with the QIAstat-Dx Gastrointestinal Panel 2 are performed on the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0. All of the sample preparation and analysis steps are performed automatically by the QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0.

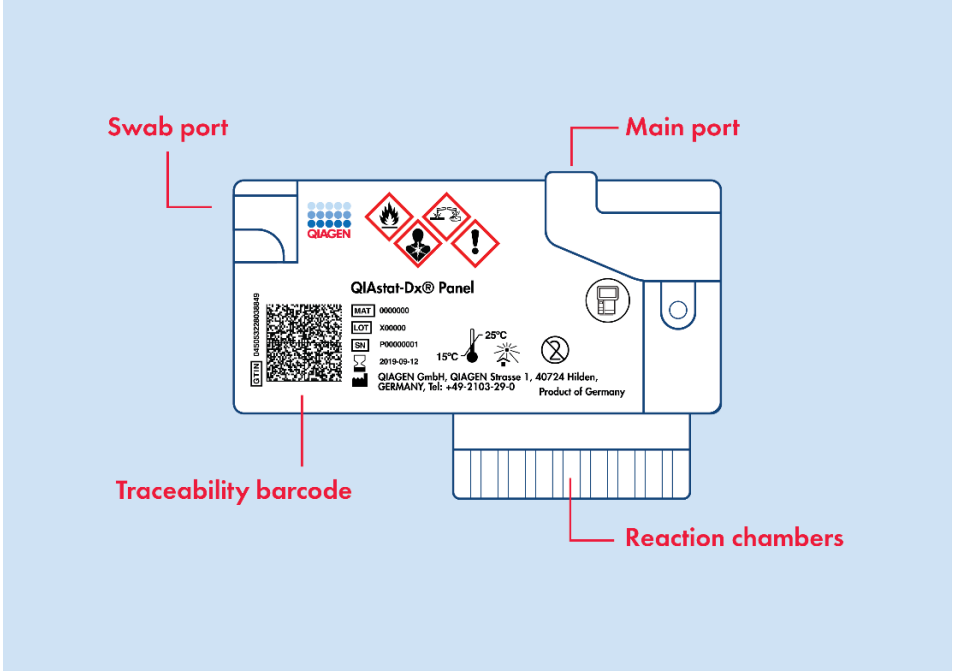


Figure 1. Layout of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge and its features.

Note: The swab port is not used for the QIAstat-Dx Gastrointestinal Panel 2.

Materials Provided

Kit contents

QIAstat-Dx Gastrointestinal Panel 2 Cartridge* **6**

QIAstat-Dx Gastrointestinal Panel 2 Cartridges* 6

Transfer pipettes† 6

QIAstat-Dx Gastrointestinal Panel 2 Product information Card 1

* 6 individually packaged cartridges containing all reagents needed for sample preparation and multiplex real-time RT-PCR, plus Internal Control.

† 6 individually packaged transfer pipettes for dispensing liquid sample into the QIAstat-Dx Gastrointestinal Panel 2 Cartridge.

Materials Required but Not Provided

Platform and software*

The QIAstat-Dx Gastrointestinal Panel 2 is designed for use with the QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0. Before beginning a test, make sure the following are available:

- QIAstat-Dx Analyzer 1.0 (at least one Operational Module and one Analytical Module) with software version 1.4 or later OR QIAstat-Dx Analyzer 2.0 (at least one Operational Module PRO and one Analytical Module) with software version 1.6 or higher.

QIAstat-Dx Gastrointestinal Panel 2 is designed for use with the QIAstat-Dx Analyzer 1.0 and QIAstat-Dx Rise. Before beginning a test, make sure the following are available:

- QIAstat-Dx Analyzer 1.0 (at least one Operational Module and one Analytical Module) with software version 1.4 or later[†] OR a QIAstat-Dx Rise (at least two Analytical Modules must be inside for the machine to work) with software version 2.2 or later.
- QIAstat-Dx Analyzer 1.0 User Manual (for use with software version 1.4 or later) OR QIAstat-Dx Rise User Manual (for use with software version 2.2 or later)
- QIAstat-Dx latest Assay Definition File software for Gastrointestinal Panel 2 installed on the Operational Module or on the QIAstat-Dx Rise.

*Prior to use, ensure that instruments have been checked and calibrated according to the manufacturer's recommendations.

[†]DiagCORE[®] Analyzer instruments running QIAstat-Dx software version 1.4 or later can be used as an alternative to QIAstat-Dx Analyzer 1.0 instruments.

Warnings and Precautions

- The QIAstat-Dx Gastrointestinal Panel 2 is for *in vitro* diagnostic use.
- For prescription use only.
- The QIAstat-Dx Gastrointestinal Panel 2 is to be used by laboratory professionals trained in the use of QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0.
- False positives and false negatives can be the result of a variety of sources and causes. A trained healthcare professional should carefully interpret the results from the QIAstat-Dx Gastrointestinal Panel 2 in conjunction with a patient's signs and symptoms, results from other diagnostic tests, and relevant epidemiological information.
- Please be aware that you may be required to consult your local regulations for reporting serious incidents that have occurred in relation to the device to the manufacturer and the regulatory authority in which the user and/or the patient is established.

Safety information

- When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate safety data sheets (SDSs). These are available online in convenient and compact PDF format at www.qiagen.com/safety where you can find, view, and print the SDS for each QIAGEN kit and kit component.
- Observe standard laboratory procedures for keeping the working area clean and contamination-free. Guidelines are outlined in publications such as the *Biosafety in Microbiological and Biomedical Laboratories* from the Centers for Disease Control and Prevention and the National Institutes of Health (72).

- Specimens and samples are potentially infectious. Discard sample and assay waste according to your local safety procedures.
- Always wear appropriate personal protective equipment and follow your institution's safety procedures for handling biological samples. Handle all samples, cartridges, and transfer pipettes as if they are capable of transmitting infectious agents.
- Always observe safety precautions as outlined in relevant guidelines, such as the Clinical and Laboratory Standards Institute® (CLSI) *Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline (M29)*, or other appropriate documents provided by local authorities.
- The QIAstat-Dx Gastrointestinal Panel 2 Cartridge is a closed, single-use device that contains all reagents needed for sample preparation and multiplex real-time RT-PCR within the QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0. Do not use a QIAstat-Dx Gastrointestinal Panel 2 Cartridge that is past its expiration date, appears damaged or leaks fluid.
- Dispose of used or damaged cartridges in accordance with all national, state and local health and safety regulations and laws.

Emergency information

CHEMTREC

USA & Canada 1-800-424-9300

Precautions

The following hazard and precautionary statements apply to components of the QIAstat-Dx Gastrointestinal Panel 2.



Contains: ethanol; guanidine hydrochloride; guanidine thiocyanate; isopropanol; proteinase K; t-Octylphenoxypolyethoxyethanol. Danger! Highly flammable liquid and vapor. Harmful if swallowed or if inhaled. May be harmful in contact with skin. Causes severe skin burns and eye damage. May cause allergy or asthma symptoms or breathing difficulties if inhaled. May cause drowsiness or dizziness. Harmful to aquatic life with long lasting effects. Contact with acids liberates very toxic gas. Corrosive to the respiratory tract. Keep away from heat/sparks/open flames/hot surfaces. No smoking. Avoid breathing dust/fume/gas/mist/vapors/spray. Wear protective gloves/protective clothing/eye protection/face protection. Wear respiratory protection. IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. IF exposed or concerned: Immediately call a POISON CENTER or doctor. Rinse mouth. Do NOT induce vomiting. Remove person to fresh air and keep comfortable for breathing. Wash contaminated clothing before reuse. Store in a well-ventilated place. Keep container tightly closed.

To reduce the risk of contamination when handling stool samples, it is recommended that the below guidelines are applied (72).

- When handling the stool sample, a biosafety cabinet, dead air box, splash shield, or face shield should be used.
- The work area used for cartridge loading should be separate from the work area used for stool pathogen testing (i.e., stool culture, EIA) to prevent cross-contamination.
- Prior to sample handling, the work area should be thoroughly cleaned using 10% bleach or similar disinfectant.
- QIAstat-Dx Gastrointestinal Panel 2 Cartridges and samples should be processed one at a time.
- Change gloves prior to removing cartridges from shipping boxes.
- Change gloves and clean the work area between processing each sample.
- Dispose of used cartridges in a biohazard container immediately after the run is complete and avoid excessive handling.

Precautions related to Public Health Reporting

National, state, and local public health organizations have published guidelines for the notification of reportable diseases. While the list of reportable conditions varies by state, the Council of State and Territorial Epidemiologists (CSTE) has recommended that state health departments report cases of selected diseases to CDC's National Notifiable Diseases Surveillance System (NNDSS). At the time of writing, the notifiable pathogens in the US per CDC included in the Gastrointestinal Panel 2 are:

- *Campylobacter* spp.
- Certain *E. coli*
 - O157:H7
 - Shiga toxin-producing (STEC)
- *Cryptosporidium* spp.
- *Cyclospora cayetanensis*
- *Giardia lamblia*
- *Salmonella* spp.
- *Salmonella* enterica serotypes Paratyphi A, B [tartrate negative], and C [S. Paratyphi]]
- *Salmonella* enterica serotype Typhi
- *Shigella* spp./EIEC

Pathogens are notifiable due to their outbreak potential or impact on public health. Laboratories are responsible for following their state or local regulations for submission of clinical material or isolates on positive specimens to their state public health laboratories.

Reagent Storage and Handling

Store the QIAstat-Dx Gastrointestinal Panel 2 Cartridges in a dry, clean storage space at room temperature (15–25°C). Do not remove the QIAstat-Dx Gastrointestinal Panel 2 Cartridges or the transfer pipettes from their individual packaging until actual use. Under these conditions, QIAstat-Dx Gastrointestinal Panel 2 Cartridges can be stored until the expiration date printed on the individual packaging. The expiration date is also included in the QIAstat-Dx Gastrointestinal Panel 2 Cartridge barcode and is read by the QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0 when the cartridge is inserted into the instrument to run a test. Once the cartridge is removed from the pouch it should be protected from sunlight.

Attention should be paid to the expiration dates and storage conditions printed on the box and labels of all components. Do not use expired or incorrectly stored components.

In-use stability

When stored under the specified storage conditions, the QIAstat-Dx Gastrointestinal Panel 2 is stable until the stated expiration date on box label.

After the cartridge package is opened, sample should be introduced into the QIAstat-Dx Gastrointestinal Panel 2 Cartridge within 30 minutes. Sample-loaded cartridges should be loaded into the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 within 90 minutes.

Specimen Storage and Handling

The QIAstat-Dx Gastrointestinal Panel 2 is for use with stool samples resuspended in transport medium (Para-Pak C&S (Meridian Bioscience) or FecalSwab (COPAN)). All samples should be treated as potentially infectious. Discard sample and assay waste according to your local safety procedures.

Specimen collection

Stool samples should be collected and handled according to the transport medium manufacturer's recommended procedures.



Recommended storage conditions for stool resuspended in transport medium (Para-Pak C&S (Meridian Bioscience) or FecalSwab (COPAN)) specimens are listed below:

- Room temperature up to 4 days at 15–25°C
- Refrigerated up to 4 days at 2–8°C

Procedure

Important point before starting

Ensure all materials required but not provided are available.

The QIAstat-Dx Gastrointestinal Panel 2 Cartridge (cat. no 691421) is identified by a purple-colored () bar on the label and an icon indicating gastrointestinal tract ( , see "Symbols" on page 128).

Protocol: Stool samples in transport medium

Sample collection, transport, and storage

Collect and resuspend the stool sample in Para-Pak C&S (Meridian) or FecalSwab (COPAN) transport media according to the manufacturer's recommended procedures.

Loading a sample into the QIAstat-Dx Gastrointestinal Panel 2 Cartridge

1. Open the package of a QIAstat-Dx Gastrointestinal Panel 2 Cartridge using the tear notches on the sides of the packaging (Figure 2).

Important: After the package is open, sample should be introduced into the QIAstat-Dx Gastrointestinal Panel 2 Cartridge within 30 minutes. Sample-loaded cartridges should be loaded into the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 within 90 minutes.

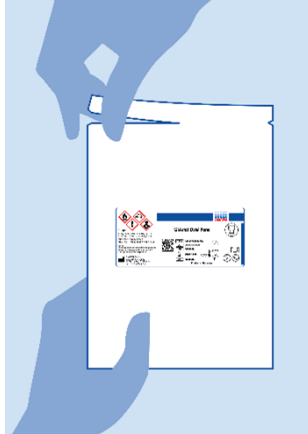


Figure 2. Opening the QIAstat-Dx Gastrointestinal Panel 2 Cartridge.

2. Remove the QIAstat-Dx Gastrointestinal Panel 2 Cartridge from the packaging and position it so that the barcode on the label faces you.
3. Manually write the sample information, or place a sample information label, on the top of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge. Make sure that the label is properly positioned and does not block the lid opening (Figure 3).

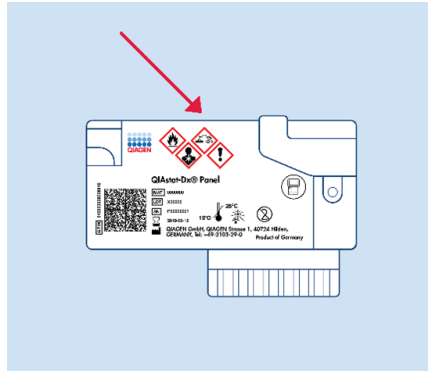


Figure 3. Sample information placement on top of QIAstat-Dx Gastrointestinal Panel 2 Cartridge.

4. Place the QIAstat-Dx Gastrointestinal Panel 2 Cartridge flat on the clean work surface so that the barcode on the label faces upwards. Open the sample lid of the main port on the front of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge (Figure 4).

Important: Do not flip the QIAstat-Dx Gastrointestinal Panel 2 Cartridge or agitate it while the main port lid is open. The main port contains silica beads used in the sample disruption. The silica beads could fall out of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge if it is agitated while the lid is open.

Note: The swab port is not used for the QIAstat-Dx Gastrointestinal Panel 2 assay.

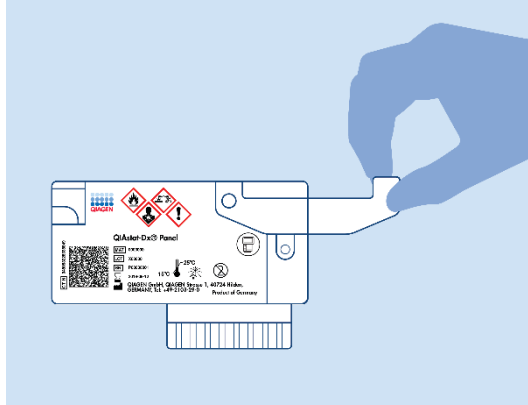


Figure 4. Opening the sample lid of main port.

5. Thoroughly mix the stool in the Para-Pak C&S or FecalSwab transport medium, for example, by vigorously agitating the tube 3 times (Figure 5).

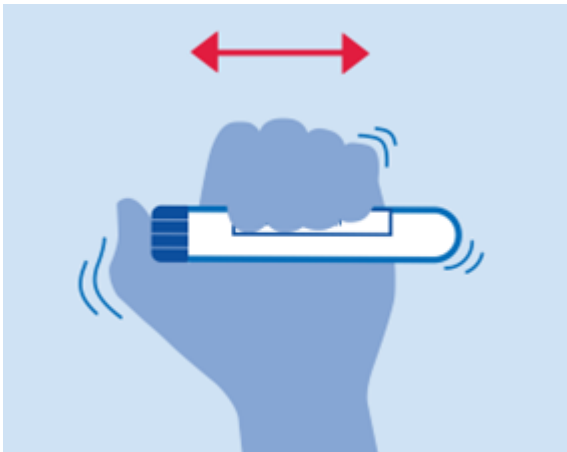


Figure 5. Mixing stool sample in transport medium.

6. Open the tube with the sample to be tested. Use the supplied transfer pipette to draw up fluid. Draw the sample to the second fill line on the pipette (i.e., 200 μL) (Figure 6).

Important: Do not draw air, mucus, or particles into the pipette. If air, mucus, or particles are drawn into the pipette, carefully expel the sample fluid in the pipette back into the sample tube and draw up fluid again. In the event that the supplied transfer pipette is lost please use another one from the package or any other commercially available pipette with a minimum volume of 200 μL .

Note: In the case the test should be repeated due to previous cartridge error related to sample concentration too high, draw the sample to the first fill line on the pipette instead (100 μL) (See "Troubleshooting Guide" on page 126 for further details on error codes and "Appendix C: Additional instructions for use" on page 136 for further instructions on repeating a sample with 100 μL).

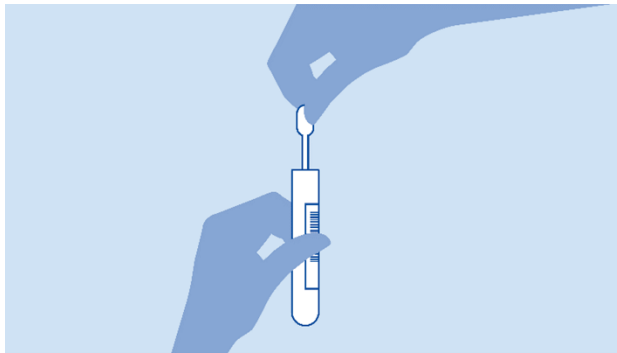


Figure 6. Drawing up sample into the supplied transfer pipette.

7. Carefully transfer the sample into the main port of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge using the supplied single-use transfer pipette (Figure 7).

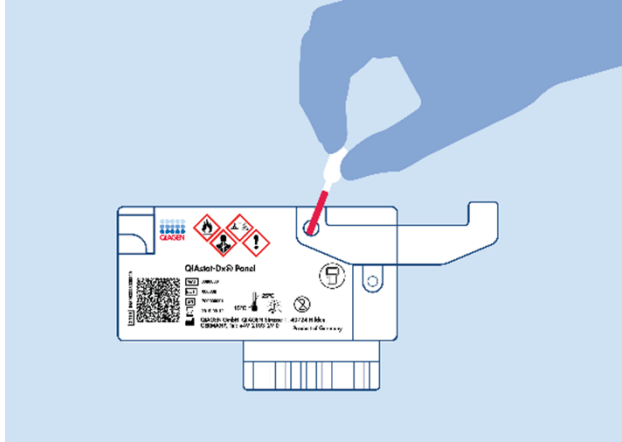


Figure 7. Transferring sample to main port of QIAstat-Dx Gastrointestinal Panel 2 Cartridge.

8. Firmly close the lid of the main port until it clicks (Figure 8).

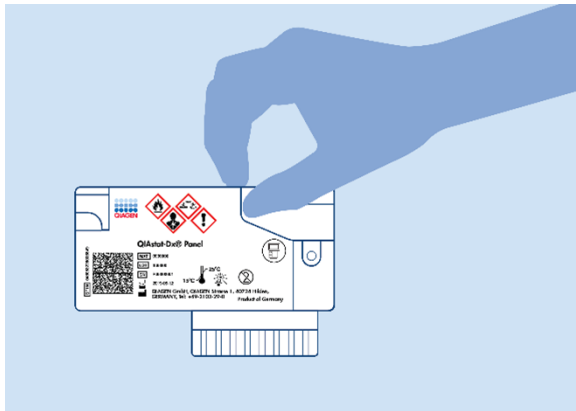


Figure 8. Closing the lid of the main port.

9. Visually confirm that the sample has been loaded by checking the sample inspection window of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge (Figure 9). A mixture of

sample and silica beads should be observed.

Important: After the sample is placed inside the QIAstat-Dx Gastrointestinal Panel 2 Cartridge, the cartridge must be loaded into the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 within 90 minutes.

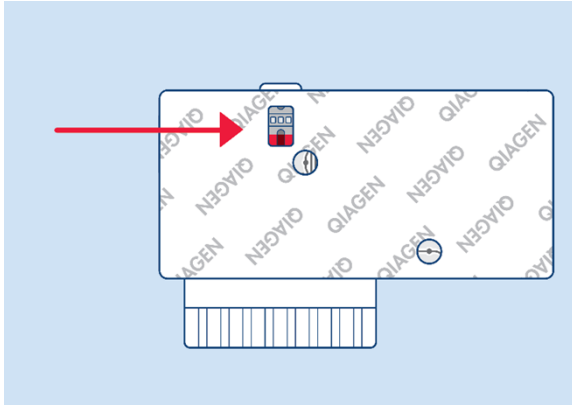


Figure 9. Sample inspection window (red arrow).

Running a test with a QIAstat-Dx Analyzer

1. Power on the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 using the **On/Off** button on the front of the instrument.

Note: The power switch at the back of the Analytical Module must be set in the “I” position. The QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 status indicators will turn blue.

2. Wait until the Main screen appears, and the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 status indicators turn green and stop blinking.

3. Enter your username and password for QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 to log in.

Note: The Login screen will appear if User Access Control is activated. If the User Access Control is disabled, username/password will not be required, and the Main screen will appear.

4. If the Assay Definition File software is not installed on the QIAstat-Dx Analyzer 1.0, or the QIAstat-Dx Analyzer 2.0 follow the installation instructions prior to running the test (see "Appendix A: Installing the Assay Definition File" on page 131 for additional information).
5. Press **Run Test** in the top right corner of the touchscreen of the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0.
6. When prompted, scan the sample ID barcode on the resuspended sample, or scan the specimen information barcode located on the top of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge (Figure 3), using the integrated front barcode reader of the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 (Figure 10).

Note: It is also possible to enter the sample ID using the virtual keyboard of the touchscreen by selecting the **Sample ID** field.

Note: Depending on the selected system configuration, entering the patient ID may also be required at this point.

Note: Instructions from the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 appear in the Instructions Bar at the bottom of the touchscreen.

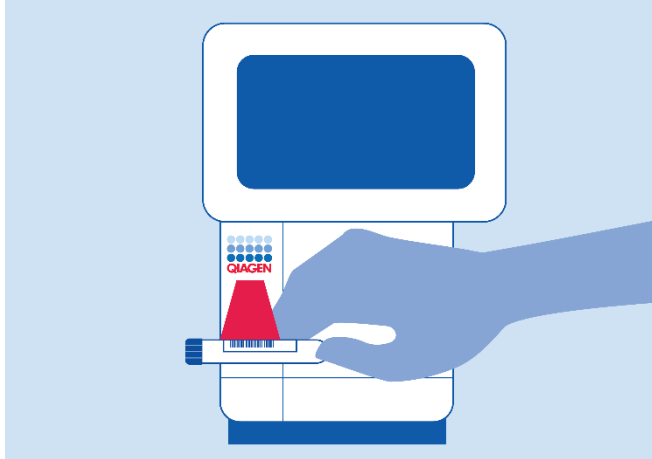


Figure 10. Scanning sample ID barcode.

7. When prompted, scan the barcode of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge to be used (Figure 11). The QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 will automatically recognize the assay to be run based on the cartridge barcode.

Note: The QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0 will not accept QIAstat-Dx Gastrointestinal Panel 2 Cartridges with lapsed expiration dates, previously used cartridges or cartridges for assays that have not been installed on the unit. An error message will be shown in these cases and the QIAstat-Dx Gastrointestinal Panel 2 Cartridge will be rejected. Refer to the *QIAstat-Dx Analyzer 1.0 User Manual* or the *QIAstat-Dx Analyzer 2.0 User Manual* or "Appendix A: Installing the Assay Definition File" on page 131 for further details on how to install assays.

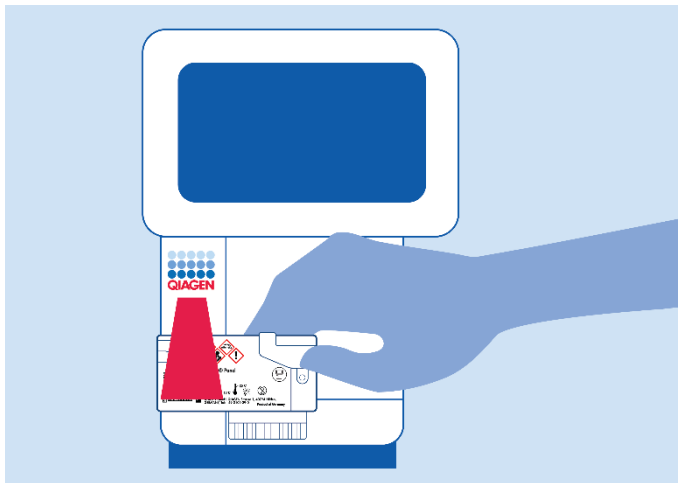


Figure 11. Scanning QIAstat-Dx Gastrointestinal Panel 2 Cartridge barcode.

8. Select sample type option from the list (Figure 12) based on the sample collection device used (Para-Pak C&S or FecalSwab).



Figure 12. Selecting sample type.

9. The Confirm screen will appear. Review the entered data and make any necessary changes by selecting the relevant fields on the touchscreen and editing the information.
10. Press **CONFIRM** when all the displayed data are correct. If needed, select the appropriate field to edit its content, or press **Cancel** to cancel the test (Figure 13).

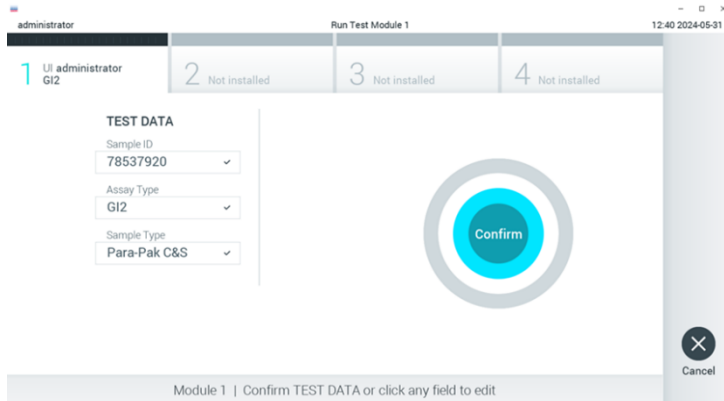


Figure 13. Confirming data entry.

11. Ensure that both sample lids of the swab port and main port of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge are firmly closed.
12. When the cartridge entrance port on the top of the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 automatically opens, insert the QIAstat-Dx Gastrointestinal Panel 2 Cartridge with the barcode facing to the left and the reaction chambers facing down (Figure 14).

Note: Depending on the system configuration, the operator may be required to re-enter their user password to start the test run.

Note: Up to this point, it is possible to cancel the test run by pressing the **Cancel** button in the bottom right corner of the touchscreen.

13. Upon detecting the QIAstat-Dx Gastrointestinal Panel 2 Cartridge, the QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0 will automatically close the lid of the cartridge entrance port and start the test run. No further action from the operator is required to start the run.

Note: There is no need to push the QIAstat-Dx Gastrointestinal Panel 2 Cartridge into the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0.

Note: The QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0 will not accept a QIAstat-Dx Gastrointestinal Panel 2 Cartridge other than the one used and scanned during the test setup. If a cartridge other than the one scanned is inserted, an error will be generated, and the cartridge will be automatically ejected.

Note: The lid of the cartridge entrance port will close automatically after 30 seconds if a QIAstat-Dx Gastrointestinal Panel 2 Cartridge is not positioned in the port. If this occurs, repeat the procedure starting with step 5.

14. While the test is running, the remaining run time is displayed on the touchscreen.

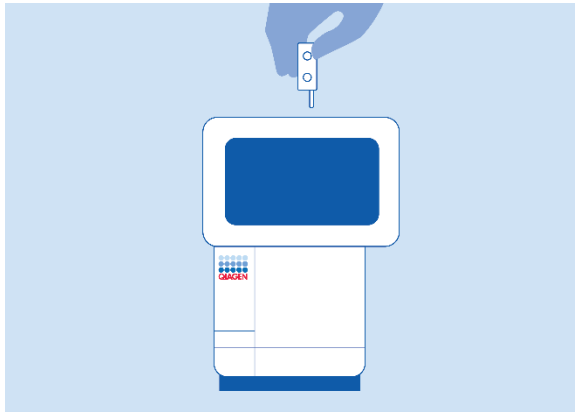


Figure 14. Inserting QIAstat-Dx Gastrointestinal Panel 2 Cartridge into QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0.


15. After the test run is completed, the Eject screen will appear (Figure 15) and the Module status bar will display the test result as one of the following options:

- TEST COMPLETED: The test was completed successfully
- TEST FAILED: An error occurred during the test
- TEST CANCELED: The user canceled the test

Important: If the test fails, refer to the “Troubleshooting” section in the *QIAstat-Dx Analyzer 1.0 User Manual* or the *QIAstat-Dx Analyzer 2.0 User Manual* for possible reasons and instructions on how to proceed. For additional information about specific QIAstat-Dx Gastrointestinal Panel 2 error codes and messages, please see “Troubleshooting Guide” on page 126 of this document.



Figure 15. Eject screen display.

16. Press  **Eject** on the touchscreen to remove the QIAstat-Dx Gastrointestinal Panel 2 Cartridge and dispose of it as biohazardous waste in accordance with all national, state and local health and safety regulations and laws. The QIAstat-Dx Gastrointestinal Panel 2 Cartridge should be removed when the cartridge entrance port opens and ejects the cartridge. If the cartridge is not removed after 30 seconds, it will automatically move back

into the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 and the cartridge entrance port lid will close. If this occurs, press **Eject** to open the lid of the cartridge entrance port again and then remove the cartridge.

Important: Used QIAstat-Dx Gastrointestinal Panel 2 Cartridges must be discarded. It is not possible to re-use cartridges for tests for which the execution was started but then subsequently canceled by the operator, or for which an error was detected.

17. After the QIAstat-Dx Gastrointestinal Panel 2 has been ejected, the results Summary screen will appear. Refer to "Interpretation of Results" on the facing page for further details. To begin the process for running another test, press **Run Test**.

Note: For further information on the use of the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0, refer to the *QIAstat-Dx Analyzer 1.0 User Manual* or the *QIAstat-Dx Analyzer 2.0 User Manual*.

Interpretation of Results

Viewing results

The QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0 automatically interprets and saves test results. After ejecting the QIAstat-Dx Gastrointestinal Panel 2 Cartridge, the results Summary screen is automatically displayed ().

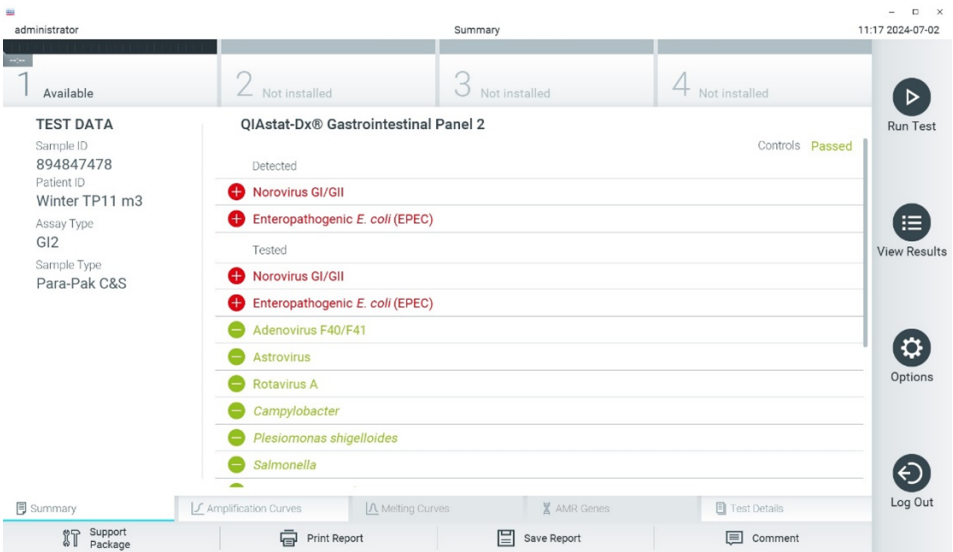






Figure 16. QIAstat-Dx Analyzer 2.0 screen.

Figure 16 shows the screen for the QIAstat-Dx Analyzer 2.0.

QIAstat-Dx Analyzer 2.0 includes an additional tab ARM Genes which is disabled for QIAstat-Dx Gastrointestinal Panel 2.

Note: From this point forward, example screen shots will be used when referring to the QIAstat-Dx Analyzer 1.0 and/or QIAstat-Dx Analyzer 2.0 where the functions being explained are the same.

The main part of the screen provides the following lists and uses color-coding and symbols to indicate the results:

- The first list, under the heading **Detected**, includes all pathogens detected and identified in the sample, which are preceded by a  sign and are colored red.
- The second list, under the heading **Tested** is includes all pathogens tested in the sample. Pathogens detected and identified in the sample are preceded by a  sign and are colored red. Pathogens that were tested but not detected are preceded by a  sign and are colored green. Invalid and not applicable pathogens are also displayed in this list.
- The third, under the heading **Tested**, includes all pathogens tested in the sample. Pathogens detected and identified in the sample are preceded by a  sign and are colored green. Invalid pathogens are also displayed in this list.

Note: Pathogens detected and identified in the sample are shown in both the **Detected** and **Tested** lists.

If the test failed to complete successfully, a message will indicate **Failed** followed by the specific Error Code.

The following Test Data is shown on the left side of the screen:

- Sample ID
- Patient ID (if available)


- Assay Type
- Sample Type

Further data about the assay is available, depending on the operator's access rights, through the tabs at the bottom of the screen (e.g., amplification plots and test details).

A report with the assay data can be exported to an external USB storage device. Insert the USB storage device into one of the USB ports of the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 and press **Save Report** in the bottom bar of the screen. This report can be exported later at any time by selecting the test from the **View Result List**.

The report can also be sent to the printer by pressing **Print Report** in the bottom bar of the screen.

Viewing amplification curves

To view test amplification curves of pathogens detected, press the  **Amplification Curves** tab (Figure 17).

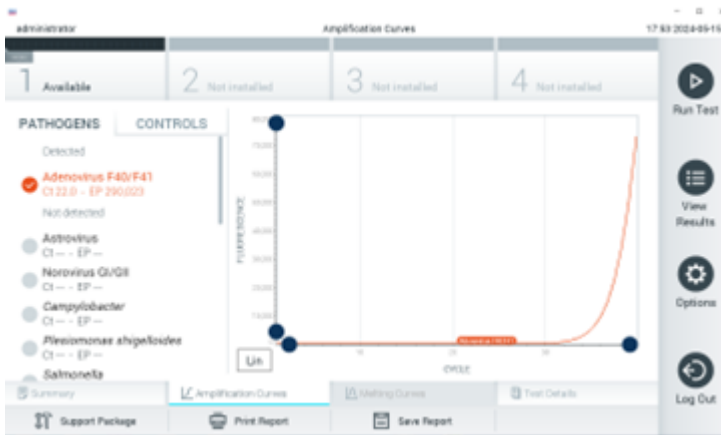


Figure 17. Amplification Curves screen (PATHOGENS tab).

Details about the tested pathogens and controls are shown on the left and the amplification curves are shown in the center.

Note: If User Access Control is enabled on the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0, the **Amplification Curves** screen is only available for operators with access rights.

Press the **PATHOGENS** tab on the left side to display the plots corresponding to the tested pathogens. Press on the pathogen name to select which pathogens are shown in the amplification plot. It is possible to select single, multiple, or no pathogens. Each pathogen in the selected list will be assigned a color corresponding to the amplification curve associated with the pathogen. Unselected pathogens will be shown in gray.

The corresponding C_T and endpoint fluorescence (EP) values are shown below each pathogen name.

Press the **CONTROLS** tab on the left side to view the controls in the amplification plot. Press the circle next to the control name to select or deselect it (Figure 18).



Figure 18. Amplification Curves screen (CONTROLS tab).

The amplification plot displays the data curve for the selected pathogens or controls. To alternate between logarithmic or linear scale for the Y-axis, press the **Lin** or **Log** button at the bottom left corner of the plot.

The scale of the X-axis and Y-axis can be adjusted using the **blue pickers** on each axis. Press and hold a blue picker and then move it to the desired location on the axis. Move a blue picker to the axis origin to return to the default values.

Viewing test details

Press **Test Details** in the Tab Menu bar at the bottom of the touchscreen to review the results in more detail. Scroll down to see the complete report.

The following Test Details are shown in the center of the screen (Figure 19):

- User ID
- Cartridge SN (serial number)
- Cartridge Expiration Date
- Module SN
- Test Status (Completed, Failed or Canceled by operator)
- Error Code (if applicable)
- Test Start Date and Time
- Test Execution Time
- Assay Name
- Test ID
- Test Result
 - **Positive** (if at least one gastrointestinal pathogen is detected/identified)
 - **Positive with warning** (if at least one pathogen is detected, but the Internal Control failed)
 - **Negative** (if no gastrointestinal pathogen is detected)
 - **Failed** (an error occurred or the test was canceled by the user)
- List of analytes tested in the assay, with C_T and endpoint fluorescence in the event of a positive signal
- Internal Control, with C_T and endpoint fluorescence

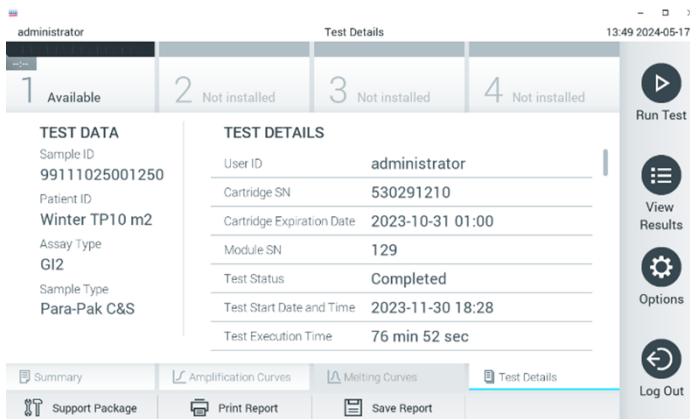


Figure 19. Example screen showing Test Data on the left panel and Test Details in the main panel.

Browsing results from previous tests

To view results from previous tests that are stored in the results repository, press **View Results** on the Main Menu bar (Figure 20).

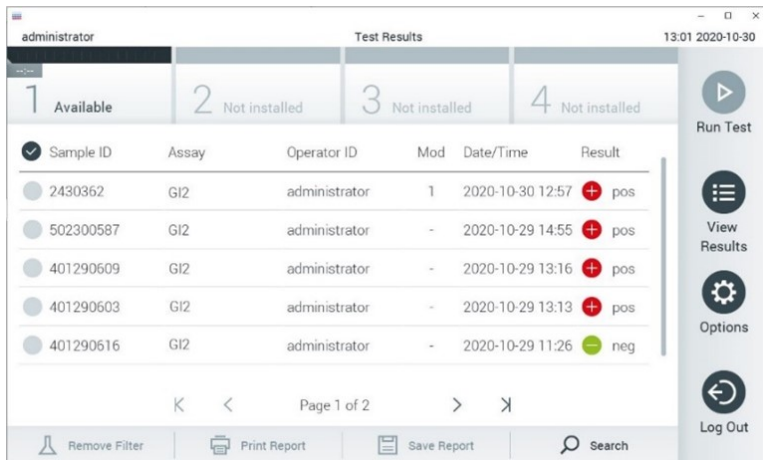



Figure 20. Example View Results screen.

The following information is available for every executed test (Figure 21):

- Sample ID
- Assay (name of test assay which is “GI2” for Gastrointestinal Panel 2)
- Operator ID
- Mod (Analytical Module on which the test was executed)
- Date/Time (date and time when the test was finished)
- Result (outcome of the test: positive [pos], negative [neg], failed [fail] or successful [suc])

Note: If User Access Control is enabled on the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0, the data for which the user has no access rights will be hidden with asterisks.

Select one or more test results by pressing the gray circle to left of the sample ID. A checkmark will appear next to selected results. Unselect test results by pressing this checkmark. The entire list of results can be selected by pressing the  **checkmark** in the top row (Figure 21).

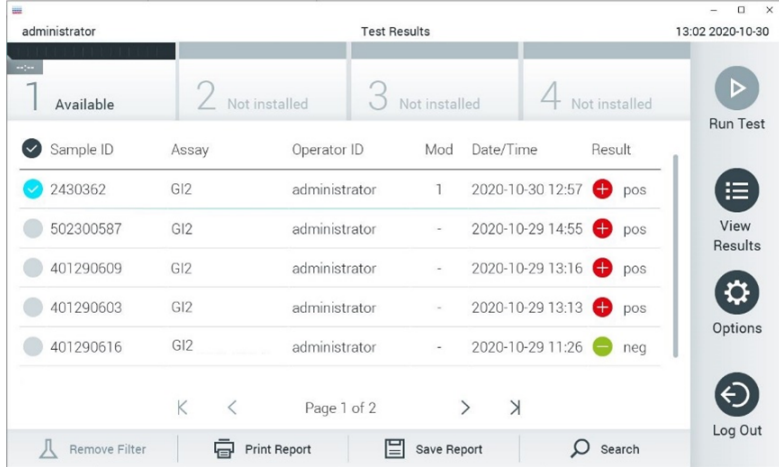


Figure 21. Example of selecting Test Results in the View Results screen.

Press anywhere in the test row to view the result for a particular test.

Press a column headline (e.g., Sample ID) to sort the list in ascending or descending order according to that parameter. The list can be sorted according to only one column at a time.

The Result column shows the outcome of each test (Table 2).

Table 2. Descriptions of the test results in View Results Screen






Outcome	Result	Description	Action
Positive	 pos	At least one pathogen is positive	Refer to the Summary Result Screen or Result Printout for pathogen specific results. Description of pathogen results can be found in Table 5.

Table 2. Descriptions of the test results in View Results Screen (continued)

Outcome	Result	Description	Action
Positive with warning	 !pos*	At least one pathogen is positive, but the Internal Control failed	Refer to the Summary Result Screen or Result Printout for pathogen specific results. Description of pathogen results can be found in Table 5.
Negative	 neg	No analytes were detected	Refer to the Summary Result Screen or Result Printout for pathogen specific results. Description of pathogen results can be found in Table 5.
Failed	 fail	The test failed because either an error occurred, the test was canceled by the user, or no pathogens were detected and the internal control failed.	Repeat the test using a new cartridge. Accept the results of the repeat testing. If the error persists, contact QIAGEN Technical Services for further instructions.
Successful	 Suc	The test is either positive or negative, but the user does not have the access rights to view the test results.	Login from a user profile with rights to view the results.

Press **Save Report** to save the report(s) for the selected result(s) in PDF format to an external USB storage device.

Select the report type: **List of Tests** or **Test Reports**.

Press **Search** to search the test results by Sample ID, Assay, and Operator ID. Enter the search string using the virtual keyboard and press **Enter** to start the search. Only the records containing the search text will be displayed in the search results.

If the results list has been filtered, the search will only apply to the filtered list.

Press and hold a column headline to apply a filter based on that parameter. For some parameters, such as Sample ID, the virtual keyboard will appear so the search string for the filter can be entered.

For other parameters, such as Assay, a dialog will open with a list of assays stored in the repository. Select one or more assays to filter only the tests that were performed with the selected assays.

The  symbol to the left of a column headline indicates that the column's filter is active.

A filter can be removed by pressing Remove Filter in the Submenu bar.

Exporting results to a USB drive

From any tab of the View Results screen, select **Save Report** to export and save a copy of the test results in PDF format to a USB drive. The USB port is located on the front of the QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0.

Backup and regular data upload to Sharepoint

The results can be exported from the instrument following these steps:

1. Press the **Options > System Configuration > System Backup** (Figure 22). Insert a USB storage device into the front USB port.
2. Press **Perform Backup**. A file with the extension .dbk will be generated in the USB with a default file name.

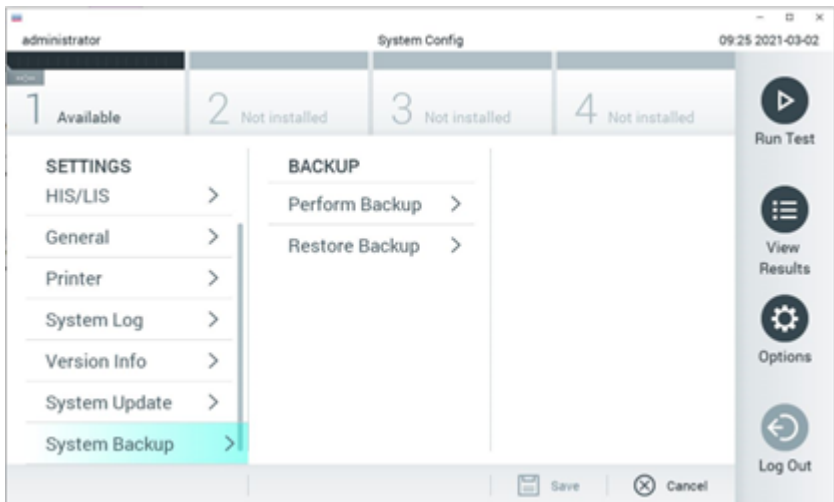


Figure 22. Perform a backup.

Printing results

Make sure a printer is connected to the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 and the proper driver is installed. Press **Print Report** to send a copy of the PDF test results to the printer.

Sample result interpretation

Note: STEC, *E. coli* O157, and EPEC results are not reported for stool specimens collected in FecalSwab transport media.

For samples in FecalSwab collection device, a result for a gastrointestinal organism is interpreted as “Positive” when the corresponding PCR assay is positive. EPEC, STEC, and *E. coli* O157 targets are not applicable and will not appear in the results section.

For samples in Para-Pak C&S collection device, a result for a gastrointestinal organism is interpreted as “Positive” when the corresponding PCR assay is positive except for EPEC, STEC and *E. coli* O157. The result interpretation for EPEC, STEC and *E. coli* O157 follows the rationale explained in Table 3.

Table 3. Interpretation of EPEC, STEC, and *E. coli* O157 results only applicable for Para-Pak C&S samples.

EPEC Result	STEC stx1/stx2 Result	<i>E. coli</i> O157 Result	Description
Negative	Negative	N/A	Enteropathogenic <i>E. coli</i> (EPEC) was not detected and Shiga-like toxin-producing <i>E. coli</i> (STEC) stx1/stx2 is negative as both stx1 and stx2 have not been detected. <i>E. coli</i> O157 result is not applicable (N/A) when STEC stx1/stx2 is not detected due to <i>E. coli</i> O157 being a specific serotype of STEC.
Positive	Negative	N/A	EPEC was detected and STEC stx1/stx2 is negative as both stx1 and stx2 have not been detected. <i>E. coli</i> O157 result is not applicable (N/A) when STEC stx1/stx2 is not detected due to <i>E. coli</i> O157 being a specific serotype of STEC.
N/A	Positive	Negative	EPEC result is not applicable because EPEC detection cannot be differentiated when STEC stx1 or stx2 is detected. <i>E. coli</i> O157 was not detected.
N/A	Positive	Positive	EPEC result is not applicable because EPEC detection cannot be differentiated when STEC stx1 or stx2 is detected. <i>E. coli</i> O157 was detected.

For both sample collection devices, internal control results are to be interpreted according to Table 4.

Table 4. Interpretation of Internal Control results

Control result	Explanation	Action
Passed	The Internal Control amplified successfully	The run was completed with success. All results are valid and can be reported. Detected pathogens are reported as positive and undetected pathogens are reported as negative.
Failed	The Internal Control failed	Positively detected pathogen(s) are reported, but all negative results (tested but not detected pathogen[s]) are invalid. Repeat the testing using a new QIAstat-Dx Meningitis/Encephalitis Panel Cartridge.

The software provides an overall test result (Table 2) as well as a result for individual pathogens. Possible results for each organism include Detected/Positive, Not Detected/Negative, N/A, and Invalid (Table 5). If the internal control has failed and no positive signal was detected or if there is an instrument error, there will be no pathogen results provided.

Table 5. Description of Pathogen results as displayed on Summary Result Screen and the Result Printout.

Result	Symbol	Explanation	Action
Positive/ Detected		A positive signal was detected for this pathogen. Result of the Internal Control is passed.	None. Report results.
Positive/ Detected with Warning	pos*	A positive signal was detected for this pathogen, but the result of the internal control has failed.	Report positive analyte. Repeat the test using a new cartridge. Accept the results of the repeat testing. If the invalid result persists, contact QIAGEN Technical Services for further instructions.
Negative/ Not Detected		No signal was detected for this pathogen. The Internal Control passed.	None. Report results.

Table 5. Description of Pathogen results as displayed on Summary Result Screen and the Result Printout. (continued)

Result	Symbol	Explanation	Action
N/A (applies to E. coli O157 and EPEC only)		The run was successfully completed and the Internal Control passed. For E. coli O157 N/A: Shiga-like toxin-producing E. coli (STEC) was not detected. For EPEC N/A: Shiga-like toxin producing E. coli (STEC) was detected.	None. Report results.
Invalid		No signal was detected for this pathogen and the Internal Control failed (but other pathogens have been detected).	Repeat the test using a new cartridge. Accept the results of the repeat testing. If the invalid result persists, contact QIAGEN Technical Services for further instructions.

Quality Control

Internal control interpretation

The QIAstat-Dx Gastrointestinal Panel 2 Cartridge includes a full process Internal Control, which is titered *Schizosaccharomyces pombe*. *Schizosaccharomyces pombe* is a yeast (fungi) that is included in the cartridge in dried form and is rehydrated upon sample loading. This Internal Control material verifies all steps of the analysis process, including sample homogenization, lysis of viral and cellular structures (by means of chemical and mechanical disruption), nucleic acid purification, reverse transcription, and real-time PCR.

A passed result for the Internal Control indicates that all processing steps performed by the QIAstat-Dx Gastrointestinal Panel 2 Cartridge were successful.

A failed result of the Internal Control does not negate any positive results for detected and identified targets, but it does invalidate all negative results in the analysis. Therefore, the test should be repeated if the Internal Control signal is negative.

External control information

All external quality control requirements and testing should be performed in accordance with local, state, and federal regulations or accreditation organizations and should follow the user's laboratory standard quality control procedures.

Blank controls are not applicable to the device because it is a single test disposable cartridge. Regular testing of negative and positive external controls is recommended by the company but controls are not provided with the QIAstat-Dx Gastrointestinal Panel 2. Use transport media as the external Negative Control, and previously characterized positive samples or negative sample spiked with well characterized target organisms as external positive controls.

Limitations

- The QIAstat-Dx Gastrointestinal Panel 2 is intended for professional use only and is not intended for self-testing. The QIAstat-Dx Gastrointestinal Panel 2 is intended for *in vitro* diagnostic use.
- Results from the QIAstat-Dx Gastrointestinal Panel 2 are not intended to be used as the sole basis for diagnosis, treatment or other patient management decisions.
- All assay results should be used and interpreted by a trained healthcare professional in the context of a full clinical evaluation, laboratory and epidemiological findings, as an aid in the diagnosis of gastrointestinal infection.
- The performance of this test has not been determined for patients without signs and symptoms of gastrointestinal illness.
- The QIAstat-Dx Gastrointestinal Panel 2 is not intended for testing of samples other than those described in this Instructions for Use (IFU). The performance of this test has only been validated with human stool preserved in transport medium (Para-Pak C&S or FecalSwab), according to the media manufacturers' instructions. It has not been validated for use with other stool transport media, rectal swabs, raw stool, vomitus, or endoscopy stool aspirates. The QIAstat-Dx Gastrointestinal Panel 2 should not be used to test Para-Pak C&S or FecalSwab collection devices that have been overfilled with stool. Only stool resuspended following the collection device manufacturer's instructions should be used. The overfilling of Para-Pak C&S or FecalSwab collection devices can result in a failed test with an error indicating 'Sample concentration too high'.
- The detection of viral, bacterial, or parasitic sequences is dependent upon proper specimen collection, handling, transportation, storage, and preparation (including extraction). Failure to observe proper procedures in any one of these steps can lead to incorrect results. There is a risk of false negative values resulting from improperly collected, transported, or handled specimens.

- Positive results do not rule out co-infection with organisms not included in the QIAstat-Dx Gastrointestinal Panel 2. The agent detected may not be the definitive cause of the disease.
- Not all agents of acute gastrointestinal infection are detected by this assay.
- The QIAstat-Dx Gastrointestinal Panel 2 is intended to be used in conjunction with standard of care culture for organism recovery, serotyping and/or antimicrobial susceptibility testing where applicable.
- The QIAstat-Dx Gastrointestinal Panel 2 can be used only with the QIAstat-Dx Analyzer 1.0 and the QIAstat-Dx Analyzer 2.0.
- The identification of multiple diarrheagenic *E. coli* pathotypes has historically relied upon phenotypic characteristics, such as adherence patterns or toxigenicity in certain tissue culture cell lines (27). The QIAstat-Dx Gastrointestinal Panel 2 targets genetic determinants characteristic of most pathogenic strains of these organisms but may not detect all strains having phenotypic characteristics of a pathotype.
- Genetic virulence markers associated with diarrheagenic *E. coli/Shigella* pathotypes are often carried on mobile genetic elements (MGEs) that can be transferred horizontally between different strains (27), therefore “Detected” results for multiple diarrheagenic *E. coli/Shigella* may be due to co-infection with multiple pathotypes or, less frequently, may be due to the presence of a single organism containing genes characteristic of multiple pathotypes. An example of the latter is the 2019 *E. coli* hybrid ETEC/STEC strains found in Sweden (73).
- The QIAstat-Dx Gastrointestinal Panel 2 detects Enteropathogenic *E. coli* (EPEC) in Para-Pak C&S collected samples through targeting of the *eae* gene, which encodes the adhesin intimin. As some Shiga-like toxin-producing *E. coli* (STEC) also carry *eae* (in particular, strains identified as enterohemorrhagic *E. coli*; EHEC) (27), the QIAstat-Dx Gastrointestinal Panel 2 cannot distinguish between STEC containing *eae* and a co-infection of EPEC and STEC. Therefore, the EPEC result is not applicable (N/A) and not reported for specimens in

which STEC has also been detected. In rare cases, STEC may be reported as EPEC when an STEC carrying *eae* (EHEC) is present in a specimen below the LoD of the STEC oligonucleotide design(s). Rare instances of other organisms carrying *eae* have been documented; e.g., *Escherichia albertii*, and *Shigella boydii* (74).

- *Shigella dysenteriae* serotype 1 possess a shiga toxin gene (*stx*) that is identical to the *stx1* gene of STEC (27) . *Stx* genes have been more recently found in other *Shigella* species (e.g., *S. sonnei* and *S. flexneri*) (75,76) . The detection of both *Shigella*/Enteroinvasive *E. coli* (EIEC) and STEC *stx1/stx2* analytes in the same specimen may indicate the presence of *Shigella* species such as *S. dysenteriae*. Rare instances of the detection of Shiga-like toxin genes in other genera/species have been reported, e.g., *Acinetobacter haemolyticus*, *Enterobacter cloacae*, and *Citrobacter freundii* (77,78,79).
- Results for STEC, *E. coli* O157, and EPEC are not reported for stool specimens preserved in FecalSwab transport media.
- *E. coli* O157 result is only reported as specific serogroup identification in association with STEC *stx1/stx2* in Para-Pak C&S collected samples. While non-STE C O157 strains have been detected in human stool (80), their role in disease has not been established (81). Serotype O157 EPEC have been identified and will be detected by the QIAstat-Dx Gastrointestinal Panel 2 (by the EPEC oligonucleotides design) due to their carriage of the *eae* gene.
- The QIAstat-Dx Gastrointestinal Panel 2 cannot distinguish between infections with a single toxigenic STEC O157 or rare co-infections of STEC (non-O157) with a *stx1/stx2*-negative *E. coli* O157 in Para-Pak C&S samples.
- This test only detects *Campylobacter jejuni*, *C. coli* and *C. upsaliensis* and does not differentiate between these three species of *Campylobacter*. Additional testing is required to differentiate between these species and to detect other *Campylobacter* species that may be present in stool specimens. In particular, the *Campylobacter upsaliensis* oligonucleotides design may cross-react with the *Campylobacter* species *C. lari* and *C.*

helveticus organisms.

- Negative results do not exclude the possibility of gastrointestinal infection. Negative test results may occur from sequence variants in the region targeted by the assay, the presence of inhibitors, technical errors, sample mix-ups, or an infection caused by an organism not detected by the panel. Test results may also be affected by use of certain medications (e.g., calcium carbonate), concurrent antimicrobial therapy or levels of organism in the sample that are below the limit of detection for the test. Sensitivity in some clinical settings may differ from that described in the Instructions for Use. Negative results should not be used as the sole basis for diagnosis, treatment, or other management decisions.
- Organism and amplicon contamination may produce erroneous results for this test. Particular attention should be given to the Laboratory Precautions noted under the Laboratory Precautions section.
- There is a risk of false-positive values resulting from cross-contamination by target organisms, their nucleic acids or the amplified product, or from non-specific signals in the assay.
- There is a risk of false negative results due to the presence of strains with sequence variability in the target regions of the oligonucleotides design. Refer to the inclusivity testing section of this document for additional information.
- Not all *Salmonella* serotypes were tested in validation studies; however, representatives of the 20 most prevalent serotypes recently circulating in the US (CDC National *Salmonella* Surveillance Annual Summary 2016) were evaluated during analytical reactivity studies. In silico sequence analysis supports detection of all subspecies and serotypes of *Salmonella*.
- The performance of the QIAstat-Dx Gastrointestinal Panel 2 has not been established in individuals who received Rotavirus A vaccine. Recent oral administration of a Rotavirus A vaccine may cause positive results for Rotavirus A if the virus is passed in the stool.
- The performance of this test has not been evaluated for immunocompromised individuals.

- Underlying polymorphisms in primer-binding regions can affect the targets being detected and subsequently the test results returned.
- Positive and negative predictive values are highly dependent on prevalence. False negative test results are more likely when prevalence of disease is high. False positive test results are more likely when prevalence is low.
- The effect of interfering substances has only been evaluated for those listed in the labeling at its indicated amount or concentration. Interference by substances other than those described in the “Interfering Substances” section of the Instruction for Use can lead to erroneous results.
- Cross-reactivity with gastrointestinal tract organisms other than those listed in the “Analytical Specificity” section of the package insert may lead to erroneous results.
- The test is a qualitative test and does not provide the quantitative value of detected organism present.
- The assay sensitivity to detect *Cyclospora cayatanensis*, Adenovirus F40/F41, *Entamoeba histolytica* and the Shiga-like toxin-producing *Escherichia coli* (STEC) might be reduced up to 3.16-fold when using half-input sample volume (100 µL) workflow detailed in "Appendix C: Additional instructions for use" on page 136.
- Due to the small number of positive specimens collected for certain analytes during the prospective clinical study, performance characteristics for Adenovirus 40/41, ETEC, *Plesiomonas shigelloides*, *Shigella*/EIEC, STEC, *E. coli* 0157, *Yersinia enterocolitica*, *Cryptosporidium*, and *Giardia lamblia* were established additionally with retrospective clinical specimens. Performance characteristics for Astrovirus and *Entamoeba histolytica* were established primarily with contrived clinical specimens.
- If four or more distinct organisms are detected in a specimen, retesting is recommended to confirm the polymicrobial result.

- Virus, bacteria, and parasite nucleic acid may persist in vivo independently of organism viability. Additionally, some organisms may be carried asymptotically. Detection of organism targets does not imply that the corresponding organisms are infectious or are the causative agents for clinical symptoms.
- The performance of this test has not been established for monitoring treatment of infection with any of the panel organisms.
- For ETEC, the assay is not predicted to detect bacteria carrier of Heat-labile enterotoxin gene subtype LT-II and/or of Heat-stable enterotoxin gene variant Stb e.
- The assay is not predicted to detect Human Astrovirus types MLB1-3 and VA1-5.
- The potential for competitive inhibition at high concentrations between on-panel analytes was evaluated for a limited number of pathogens (See Table 14). The potential for competitive inhibition between other on-panel analytes is unknown.

Performance Characteristics

Analytical performance

Limit of detection

The Limit of Detection (LoD) is defined as the lowest concentration at which $\geq 95\%$ of the tested samples generate a positive call.

The LoD for each of the QIAstat-Dx Gastrointestinal Panel 2 target pathogenic organisms was assessed, using in total 48 pathogen strains, by analyzing serial dilutions of analytical samples prepared from culture isolates from commercial suppliers (e.g., ZeptoMetrix[®] and ATCC[®]), confirmed clinical isolates, or artificial samples for target analytes commercially unavailable. Each sample tested was prepared in human stool matrix, which consists of a pool of previously tested negative clinical stool specimens resuspended in Cary-Blair transport medium.

Each of the 48 strains was tested in human stool matrix prepared following the manufacturer's instructions for the Para-Pak C&S[®] collection device. A matrix equivalency study between Para-Pak C&S and FecalSwab transport media was conducted to support the conclusions in the section.

Individual LoD values for each QIAstat-Dx Gastrointestinal Panel 2 target is shown in Table 6.

Table 6. LoD values obtained for the different gastrointestinal target strains tested with the QIAstat-Dx Gastrointestinal Panel 2

Pathogen	Strain	Source	Concentration (molecular units)* copies/mL	Concentration (microbiological units)	Detection rate
<i>Campylobacter</i>	<i>Campylobacter coli</i> 76-GA2 [LMG 21266]	ATCC 43478	5802	1.2 CFU/mL	20/20
	<i>Campylobacter coli</i> CIP 7080	ATCC 33559	8941	0.6 CFU/mL	20/20
	<i>Campylobacter jejuni</i> Z086	ZeptoMetrix 801650	14491	1660 CFU/mL	20/20
	<i>Campylobacter jejuni</i> subsp. <i>Jejuni</i> RM3193	ATCC BAA-1234	7210	110 CFU/mL	19/20
	<i>Campylobacter upsaliensis</i> NCTC 11541	ZeptoMetrix 0801999	56165	2259.4 CFU/mL	20/20
	<i>Campylobacter upsaliensis</i> RM3195	ATCC BAA-1059	7631	35 CFU/mL	19/20
<i>Clostridium difficile</i> toxin A/B	(NAP1A) Toxinotype III A+ B+	ZeptoMetrix 0801619	11083	515 CFU/mL	19/20
	Toxinotype 0 A+ B+	ATCC 9689	101843	853.2 CFU/mL	20/20
<i>Plesiomonas shigelloides</i>	Z130	ZeptoMetrix 801899	481	2291 CFU/mL	20/20
	Bader	ATCC 14029	116	2.7 CFU/mL	19/20

Table 6. LoD values obtained for the different gastrointestinal target strains tested with the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Strain	Source	Concentration (molecular units)* copies/mL	Concentration (microbiological units)	Detection rate
<i>Salmonella</i>	<i>Salmonella enterica</i> Serovar choleraesuis	ATCC 13312	647	91.6 CFU/mL	20/20
	<i>Salmonella enterica</i> Serovar Typhimurium Z005	ZeptoMetrix 801437	1441	4518.8 CFU/mL	20/20
<i>Vibrio cholerae</i>	Z132; toxigenic	ZeptoMetrix 0801901	28298	13600 CFU/mL	20/20
	Z133; non-toxigenic	ZeptoMetrix 0801902	79749	54668 CFU/mL	20/20
<i>Vibrio parahaemolyticus</i>	EB 101	ATCC 17802	12862	1600 CFU/mL	20/20
	Z134	ZeptoMetrix 0801903	8904	143 CFU/mL	20/20
<i>Vibrio vulnificus</i>	329 [CDC B3547]	ATCC 33817	109131	260 CFU/mL	20/20
	324 [CDC B629]	ATCC 27562	2983	1905.1 CFU/mL	20/20
<i>Yersinia enterocolitica</i>	Z036	ZeptoMetrix 0801734	719	2070 CFU/mL	20/20
	subsp. enterocolitica NTCC 11175, Biotype 4, serotype 3	ATCC 700822	2496	120.1 CFU/mL	20/20

Table 6. LoD values obtained for the different gastrointestinal target strains tested with the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Strain	Source	Concentration (molecular units)* copies/mL	Concentration (microbiological units)	Detection rate
Enteroinvasive <i>E. coli</i> (EIEC)/ <i>Shigella</i>	<i>Shigella sonnei</i> NCDC 1120-66	ATCC 25931	488	0.2 CFU/mL	20/20
	<i>Escherichia coli</i> CDC EDL 1282, O29:NM	ATCC 43892	1431	41.3 CFU/mL	20/20
Enteropathogenic <i>E. coli</i> (EPEC)*	<i>Escherichia coli</i> O111:NM (EPEC)	ZeptoMetrix 0801747	1817	2581.7 CFU/mL	20/20
	<i>Escherichia coli</i> 7.1493; EPEC; O84:H28	Zeptomatrix 801938	29021	1190 CFU/mL	20/20
Enterotoxigenic <i>E. coli</i> (ETEC) lt/st	<i>Escherichia coli</i> H10407, O78:H11	ATCC 35401	367	10.1 CFU/mL	19/20
	<i>Escherichia coli</i> ETEC; ST+, LT+	ZeptoMetrix 801624	855	567 CFU/mL	20/20
Shiga-like toxin-producing <i>E. coli</i> (STEC) stx1/stx2*	<i>Escherichia coli</i> O26:H4	ZeptoMetrix 801748	2012	726.8 CFU/mL	20/20
Shiga-like toxin-producing <i>E. coli</i> (STEC) <i>E. coli</i> O157*	<i>Escherichia coli</i> O157:H7; EDL933	ZeptoMetrix 801622	1217	2281.5 CFU/mL	STEC stx1/stx2: 19/20 O157: 19/20
<i>Cryptosporidium</i>	<i>Cryptosporidium hominis</i>	Public Health Wales UKM 84	357 661	N/A N/A	20/20 20/20
	<i>Cryptosporidium parvum</i> – Iowa isolate	Waterborne® P102C			

Table 6. LoD values obtained for the different gastrointestinal target strains tested with the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Strain	Source	Concentration (molecular units)* copies/mL	Concentration (microbiological units)	Detection rate
<i>Cyclospora cayentensis</i>	N/A	LACNY- Clinical sample LAC2825	53	N/A	19/20
	N/A	LACNY Clinical sample LAC2827	137	N/A	20/20
<i>Entamoeba histolytica</i>	HM-1:IMSS (Mexico City 1967)	ATCC 30459	7	0.2 cells/mL	20/20
	HK-9 (Korea)	ATCC 30015	1	0.13 cells/mL	19/20
<i>Giardia lamblia</i>	WB (Bethesda)	ATCC 30957	11850	790 cells/mL	19/20
	Portland-1	ATCC 30888	14500	635 cells/mL	20/20
Adenovirus F40/F41	Type 40 (Dugan)	ZeptoMetrix 0810084CF	11726	0.1 TCID50/mL	20/20
	Type 41 (Tak)	ZeptoMetrix 0810085CF	979	0.05 TCID50/mL	19/20
Astrovirus	ERE IID 2371 (type 8)	Zeptomatrix 0810277CF	11586371	11.7 TCID50/mL	20/20
	ERE IID 2868 (type 4)	Zeptomatrix 0810276CF	52184	1.3 TCID50/mL	19/20
Norovirus GI/GII	GI.1 (recombinant)	ZeptoMetrix 0810086CF	24629 copies/mL	891.1 TCID50/mL	19/20
	GII.4 (recombinant)	ZeptoMetrix 0810087CF	8998 copies/mL	10.5 TCID50/mL	20/20
Rotavirus A	69M	ZeptoMetrix 0810280CF	5787	436.1 TCID50/mL	19/20
	Wa	ZeptoMetrix 0810041CF	5201	14.1 TCID50/mL	19/20

Table 6. LoD values obtained for the different gastrointestinal target strains tested with the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Strain	Source	Concentration (molecular units)* copies/mL	Concentration (microbiological units)	Detection rate
Sapovirus	Genogroup I, genotype 1	QIAGEN Barcelona -Clinical sample GI-88	187506	N/A	20/20
	Genogroup V	Universitat de Barcelona 160523351	3007	N/A	20/20

Exclusivity (analytical specificity)

The analytical specificity study was carried out by *in vitro* testing and *in silico* analysis to assess the potential cross-reactivity and exclusivity of the QIAstat-Dx Gastrointestinal Panel 2. On-panel organisms were tested to assess the potential for intra-panel cross-reactivity and Off-panel organisms were tested to evaluate cross-reactivity with organisms not covered by the panel content. The On-panel and Off-panel organisms tested are shown in Table 8 and Table 9, respectively.

Samples were prepared by single spiking organisms into negative stool resuspended in Cary-Blair at the highest concentration possible based on the organism stock, preferably at 10⁵ TCID₅₀/mL for viral, 10⁵ cells/mL for parasite targets and 10⁶ CFU/mL for bacterial targets. The pathogens were tested in 3 replicates. There was no intra-panel or Off-panel cross-reactivity for all pathogens tested L, except for two non-targeted L species (*C. helveticus* and *C. lari*) that cross-reacted with the Campylobacter assay oligonucleotides included in the QIAstat-Dx Gastrointestinal Panel 2.

Table 7. List of Analytical Specificity on-panel pathogens tested.

Type	Pathogen	
Bacteria	<i>Campylobacter coli</i>	<i>Plesiomonas shigelloides</i>
	<i>Campylobacter jejuni</i>	<i>Salmonella enterica</i>
	<i>Campylobacter upsaliensis</i>	<i>Shigella sonnei</i>
	<i>Clostridium difficile</i>	<i>Vibrio cholerae</i>
	<i>Escherichia coli</i> (EAEC)	<i>Vibrio parahaemolyticus</i>
	<i>Escherichia coli</i> (EPEC)*	<i>Vibrio vulnificus</i>
	<i>Escherichia coli</i> (ETEC)	<i>Yersinia enterocolitica</i>
	<i>Escherichia coli</i> (STEC)*	
Parasites	<i>Cryptosporidium parvum</i>	<i>Entamoeba histolytica</i>
	<i>Cyclospora cayetanensis</i>	<i>Giardia lamblia</i>
Viruses	Adenovirus F41	Norovirus GII
	Astrovirus	Rotavirus A
	Norovirus GI	Sapovirus

Table 8. List of Analytical Specificity off-panel pathogens tested

Type	Pathogen (potential cross-reactant)	
Bacteria	<i>Abiotrophia defectiva</i>	<i>Enterobacter aerogenes</i>
	<i>Acinetobacter baumannii</i>	<i>Enterobacter cloacae</i>
	<i>Aeromonas hydrophila</i>	<i>Enterococcus faecalis</i>
	<i>Arcobacter cryaerophilus</i>	<i>Enterococcus faecium</i>
	<i>Bacillus subtilis</i>	<i>Escherichia fergusonii</i>
	<i>Bifidobacterium bifidum</i>	<i>Escherichia hermannii</i>
	<i>Campylobacter fetus</i>	<i>Escherichia vulneris</i>
	<i>Campylobacter gracilis</i>	<i>Faecalibacterium prausnitzii</i>
	<i>Campylobacter helveticus</i> (Cross-reactive for <i>Campylobacter</i> target)	<i>Gardnerella vaginalis</i>
	<i>Campylobacter hominis</i>	<i>Haemophilus influenzae</i>
	<i>Campylobacter lari</i> (Cross-reactive for <i>Campylobacter</i> target)	<i>Helicobacter pylori</i>
	<i>Campylobacter mucosalis</i>	<i>Klebsiella pneumoniae</i>
	<i>Campylobacter rectus</i>	<i>Lactobacillus casei</i>
	<i>Chlamydia trachomatis</i>	<i>Listeria monocytogenes</i>
	<i>Citrobacter freundii</i>	<i>Proteus mirabilis</i>
	<i>Clostridium difficile</i> non-toxigenic	<i>Proteus vulgaris</i>
	<i>Clostridium perfringens</i>	<i>Pseudomonas aeruginosa</i>
	<i>Clostridium septicum</i>	<i>Staphylococcus aureus</i>
	<i>Clostridium tetani</i>	<i>Staphylococcus aureus</i> subsp. <i>Aureus</i>
	<i>Corynebacterium genitalium</i>	<i>Staphylococcus epidermidis</i>
Fungi	<i>Aspergillus fumigatus</i>	<i>Saccharomyces boulardii</i>
	<i>Candida albicans</i>	<i>Saccharomyces cerevisiae</i>
Parasites	<i>Babesia microti</i>	<i>Toxoplasma gondii</i>
	<i>Blastocystis hominis</i>	<i>Trichomonas tenax</i>
	<i>Giardia muris</i>	

Table 8. List of Analytical Specificity off-panel pathogens tested (continued)

Type	Pathogen (potential cross-reactant)	
Viruses	Adenovirus C:2	Coronavirus 229E
	Adenovirus B:34	Coxsackievirus B3
	Adenovirus B3	Cytomegalovirus
	Adenovirus E:4a	Enterovirus 6 (Echovirus)
	Adenovirus serotype 1	Enterovirus 68
	Adenovirus serotype 5	Herpes Simplex Virus Type 2
	Adenovirus serotype 8	Rhinovirus 1A
	Bocavirus Type 1	

In silico predictions of potential cross-reactions showed that the following cross-reactions may occur when testing stool samples with the QIAstat-Dx Gastrointestinal Panel 2 (Table 9).

Table 9. Potential cross-reactions based on *in silico* analysis.

QIAstat-Dx Gastrointestinal Panel 2 Target	Potential cross-reactive organisms
Enteropathogenic <i>E. coli</i> (EPEC)*	<i>Shigella boydii</i> †‡§, <i>Escherichia albertii</i> †‡
<i>Campylobacter</i> spp.	<i>Campylobacter lari</i> §, <i>Campylobacter helveticus</i> §
Shiga-like toxin-producing <i>E. coli</i> (STEC) stx1/stx2*	<i>Shigella sonnei</i> †§, <i>Shigella dysenteriae</i> †§, <i>Acinetobacter haemolyticus</i> †**, <i>Citrobacter freundii</i> †**, <i>Enterobacter cloacae</i> †**, <i>Aeromonas caviae</i> †**, <i>Escherichia albertii</i> †¶
<i>E. coli</i> O157*	Non-STEC <i>E. coli</i> O157 strains††

* Only applicable for samples with Para-Pak C&S collection device.

† Note that these predicted cross-reactivity identified by *in silico* analysis reflects sequences which can be acquired between species by horizontal gene transfer(27,82)

‡ Rare or less common eae intimin carrier organisms (74).

§ On-panel target.

¶ Rare or less common Stx toxins producers (77,83,84,85,86,87)

Table 9. Potential cross-reactions based on in silico analysis. (continued)

QIAstat-Dx Gastrointestinal Panel 2 Target	Potential cross-reactive organisms
** <i>In vitro</i> testing of <i>Campylobacter lari</i> and <i>Campylobacter helveticus</i> strains at high concentration confirmed potential cross-reactivity of these <i>Campylobacter</i> species with the QIAstat-Dx Gastrointestinal Panel 2 assay.	
†† <i>E. coli</i> O157 will only be reported by the QIAstat-Dx Gastrointestinal Panel 2 when there is a positive amplification for the <i>E. coli</i> (STEC) design according to the calling algorithm. An infrequent case of an <i>E. coli</i> (STEC) and an <i>E. coli</i> O157 co-infection will not be differentiated from a single infection caused by an STEC O157:H7 strain.	

Inclusivity (analytical reactivity)

Analytical Reactivity (Inclusivity) was evaluated with gastrointestinal pathogen isolates/strains that were selected based on clinical relevance and genetic, temporal and geographical diversity. Samples were prepared by spiking organisms into negative stool matrix resuspended in Para-Pak C&S transport media. Based on in vitro (wet) testing and in silico analysis, the QIAstat-Dx Gastrointestinal Panel 2 primers and probes are specific and inclusive for clinically prevalent and relevant strains for each pathogen tested.

***In vitro* (wet) testing**

QIAstat-Dx Gastrointestinal Panel 2 is inclusive for 100% (114 out of 114) of the pathogen strains tested in vitro. Most pathogen strains evaluated in wet testing were detected at ≤ 3 -fold (104/114), of the corresponding LoD reference strain. Less than 100% detection was observed for one strain each of ETEC, EIEC/*Shigella* and Rotavirus and two strains each of STEC (one STEC O157), Adenovirus and Norovirus at 3x LoD. Testing of these strains at 10x LoD generated the expected positive result for all replicates (Table 10).

Inclusivity test results for all the pathogens tested with the QIAstat-Dx Gastrointestinal Panel 2 Assay. LoD reference strain for every pathogen is written in bold

Table 10a. Inclusivity test results for *Campylobacter* strains.

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
<i>Campylobacter</i>	<i>Campylobacter coli</i>	76-GA2 [LMG 21266]	ATCC	43478	1x LoD
	<i>Campylobacter coli</i>	Z293	ZeptoMetrix	0804272	1x LoD
	<i>Campylobacter coli</i>	CIP 7080 [1407, CIP 70.80]	ATCC	33559*	3x LoD
	<i>Campylobacter jejuni</i>	Z086	ZeptoMetrix	0801650*	1x LoD
	<i>Campylobacter jejuni</i>	subsp. jejuni RM3193	ATCC	BAA-1234*	0.1x LoD
	<i>Campylobacter jejuni</i> subsp. jejuni	O:19 HL7; D3180	ATCC	BAA-218	0.1x LoD
	<i>Campylobacter jejuni</i> subsp. jejuni	AS-83-79	ATCC	33291	0.1x LoD
	<i>Campylobacter jejuni</i> subsp. doylei	NCTC 11951	ATCC	49349	0.1x LoD
	<i>Campylobacter upsaliensis</i>	NCTC 11541	ZeptoMetrix	0801999*	1x LoD
	<i>Campylobacter upsaliensis</i>	RM 3195 (1994)	ATCC	BAA-1059*	0.3x LoD
<i>Campylobacter upsaliensis</i>	NCTC 11541 [C231]	ATCC	43954	1x LoD	

* Strain tested during LoD verification study.

Table 10b. Inclusivity test results for *Plesiomonas shigelloides* strains.

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Plesiomonas shigelloides	Plesiomonas shigelloides	Z130	ZeptoMetrix	0801899*	1x LoD
	Plesiomonas shigelloides	GNI 14	ATCC	51903	1x LoD
	Plesiomonas shigelloides	CDC 3085-55 [Bader M51, NCIB 9242, NCTC 10360, RH 798]	ATCC	14029*	0.3x LoD

*Strain tested during LoD verification study.

Table 10c. Inclusivity test results for *Salmonella* strains.

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
<i>Salmonella</i>	<i>Salmonella enterica</i>	Serovar Typhimurium Z005	ZeptoMetrix	0801437*	1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Bareilly	NCTC	NC05745	1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar typhi, Z152	ZeptoMetrix	0801933	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Enteridis, CDC K-1891 [ATCC 25928]	ATCC	13076	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Infantis, MZ1479 [SARB27]	ATCC	BAA-1675	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Montevideo, G4639	ATCC	BAA-710	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Javiana	NCTC	NC06495	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Thompson	NCTC	NC08496	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Saintpaul	ATCC	9712	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Berta	NCTC	NC05770	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Salame, II NCTC 10310 [JT945, SS140/61]	ATCC	700151	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. diarizonae IIIb, 62	ATCC	29934	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. houtenae IV, CIP 82.32 [264.66]	ATCC	43974	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Indica VI, CIP 102501 [F. Kauffmann 1240]	ATCC	43976	0.1x LoD

Table 10c. Inclusivity test results for Salmonella strains. (continued)

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Agona, CDC 873 [CDC 11111-61]	ATCC	51957	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Muenchen, 54	ATCC	8388	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Oranienburg, E1093	ATCC	9239	0.1x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Paratyphi B var. Java, CDC 5	ATCC	51962	0.1x LoD
	<i>Salmonella enterica</i>	CIP 82.33 [1224.72]	ATCC	43975	0.3x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Choleraesuis, NCTC 5735 [1348, K.34]	ATCC	13312*	0.3x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Newport, C487-69	ATCC	27869	0.3x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, 4, 5, 12:7:-, serovar Typhimurium	NCTC	NC13952	0.3x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Braenderup	ATCC	700136	0.3x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Anatum	NCTC	NC05779	0.3x LoD
	<i>Salmonella enterica</i>	Subsp. arizonae IIIa, NCTC 7311 [CDAI 426]	ATCC	700156	0.3x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Heidelberg, [16]	ATCC	8326	0.3x LoD
	<i>Salmonella enterica</i>	Subsp. Enterica, serovar Mississippi, CDC 2012K-0487	ATCC	BAA-2739	0.3x LoD

* Strain tested during LoD verification study.

Table 10d. Inclusivity test results for *Yersinia enterocolitica* strains.

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
<i>Yersinia enterocolitica</i>	<i>Yersinia enterocolitica</i>	Z036	ZeptoMetrix	0801734*	1x LoD
	<i>Yersinia enterocolitica</i>	NTCC 11175, Biotype 4, serotype 3 (O:3)	ATCC	700822*	1x LoD
	<i>Yersinia enterocolitica</i>	33114 [CCUG 11291, CCUG 12369, CIP 80.27, DSM 4780, LMG 7899, NCTC 12982], Biovar 1, O:8	ATCC	9610	1x LoD
	<i>Yersinia enterocolitica</i>	O:9	ATCC	55075	3x LoD

* Strain tested during LoD verification study.

Table 10e. Inclusivity test results for Enteropathogenic *E. coli* (EPEC) strains. Only applicable for Para-Pak C&S samples

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Enteropathogenic <i>E. coli</i> (EPEC)	Enteropathogenic <i>E. coli</i> (EPEC)	O111:NM	ZeptoMetrix	0801747*	1x LoD
	Enteropathogenic <i>E. coli</i> (EPEC)	7.1493,O84:H28	ZeptoMetrix	0801938*	1x LoD
	Enteropathogenic <i>E. coli</i> (EPEC)	Stoke W,O111:K58 (B4):H-	ATCC	33780	1x LoD

* Strain tested during LoD verification study.

Table 10f. Inclusivity test results for Enterotoxigenic *E. coli* (EPEC) strains

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Enterotoxigenic <i>E. coli</i> (EPEC) It/st	Enterotoxigenic <i>E. coli</i> (EPEC) It/st	ST+, LT+	ZeptoMetrix	0801624*	1x LoD
	Enterotoxigenic <i>E. coli</i> (EPEC) It/st	H10407,O78:H11,LT (+)/ctx A11(+)	ATCC	35401*	0.3x LoD
	Enterotoxigenic <i>E. coli</i> (EPEC) It/st	O27:H7,ST (+)/LT (-)	SSI Diagnostica	82173	0.1x LoD
	Enterotoxigenic <i>E. coli</i> (EPEC) It/st	O115:H15,ST (+)/LT (-)	SSI Diagnostica	82174	3x LoD
	Enterotoxigenic <i>E. coli</i> (EPEC) It/st	O169:H-,ST (-)/LT (+)	SSI Diagnostica	82172	10x LoD†

* Strain tested during LoD verification study.

† Testing at a lower concentration resulted in a detection rate of <100%.

Table 10g. Inclusivity test results for Enteroinvasive *E. coli* (EIEC)/*Shigella* strains.

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Enteroinvasive <i>E. coli</i> (EIEC)/ <i>Shigella</i>	Enteroinvasive <i>E. coli</i> (EIEC)	CDC EDL 1282, O29:NM	ATCC	43892*	1x LoD
	Enteroinvasive <i>E. coli</i> (EIEC)	O172:H-	SSI Diagnostica	82171	3x LoD
	<i>Shigella sonnei</i>	NCDC 1120-66	ATCC	25931*	1x LoD
	<i>Shigella boydii</i> (Serogroup C)	Z131	ZeptoMetrix	0801900	1x LoD
	<i>Shigella flexneri</i> (Serogroup B)	AMC 43-G-68 [EVL 82, M134]	ATCC	9199	1x LoD
	<i>Shigella flexneri</i> (Serogroup B)	Z046	ZeptoMetrix	0801757	1x LoD
	<i>Shigella sonnei</i> (Serogroup D)	WR AIR 1 virulent	ATCC	29930	1x LoD
	<i>Shigella sonnei</i> (Serogroup D)	Z004	ZeptoMetrix	0801627	3x LoD
	<i>Shigella boydii</i> (Serogroup C)	AMC 43-G-58 [M44 (Type 170)]	ATCC	9207	10x LoD†

* Strain tested during LoD verification study

† Testing at a lower concentration resulted in a detection rate of <100%.

Table 10h. Inclusivity test results for Shiga-like toxin-producing *E. coli* (STEC)(stx1/stx2-carrier strains). Only applicable for Para-Pak C&S samples

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1/2	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1/stx2	O157:H7; EDL933	ZeptoMetrix	0801622*	1x LoD

Table 10h. Inclusivity test results for Shiga-like toxin-producing *E. coli* (STEC)(stx1/stx2-carrier strains). Only applicable for Para-Pak C&S samples (continued)

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1	O26:H4, stx1 (+)	ZeptoMetrix	0801748*	1 x LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1/stx2	Reference ATCC® 35150 (EDL 931), O157:H7, stx1 (+), stx2 (+)	Microbiologics	617	3x LoD

Table 10h. Inclusivity test results for Shiga-like toxin-producing *E. coli* (STEC)(stx1/stx2-carrier strains). Only applicable for Para-Pak C&S samples (continued)

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1	Reference CDC 00-3039,O45:H2,unknown	Microbiologics	1098	1x LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1	O103:H2,stx1 (+)	SSI Diagnostica	82170	3x LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1/stx2	O22:H8,stx1c (+), stx2b (+)	SSI Diagnostica	91350	1x LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx2	O92,O107:K+:H48,stx2d (+)	SSI Diagnostica	91352	10x LoD†
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx2	O101:K32:H,stx2e (+)	SSI Diagnostica	91354	0.3x LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1/stx2	O128ac:H,stx2f (+)	SSI Diagnostica	91355	10x LoD†
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx2	O26:H11,stx2a (+)	SSI Diagnostica	95211	1x LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) - stx1	O8 ,stx1d (+)	SSI Diagnostica	91349	1x LoD

* Strain tested during LoD verification study

† Testing at a lower concentration resulted in a detection rate of <100%.

Table 10i. Inclusivity test results for Shiga-like toxin producing *E. coli* (STEC) stx1/stx2 O157 strains. Only applicable for Para-Pak C&S samples

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalogue ID	Times LoD
Shiga-like toxin producing <i>E. coli</i> (STEC) O157	Shiga-like toxin producing <i>E. coli</i> (STEC) - O157	O157:H7; EDL933	ZeptoMetrix	0801622*	1x LoD
	Shiga-like toxin producing <i>E. coli</i> (STEC) O157	O128ac:H; stx2f (+)	SSI Diagnostica	91355†	10x LoD‡
	Shiga-like toxin producing <i>E. coli</i> (STEC) O157	Reference ATCC 35150 (EDL 931), O157:H7, stx1 (+), stx2 (+)	Microbiologics	617	1x LoD

* Strain tested during LoD verification study.

† The *E. coli* strain 91355 from SSI Diagnostica is reported as following in its catalog: vtx2f+, eae+. However, it was found to amplify for *E. coli* O157 in both QIAstat-Dx and an FDA-cleared test .

‡ Testing at a lower concentration resulted in a detection rate of <100%.

Table 10j. Inclusivity test results for *Cryptosporidium* strains

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
<i>Cryptosporidium</i>	<i>Cryptosporidium parvum</i>	Iowa isolate	Waterborne	P102C*	1x LoD
	<i>Cryptosporidium hominis</i>	n/a	Public Health Wales	Clinical sample; UKM 84*	0.01x LoD
	<i>Cryptosporidium parvum</i>	–	ATCC	PRA-67DQ (isolated genomic DNA)	<0.01 LoD
	<i>Cryptosporidium meleagridis</i>	–	Public Health Wales	Clinical sample; UKMEL 14	<0.01 LoD

* Strain tested during LoD verification study.

Table 10k. Inclusivity test results for *Cyclospora cayetanensis* strains.

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalogue ID	Times LoD
<i>Cyclospora cayetanensis</i>	<i>Cyclospora cayetanensis</i>	n/a	Clinical sample	LAC2825*	1x LoD
	<i>Cyclospora cayetanensis</i>	n/a	Clinical sample	LAC2827*	1x LoD
	<i>Cyclospora cayetanensis</i>	–	ATCC	PRA-3000SD	1x LoD

* Strain tested during LoD verification study.

Table 10l. Inclusivity test results for *Entamoeba histolytica* strains

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalogue ID	Times LoD
<i>Entamoeba histolytica</i>	<i>Entamoeba histolytica</i>	HM-1:IMSS (Mexico City 1967)	ATCC	30459*	1x LoD
	<i>Entamoeba histolytica</i>	HK-9 (Korea)	ATCC	30015*	1x LoD
	<i>Entamoeba histolytica</i>	–	Vall d'Hebrón	Clinical sample; 1	1x LoD

* Strain tested during LoD verification study.

Table 10m. Inclusivity test results for *Giardia lamblia* strains

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalogue ID	Times LoD
<i>Giardia lamblia</i>	<i>Giardia lamblia</i>	Portland -1 (Portland, OR, 1971)	ATCC	30888*	1x LoD
	<i>Giardia lamblia</i>	WB (Bethesda, MD, 1979)	ATCC	30957*	1x LoD
	<i>Giardia intestinalis</i>	H3 isolate	Waterborne	P101	1x LoD

* Strain tested during LoD verification study.

Table 10n. Inclusivity test results for Adenovirus F40/F41 targets

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Adenovirus F40/F41	Human Adenovirus F41	Tak	ZeptoMetrix	0810085CF*	1x LoD
	Human Adenovirus F41	Tak (73-3544)	ATCC	VR-930	10x LoD†
	Human Adenovirus F40	Dugan [79-18025]	ATCC	VR-931	10x LoD†
	Human Adenovirus Type 40	Dugan	ZeptoMetrix	0810084CF*	3x LoD

* Strain tested during LoD verification study.

† Testing at a lower concentration resulted in a detection rate of <100%.

Table 10o. Inclusivity test results for Astrovirus strains

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Astrovirus	Human Astrovirus	ERE IID 2371 (type 8)	ZeptoMetrix	0810277CF*	1x LoD
	Human Astrovirus	HAsIV-1	Universitat de Barcelona	Clinical sample; 160521599	1x LoD
	Human Astrovirus	ERE IID 2868 (type 4)	ZeptoMetrix	0810276CF*	1x LoD
	Human Astrovirus	HAsIV-3	Universitat de Barcelona	Clinical sample; 151601306	1x LoD

* Strain tested during LoD verification study.

Table 10p. Inclusivity test results for Norovirus GI/GII strains

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Norovirus GI/GII	Human Norovirus Genogroup 1	Recombinant GI.1	ZeptoMetrix	0810086CF*	1x LoD
	Human Norovirus Genogroup 1	–	Indiana University Health	Clinical sample; IU3156	1x LoD
	Human Norovirus Genogroup 1	–	Indiana University Health	Clinical sample; IU3220	1x LoD
	Human Norovirus Genogroup 1	–	TriCore Reference Laboratories	Clinical sample; TC4274	3x LoD
	Human Norovirus Genogroup 2	Recombinant GII.4	ZeptoMetrix	0810087CF*	1x LoD
	Human Norovirus Genogroup 2	GII.2	Vall d'Hebrón	Clinical sample; 198058327	1x LoD
	Human Norovirus Genogroup 2	GII.4	Universitat de Barcelona	Clinical sample; N26.2TA	1x LoD
	Human Norovirus Genogroup 2	–	Lacny Hospital	Clinical sample; LAC2019	1x LoD
	Human Norovirus Genogroup 2	–	Nationwide Children's Hospital	Clinical sample; NWC6063	1x LoD
	Human Norovirus Genogroup 2	GII.6	QIAGEN Barcelona (STAT-Dx)	Clinical sample; GI 12	3x LoD
	Human Norovirus Genogroup 2	–	Lacny Hospital	Clinical sample; LAC2133	10x LoD#
	Human Norovirus Genogroup 2	–	Lacny Hospital	Clinical sample; LAC2074	10x LoD#

* Strain tested during LoD verification study.

Testing at a lower concentration resulted in a detection rate of <100%.

Table 10q. Inclusivity test results for Rotavirus A strains

QIAstat-Dx target	Pathogen	Strain	Supplier	Catalog ID	Times LoD
Rotavirus A	Human Rotavirus A	69M	ZeptoMetrix	0810280CF*	1x LoD
	Human Rotavirus A	Wa, G1P1A[8]	ZeptoMetrix	0810041CF*	1x LoD
	Human Rotavirus A	DS-1, G2P1B[4]	ATCC	VR-2550	1x LoD
	Human Rotavirus A	Va70	ZeptoMetrix	0810281CF	1x LoD
	Human Rotavirus A	RRV	ZeptoMetrix	0810530CF	10x LoD†

* Strain tested during LoD verification study.

† Testing at a lower concentration resulted in a detection rate of <100%.

In silico analysis

In silico analysis of potential reactivity showed that the following organisms (including species, subspecies, subtypes, serotypes or serovars) are predicted to be detected with the QIAstat-Dx Gastrointestinal Panel 2 (Table 11). *In silico* analysis of potential reactivity showed that the following organisms (including species, subspecies, subtypes, serotypes or serovars) are predicted to be detected with the QIAstat-Dx Gastrointestinal Panel 2 (Table 11).

Table 11. Organisms with predicted reactivity based on *in silico* analysis

QIAstat-Dx GI Panel	Organisms with predicted reactivity
2	
Bacteria	
Campylobacter	Campylobacter coli*, Campylobacter jejuni, Campylobacter jejuni subsp. jejuni, Campylobacter jejuni subsp. doylei, Campylobacter upsaliensis

Table 11. Organisms with predicted reactivity based on in silico analysis (continued)

QIAstat-Dx GI Panel

2	Organisms with predicted reactivity
Salmonella	Salmonella bongori*, Salmonella enterica subsp. salamae II (e.g. serovar 55:k:z39), Salmonella enterica subsp. arizonae IIIa (e.g. serovar 63:g:z51), Salmonella enterica subsp. diarizonae IIIb (e.g. serovar 47:l,v:z), Salmonella enterica subsp. houtenae IV (e.g. serovar 43:z4), Salmonella enterica subsp. indica VI. Salmonella enterica subsp. enterica (up to 92 different serovars including Agona, Anatum, Bareilly, Choleraesuis, Enteritidis, Heidelberg, Infantis, Kentucky, Montevideo, Newport, Paratyphi A*, Senftenberg, Tennessee, Thompson, Typhi, Typhimurium, Weltevreden*)
Plesiomonas shigelloides	Plesiomonas shigelloides (e.g. strains NCTC10360, ATCC 14029T, R4605035)
Yersinia enterocolitica	Yersinia enterocolitica, Yersinia enterocolitica subsp. palearctica, Yersinia enterocolitica subsp. enterocolitica
Enteroinvasive E. coli (EIEC)/Shigella	Enteroinvasive E. coli (EIEC), Escherichia coli sp., Shigella flexneri, Shigella dysenteriae, Shigella boydii, Shigella sonnei.
Enteropathogenic E. coli (EPEC)β	Enteropathogenic E. coli (EPEC) (e.g. including serotypes OUT: HND, OUT:H6, OUT:H34, OUT:H21, O55:H7, O119:HNM, O117)
Enterotoxigenic E. coli (ETEC)†	Enterotoxigenic E. coli (ETEC) (including H10407 and E24377A strains and serotypes O169:H41, O25:H42, O148:H28, O6:H16) carrier of: Heat-labile enterotoxin gene subtype LT-I and Heat-stable enterotoxin gene variant Sta, subtypes STp and STh
Shiga-like toxin-producing E. coli (STEC) - stx1/stx2β	Shiga-like toxin-producing E. coli (STEC) including O157:H7 and O157:NM serotype and non-O157 serotypes (O111:NM, O111:H, O26:H11, O145:NM, O145:H28, O45:H2, O26:H11, ONT:NM, O104:H4, O121:H19, O145:H34, O113:H21, ONT:H-, O128:H2, OUT:HNM, O124:HNM E. coli strains carrier of: stx1a, stx1c, stx1d, stx2a, stx2b, stx2c, stx2d, stx2d, stx2e, stx2f, stx2g, stx2h, stx2i, stx2j, stx2k and stx2l. Other stx-carrying bacteria: Shigella sonnei, Shigella dysenteriae
Shiga-like toxin-producing E. coli (STEC) O157β	Escherichia coli O157 including: STEC O157:H7 strains (e.g. EDL933) and E. coli O157: non-H7 groups including non-Shiga-toxigenic E. coli O157 bacteria (e.g. serotype O157:H45)
Parasites	

Table 11. Organisms with predicted reactivity based on in silico analysis (continued)

QIAstat-Dx GI Panel

2	Organisms with predicted reactivity
Cryptosporidium‡	Common Cryptosporidium species involved in human disease: <i>C. parvum</i> , <i>C. hominis</i> . Less common Cryptosporidium species involved in human infections: <i>C. meleagridis</i> , <i>C. felis</i> , <i>C. bovis</i> , <i>C. viatorum</i> , <i>C. ubiquitum</i> , <i>C. tyzzeri</i> , <i>C. cuniculus</i> , <i>Cryptosporidium</i> sp. Chipmunk genotype I, <i>C. canis</i> *. Rare or non-human species: <i>Cryptosporidium wrairi</i>
Cyclospora cayetanensis	<i>Cyclospora cayetanensis</i> (including strains LG, CY9, NP20 and NP21) *
Entamoeba histolytica	<i>Entamoeba histolytica</i> (e.g. strains HM-1: IMSS, EHMfas1, HK-9)*
Giardia lamblia	<i>Giardia lamblia</i> (aka <i>Giardia duodenalis</i> , <i>Giardia intestinalis</i>)*

Viruses

Adenovirus	Human Adenovirus F40/41
Astrovirus§	Human Astrovirus (including types 1, 2, 3, 4, 5, 6, 7, 8)
Norovirus GI/GII	Norovirus genogroup II genotypes: GII.1, GII.2, GII.3*, GII.4*, GII.5, GII.6, GII.7, GII.8, GII.9, GII.10, GII.12, GII.13, GII.14, GII.16, GII.17, GII.20, GII.21, GII.22, GII.23, GII.24*, GII.25, GII.26, GII.27, GII.NA1 and GII.NA2* Norovirus genogroup I genotypes: GI.1, GI.2, GI.3*, GI.4*, GI.5, GI.6*, GI.7*, GI.8, GI.9.
Rotavirus	Rotavirus A including genotypes: G1P[8]*, G2P[4]*, G3P[8]*, G4P[8]*, G9P[6], G9P[8]*, G12P[6]* and G12P[8]*

* Certain sequences are predicted to be detected with reduced sensitivity due to the presence of a reduced number of mismatches at critical positions of the primer-probe design.

‡The assay is not predicted to detect bacteria carrier of Heat-labile enterotoxin gene subtype LT-II and/or of Heat-stable enterotoxin gene variant Stb e.

‡The assay is not predicted to detect other *Cryptosporidium* spp. less involved in human disease: *C. andersoni* and *C. muris*. (88)

§ The assay is not predicted to detect Human Astrovirus types MLB1-3 and VA1-5.

¶ Only applicable for samples with Para-Pak C&S collection device

Interfering substances

The effect of potentially interfering substances on the detectability of the QIAstat-Dx Gastrointestinal Panel 2 organisms was evaluated. Thirty-four (34) potentially interfering substances were spiked into the sample mixes at a level predicted to be above the concentration of the substance likely to be found in stool specimens. Endogenous substances such as human whole blood, human genomic DNA and several pathogens were tested alongside exogenous substances like antibiotics, other gastrointestinal-related medications and different technique-specific substances.

Testing included samples containing negative clinical stool matrix in Para-Pak C&S media with and without addition of each potentially interfering substance. Samples containing organism mixes with one strain for each targeted pathogen were tested at 3x LoD. Testing was performed in triplicates. Additionally, for endogenous substances, negative specimens (stool matrix in Para-Pak C&S media matrix with no organism mix) were spiked with only the test substance to evaluate the potential for false positive results due to the test substance itself.

For the vast majority of substances tested, no interference was observed, with the exceptions of mucin, calcium carbonate, nonoxynol-9 and Rotavirus reassortants, that demonstrated interference at high concentration.

Mucin at 5% w/v was found to generate false positives results for the Yersinia target. These signals were investigated by testing the interfering substance with and FDA-cleared method and they were confirmed to be present in the endogenous substance.

Calcium carbonate at concentrations above 0.5% w/v was found to generate false negative results for all the QIAstat-Dx Gastrointestinal Panel 2 targets and the internal control.

Nonoxynol-9 at concentrations above 0.02% v/v was found to generate false negative results for detection of Entamoeba.

As predicted, Rotavirus reassortants WC3:2-5, R574(9) and WI79-4,9 used in Rotavirus A vaccines generated positive results for Rotavirus A in the QIAstat-Dx Gastrointestinal Panel 2. Final concentrations without interference (i.e., no false positive results for Rotavirus) for WC3:2-5, R574(9) and WI79-4,9 were 8.89×10^{-5} TCID50/mL and 1.10 PFU/mL, respectively (Table 12) for other concentrations tested.

Results from the 34 interfering substances that could be present or introduced in a stool specimen are provided in (Table 12).

Table 12. Final highest concentration without observable inhibitory effect

Substance tested	Concentration tested	Result
Endogenous substances		
Bovine and ovine bile	12% w/v	No Interference
Cholesterol	1.5% w/v	No Interference
Fatty acids (palmitic acid)	0.2% w/v	No Interference
Fatty acids (stearic acid)	0.4% w/v	No Interference
Human genomic DNA	20 µg/mL	No Interference
Human stool (overflow of Cary Blair vial)	300 mg/mL	No Interference
Human urine	50% v/v	No Interference
Human whole blood with Na Citrate	40% v/v	No Interference
Mucin from bovine submaxillary	5% w/v	Interference†
	2.5% w/v	No Interference
Triglycerides	5% w/v	No Interference
Exogenous substances		
Bacitracin	250U/mL	No Interference
Bisacodyl	0.3% w/v	No Interference
Bismuth subsalicylate	0.35% w/v	No Interference

Table 12. Final highest concentration without observable inhibitory effect (continued)

Substance tested	Concentration tested	Result
Calcium carbonate (TUMS® Extra Strength 750)	5%w/v	Interference
	0.5% w/v	No Interference
Docusate sodium	2.5% w/v	No Interference
Doxycycline hydrochloride	0.05% w/v	No Interference
Glycerin	50% v/v	No Interference
Hydrocortisone	0.5% w/v	No Interference
Loperamide hydrochloride	0.078% w/v	No Interference
Magnesium hydroxide	0.1% w/v	No Interference
Metronidazole	1.5% w/v	No Interference
Mineral oil	50% v/v	No Interference
Naproxen sodium	0.7% w/v	No Interference
Nonoxynol-9	1.2% v/v	Interference
	0.6% v/v	Interference
	0.3% v/v	Interference
	0.15% v/v	Interference
	0.075% v/v	Interference
	0.02% v/v	No Interference
Nystatin	10000 USP units/mL	No Interference
Phenylephrine hydrochloride	0.075% w/v	No Interference
Sodium phosphate	5% w/v	No Interference

Vaccine components

Table 12. Final highest concentration without observable inhibitory effect (continued)

Substance tested	Concentration tested	Result
Rotavirus reassortant WC3:2-5, R574(9) - VR 2195	8.89 x 10 ⁻³ TCID50/mL	Interference
		Interference
	8.89 x 10 ⁻⁴ TCID50/mL	No Interference
Rotavirus reassortant WI79-4,9 - VR 2415	8.89 x 10 ⁻⁵ TCID50/mL	
	1.10 x 10 ² pfu/mL	Interference
	1.10 x 10 ¹ pfu/mL	Interference
	1.10 pfu/mL	No Interference

Technique-specific Substances, Transport Media

Bleach	0.5% v/v	No Interference
Ethanol	0.2% v/v	No Interference
Puritan Fecal Opti-Swab Collection & Transport System with Cary-Blair Medium*	100%	No Interference
Puritan PurSafe® DNA/RNA Preservative*	100%	No Interference
Sigma Fecal Transwab*	1 swab/2mL Cary Blair	No Interference

* Performance not established for this transport media

† This substance was tested by another FDA-cleared test that also detected Yersinia positive signals.

Microbial interference

A microbial interference study was conducted to assess the inhibitory effects of select non-target organisms on the ability to detect the QIAstat-Dx Gastrointestinal Panel 2 targets. Clinically relevant and challenging concentrations of non-target organisms (1 x 10⁶ CFU/mL for bacteria, 1 x 10⁵ cells/mL for yeast and 1 x 10⁵ TCID50/mL for viruses) were individually mixed with negative clinical stool matrix with spiked targeted pathogens at 3x LoD. Testing was performed in triplicate. All combinations and replicates successfully detected all the

QIAstat-Dx Gastrointestinal Panel 2 targets. See Table 13 for a list of the non-target organisms tested and the result summary.

Table 13. Final highest concentration without observable inhibitory effect

Substance tested	Concentration tested	Result
<i>Non-target microorganisms</i>		
<i>Aeromonas hydrophila</i>	1 x 10 ⁶ units/mL	No Interference
<i>Bacteroides vulgatus</i>	1 x 10 ⁶ units/mL	No Interference
<i>Bifidobacterium bifidum</i>	1 x 10 ⁶ units/mL	No Interference
<i>Enterovirus Species D, Serotype EV-D68</i>	1 x 10 ⁵ units/mL	No Interference
<i>Non-pathogenic E. coli</i>	1 x 10 ⁶ units/mL	No Interference
<i>Helicobacter pylori</i>	1 x 10 ⁶ units/mL	No Interference
<i>Saccharomyces cerevisiae (deposited as S. boulardii)</i>	1 x 10 ⁵ units/mL	No Interference

Competitive interference

Competitive interference was tested in a subset of pathogens. No interference was observed when evaluating competitive interference by target pathogens when two QIAstat-Dx Gastrointestinal Panel 2 target pathogens were tested by spiking samples with one pathogen target at 3x LoD and one at 50x LoD. Results from the pathogen targets tested are provided in Table 14.

Table 14. QIAstat-Dx Gastrointestinal Panel 2 results for competitive interference

Sample Mix	Target	Final concentration (molecular units)*	Final concentration tested x LoD	Co-infection detected
Norovirus 50x - Rotavirus 3x	Norovirus	4.5E+05 copies/mL	50x	Yes
	GI/GII Rotavirus A	1.7E+04 copies/mL	3x	

Table 14. QIAstat-Dx Gastrointestinal Panel 2 results for competitive interference (continued)

Sample Mix	Target	Final concentration (molecular units)*	Final concentration tested x LoD	Co-infection detected
Norovirus 3x - Rotavirus 50x	Norovirus	2.7E+04 copies/mL	3x	Yes
	GI/GII	2.9E+05 copies/mL	50x	
	Rotavirus A			
Giardia 50x - Adenovirus 3x	Giardia lamblia	7.2 E+05 copies/mL	50x	Yes
	Adenovirus F40/F41	2.9E+03 copies/mL	3x	

*Molecular unit titers were determined using in-house developed and validated qPCR assays.

Carryover

A carryover study was performed to evaluate the potential occurrence of cross-contamination between consecutive runs when using the QIAstat-Dx Gastrointestinal Panel 2 on the QIAstat-Dx Analyzer 1.0.

Pathogen samples of stool sample matrix in Para-Pak C&S transport media, with alternating high-positive (10⁶ CFU/mL for bacteria-, 10⁵ TCID₅₀ or organism/mL for viruses and parasites) and negative samples, were conducted on two QIAstat-Dx Analyzer 1.0 instruments.

No carryover between samples was observed in the QIAstat-Dx Gastrointestinal Panel 2, demonstrating that the system design and recommended sample handling and testing practices are effective in preventing false-positive results due to carryover or cross-contamination between samples.

Reproducibility

Reproducibility testing of contrived samples was performed at three test sites including one internal site (Site A) and two external sites (Site B and Site C). The study incorporated a range

of potential variation introduced by sites, days, replicates, cartridge lots, operators, and QIAstat-Dx analyzers. For each site, testing was performed across 5 non-consecutive days with 6 replicates per day (leading to a total of 30 replicates per target, concentration and site), 4 QIAstat-Dx Analyzers (2 analyzers per operator and per site), and at least 2 operators on each testing day. A total of 5 sample mixes (two combined samples at 1x LoD and 3x LoD plus one negative sample) were prepared. For each mix, 6 replicates were tested and evaluated.

Table 15 shows the detection rate per target and concentration for each site of the Reproducibility study. In addition, data obtained at all three sites have been compiled to calculate the exact 2-sided 95% Confidence Interval by target and concentration. During the reproducibility study, potential variation introduced by sites, days, replicates, cartridge lots, operators, and QIAstat-Dx analyzers was analyzed showing no significant contribution to variability (Standard Deviation and Coefficient of Variation values below 1 and 5%, respectively) caused by any of the assessed variables.

Table 15. Detection rate per target and concentration for each site of the Reproducibility study and exact 2-sided 95% Confidence Interval by target and concentration

% Agreement with Expected Result						
Pathogen Tested	Concentration Tested	Expected Result	Site A	Site B	Site C	All Sites (95% Confidence Interval)
Adenovirus F41 ZeptoMetrix 0810085CF	3x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	1x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	None	Not Detected	29/30 96.67%	29/30 96.67%	29/30 96.67%	87/90* 96.7% (90.98 - 98.9%)
Campylobacter ZeptoMetrix 801650	3x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	1x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	None	Not Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)

Table 15. Detection rate per target and concentration for each site of the Reproducibility study and exact 2-sided 95% Confidence Interval by target and concentration (continued)

% Agreement with Expected Result						
Pathogen Tested	Concentration Tested	Expected Result	Site A	Site B	Site C	All Sites (95% Confidence Interval)
<i>Escherichia coli</i> EPEC § ZeptoMetrix 801747	3x LoD	Detected	30/30	30/30	30/30	90/90
			100%	100%	100%	100%
						(95.98 - 100.00%)
	1x LoD	Detected	30/30	29/30	30/30	89/90
			100%	96.67%	100%	98.89%
						(93.96 – 99.97%)
	None	Not Detected	30/30	30/30	30/30	90/90
			100%	100%	100%	100%
						(95.98 - 100.00%)
<i>Entamoeba histolytica</i> ATCC 30459	3x LoD	Detected	30/30	30/30	30/30	90/90
			100%	100%	100%	100%
						(95.98 - 100.00%)
	1x LoD	Detected	30/30	30/30	29/30	89/90
			100%	100%	96.67%	98.89%
						(93.96 – 99.97%)
	None	Not Detected	30/30	30/30	30/30	90/90
			100%	100%	100%	100%
						(95.98 - 100.00%)

Table 15. Detection rate per target and concentration for each site of the Reproducibility study and exact 2-sided 95% Confidence Interval by target and concentration (continued)

% Agreement with Expected Result						
Pathogen Tested	Concentration Tested	Expected Result	Site A	Site B	Site C	All Sites (95% Confidence Interval)
<i>Giardia lamblia</i> ATCC 30888	3x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	1x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	None	Not Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
<i>Norovirus GI</i> ZeptoMetrix 0810087CF	3x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	1x LoD	Detected	29/30 96.67%	30/30 100%	30/30 100%	89/90 98.89% (93.96 - 99.97%)
	None	Not Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)

Table 15. Detection rate per target and concentration for each site of the Reproducibility study and exact 2-sided 95% Confidence Interval by target and concentration (continued)

% Agreement with Expected Result						
Pathogen Tested	Concentration Tested	Expected Result	Site A	Site B	Site C	All Sites (95% Confidence Interval)
Rotavirus A † ZeptoMetrix 0810280CF	3x LoD	Detected	29/30 96.67%	29/30 96.67%	30/30 100%	88/90 97.8% (92.20 - 99.73%)
	1x LoD	Detected	23/30 76.67%	26/30 86.67%	12/12 100%	61/72 84.7% (74.31 - 92.12%)
	None	Not Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
Escherichia coli (STEC) O157:H7§ ZeptoMetrix 0801622	3x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	1x LoD	Detected	30/30 100%	30/30 100%	29/30 96.67%	89/90 98.89% (93.96 - 99.97%)
	None	Not Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)

Table 15. Detection rate per target and concentration for each site of the Reproducibility study and exact 2-sided 95% Confidence Interval by target and concentration (continued)

% Agreement with Expected Result						
Pathogen Tested	Concentration Tested	Expected Result	Site A	Site B	Site C	All Sites (95% Confidence Interval)
<i>Escherichia coli</i> (STEC)§ stx1/stx2 ZeptoMetrix 0801622	3x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	1x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	None	Not Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
<i>Salmonella enterica</i> ZeptoMetrix 0801437	3x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	1x LoD	Detected	30/30 100%	29/30 96.67%	29/30 96.67%	88/90 97.78% (92.20 - 99.73%)
	None	Not Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)

Table 15. Detection rate per target and concentration for each site of the Reproducibility study and exact 2-sided 95% Confidence Interval by target and concentration (continued)

% Agreement with Expected Result						
Pathogen Tested	Concentration Tested	Expected Result	Site A	Site B	Site C	All Sites (95% Confidence Interval)
<i>Yersinia enterocolitica</i> Zeptomatrix 0801734	3x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	1x LoD	Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)
	None	Not Detected	30/30 100%	30/30 100%	30/30 100%	90/90 100% (95.98 - 100.00%)

* Three (3) Adenovirus F40/41 false positives were observed when testing negative sample. Retesting of the three samples resulted in the expected negative results.

† One (1) Giardia lamblia false positive was observed when testing a positive sample not containing the pathogen. Repeat testing of this sample resulted in the expected negative result.

‡ The Reproducibility study was fully re-tested for Rotavirus A with a new sample set due to an unexpected number of false negatives for Rotavirus A at the 1x LoD concentration. This was observed with during an interim data evaluation (61/72, 84.7%) and that was attributed to the sample manufacture and while unrelated to the study workflow variables (operator, lot, day, instrument and site). Test runs derived from Rotavirus A new sample set resulted in 90/90 (100%; 95.98-100% CI) for the 3x LoD and 89/90 (98.89%; 93.96-99.97% CI) for the 1x LoD. During this testing, one (1) *Campylobacter* false positive was observed Retesting of this sample resulted in the expected negative result.

§ Only applicable for Para-Pak C&S samples

Repeatability

A repeatability study was conducted on the QIAstat-Dx Analyzer 1.0 instruments using a set of samples composed of low-concentrated analytes spiked into stool matrix (3x LoD and 1x LoD) and negative stool samples. QIAstat-Dx Gastrointestinal Panel 2 detected pathogens included in the positive samples were Adenovirus, *Campylobacter*, *Entamoeba histolytica*, *Giardia lamblia*, Norovirus GII, Rotavirus, *Salmonella enterica*, and *Yersinia enterocolitica*, and additionally Enteropathogenic *E. coli* (EPEC), STEC stx1/stx2, and *E. coli* O157 which are only applicable for Para-Pak C&S samples. Each sample was tested with the same instrument over 12 days. In total, 60 replicates of 1x LoD and 60 replicates of 3x LoD per each of the tested targets and 60 replicates of negative samples were run. Overall results showed a 93.33-100.00-% and 95.00-100.00% detection rate for 1x LoD and 3x LoD samples, respectively. Negative samples showed 100% of negative calls for all panel analytes.

Expected values

The number and percentage of positive results as determined by the QIAstat-Dx Gastrointestinal Panel 2 in the prospective clinical evaluation, stratified by age group, are presented in Table 16. Overall, the QIAstat-Dx Gastrointestinal Panel 2 detected at least 1 organism 17.4% (213/1222) and 23.8% (171/717) of the prospectively collected stool specimens in FecalSwab and Para-Pak C&S, respectively.

Table 16. Expected values Summary by Age Group for the Prospective Clinical study as determined by the QIAstat-Dx Gastrointestinal Panel 2

Pathogen	Medium Brand	Overall	0-5 years	6-21 years	22-49 years	50+ years	Not Reported
Viruses							
Adenovirus F40/F41	FecalSwab	5 (0.4%)	3 (1.7%)	2 (1.7%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	Para-Pak C&S	2 (0.3%)	1 (3.2%)				0 (0.0%)
					0 (0.0%)	0 (0.0%)	1 (0.2%)

Table 16. Expected values Summary by Age Group for the Prospective Clinical study as determined by the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Medium Brand	Overall	0-5 years	6-21 years	22-49 years	50+ years	Not Reported
Astrovirus	FecalSwab	3 (0.2%)	3 (1.6%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	Para-Pak	6 (0.8%)	2 (6.5%)				0 (0.0%)
	C&S			0 (0.0%)	3 (1.4%)	1 (0.2%)	
Norovirus GI/GII	FecalSwab	43 (3.5%)	22 (12.1%)	1 (0.8%)	14 (4.8%)	6 (1.0%)	0 (0.0%)
	Para-Pak						0 (0.0%)
	C&S	16 (2.3%)	3 (9.7%)	1 (2.8%)	3 (1.4%)	9 (2.2%)	
Rotavirus A	FecalSwab	23 (1.9%)	13 (7.1%)	2 (1.7%)	7 (2.4%)	1 (0.2%)	0 (0.0%)
	Para-Pak						0 (0.0%)
	C&S	4 (0.6%)	2 (6.5%)	0 (0.0%)	0 (0.0%)	2 (0.5%)	
Bacteria							
Campylobacter	FecalSwab	69 (5.6%)	25 (13.7%)	7 (5.8%)	17 (5.9%)	20 (3.2%)	0 (0.0%)
	Para-Pak						0 (0.0%)
	C&S	30 (4.2%)	2 (6.5%)	0 (0.0%)	10 (4.7%)	18 (4.3%)	
Plesiomonas shigelloides	FecalSwab	2 (0.2%)	0 (0.0%)	0 (0.0%)	2 (0.7%)	0 (0.0%)	0 (0.0%)
	Para-Pak	7 (1.0%)	1 (3.2%)	0 (0.0%)			0 (0.0%)
	C&S				4 (1.9%)	2 (0.5%)	
Salmonella	FecalSwab	14 (1.1%)	5 (2.7%)	4 (3.3%)	3 (1.0%)	2 (0.3%)	0 (0.0%)
	Para-Pak		4 (12.9%)				0 (0.0%)
	C&S	17 (2.4%)		0 (0.0%)	3 (1.4%)	10 (2.4%)	
Yersinia enterocolitica	FecalSwab	22 (1.8%)	3 (1.6%)	2 (1.7%)	9 (3.1%)	8 (1.3%)	0 (0.0%)
	Para-Pak		0 (0.0%)				0 (0.0%)
	C&S	8 (1.1%)		0 (0.0%)	4 (1.9%)	4 (1.0%)	

Table 16. Expected values Summary by Age Group for the Prospective Clinical study as determined by the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Medium Brand	Overall	0-5 years	6-21 years	22-49 years	50+ years	Not Reported
Diarrheagenic <i>E. coli</i>/Shigella							
Enteropathogenic <i>E. coli</i> (EPEC)	Para-Pak C&S	56 (7.9%)	9 (29.0%)	2 (5.6%)	18 (8.4%)	27 (6.5%)	0 (0.0%)
	FecalSwab	18 (1.5%)	2 (1.1%)	2 (1.7%)	11 (3.8%)	3 (0.5%)	0 (0.0%)
Enterotoxigenic <i>E. coli</i> (ETEC) lt/st	Para-Pak C&S	17 (2.4%)	1 (3.2%)	0 (0.0%)	7 (3.3%)	9 (2.2%)	0 (0.0%)
	FecalSwab	10 (0.8%)	1 (0.5%)	0 (0.0%)	6 (2.1%)	3 (0.5%)	0 (0.0%)
Shiga-like toxin <i>E. coli</i> (STEC) stx1/stx2 <i>E. coli</i> O157	Para-Pak C&S	9 (1.3%)	0 (0.0%)	0 (0.0%)	6 (2.8%)	3 (0.7%)	0 (0.0%)
	Para-Pak C&S	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
Shigella/Enteroinvasive <i>E. coli</i> (EIEC)	FecalSwab	10 (0.8%)	1 (0.5%)	0 (0.0%)	6 (2.1%)	3 (0.5%)	0 (0.0%)
	Para-Pak C&S	3 (0.4%)	0 (0.0%)	0 (0.0%)	1 (0.5%)	2 (0.5%)	0 (0.0%)
Parasites							
<i>Cryptosporidium</i>	FecalSwab	2 (0.2%)	0 (0.0%)	1 (0.8%)	1 (0.3%)	0 (0.0%)	0 (0.0%)
	Para-Pak C&S	7 (1.0%)	0 (0.0%)	1 (2.8%)	4 (1.9%)	2 (0.5%)	0 (0.0%)
<i>Cyclospora cayetanensis</i>	FecalSwab	3 (0.2%)	0 (0.0%)	1 (0.8%)	2 (0.7%)	0 (0.0%)	0 (0.0%)
	Para-Pak C&S	18 (2.5%)	0 (0.0%)	0 (0.0%)	6 (2.8%)	12 (2.9%)	0 (0.0%)
<i>Giardia lamblia</i>	FecalSwab	15 (1.2%)	3 (1.6%)	1 (0.8%)	7 (2.4%)	4 (0.6%)	0 (0.0%)
	Para-Pak C&S	1 (0.1%)	1 (3.2%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)

Table 16. Expected values Summary by Age Group for the Prospective Clinical study as determined by the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Pathogen	Medium Brand	Overall	0-5 years	6-21 years	22-49 years	50+ years	Not Reported
<i>Entamoeba histolytica</i>	FecalSwab	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)
	Para-Pak C&S	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)

Clinical performance

The clinical performance shown below was demonstrated using QIAstat-Dx Analyzer 1.0. The QIAstat-Dx Analyzer 2.0 use the same Analytical Modules as QIAstat-Dx Analyzer 1.0; therefore, the performance is not impacted by the QIAstat-Dx Analyzer 2.0. The clinical performance of QIAstat-Dx Gastrointestinal Panel 2 was established during a multi-center international prospective study conducted at thirteen clinical settings representatives of different geographical areas within USA and Europe (9 sites in USA and 4 sites in Europe) between May and July 2021. All study sites were hospital-associated or independent clinical diagnostics laboratories that perform routine diagnostics of GI infections. A total of 1939 prospectively collected stool specimens (stool preserved in Para-Pak C&S (Meridian Bioscience) or FecalSwab (COPAN)) were obtained from patients with clinical indications of diarrhea caused by gastrointestinal infection. Table 17 provides a summary of prospective specimen’s distribution across all study sites.

Table 17. Prospective Specimens Distribution Across the study sites

Site/Country	FecalSwab	Para-Pak C&S	Total
Germany	293	46	339
Denmark	293	0	293
Spain	247	0	247
France	63	0	63

Table 17. Prospective Specimens Distribution Across the study sites (continued)

Site/Country	FecalSwab	Para-Pak C&S	Total
USA site 1	0	186	186
USA site 2	0	43	43
USA site 3	282	0	282
USA site 4	0	177	177
USA site 5	44	0	44
USA site 6	0	39	39
USA site 7	0	0	0*
USA site 8	0	131	131
USA site 9	0	95	95
Total	1222	717	1939

* The specimens from this site were excluded from the analysis because they were collected with another device different to Para-Pak C&S or FecalSwab.

The demographic information for the 1,939 specimens evaluated in the prospective study is summarized in Table 18 below.

Table 18. Demographic data for prospective evaluated specimens

Demographic data	FecalSwab		Para-Pak C&S	
	N	%	N	%
Gender				
Female	628	32.4	442	22.8
Male	594	30.6	275	14.2
Age Group				
0-5 years	182	9.4	31	1.6
6-21 years	121	6.2	38	2.0

Table 18. Demographic data for prospective evaluated specimens (continued)

Demographic data	FecalSwab		Para-Pak C&S	
	N	%	N	%
22-49 years	290	15.0	215	11.1
50+ years	629	32.4	426	22.0
Not Reported	0	0.0	7	0.4
Patient population				
Emergency room	46	2.4	29	1.5
Hospitalized	342	17.6	143	7.4
Immunocompromised	3	0.2	0	0.0
Outpatient	491	25.3	325	16.8
No information available	340	17.5	220	11.3
No. Days between Symptom Onset and QIAstat-Dx Testing				
> 7 days	89	4.6	0	0.0
≤ 7 days	146	7.5	16	0.8
Not Reported	987	50.9	701	36.2

The performance of the QIAstat-Dx Gastrointestinal Panel 2 was evaluated for each panel test result using one FDA-cleared test as comparator for the most analytes. A composite comparator consisting of either three independent FDA-cleared test methods or two independent FDA-cleared tests methods and two validated PCR assays followed by bi-directional sequencing was used for Norovirus GI/GII, ETEC, STEC and Giardia lamblia (Table 19).

Table 19. Comparator Methods for the Clinical Evaluation of QIAstat-Dx Gastrointestinal Panel 2

QIAstat-Dx GI Panel 2 Test Result	Comparator Method
<i>Adenovirus F40/F41</i>	One FDA-cleared test method
<i>Astrovirus</i>	
<i>Rotavirus A</i>	
<i>Campylobacter</i>	
<i>Plesiomonas shigelloides</i>	
<i>Salmonella</i>	
<i>Yersinia enterocolitica</i>	
<i>E. coli O157</i> *	
Enteropathogenic <i>E. coli</i> (EPEC) *	
<i>Shigella</i> /Enteroinvasive <i>E. coli</i> (EIEC)	
<i>Cryptosporidium</i>	
<i>Cyclospora cayetanensis</i>	
<i>Entamoeba histolytica</i>	
Norovirus GI/GII	Composite of three FDA-cleared test methods
Enterotoxigenic <i>E. coli</i> (ETEC) lt/st	Composite of three FDA-cleared test methods
Shiga-like toxin- <i>E. coli</i> (STEC) stx1/stx2*	Composite of three FDA-cleared test methods
<i>Giardia lamblia</i>	Composite of two FDA-cleared test methods and two validated PCR tests followed by bi-directional sequencing†

* Targets evaluated in Para-Pak C&S specimens only.

† Each of the two PCR assays used were well-characterized and validated nucleic acid amplification tests (NAAT) followed by bi-directional sequencing analysis. Both assays were designed to amplify different sequences than those targeted by the QIAstat-Dx Gastrointestinal Panel 2. Positive results required to generate sequences from bi-directional sequencing with at least 200 bases of adequate quality that by BLAST analyses matched a sequence of the expected organism or gene from NCBI GenBank database with at least 95% query coverage and at least 95% identity compared to the reference.

Additional prospective archived samples were collected for Norovirus GI/GII (81 samples) and STEC (18 samples). These were prospectively collected samples from four different collection sites (3 US and 1 EU), where only those positive for the pathogen by standard of care method were archived for analysis alongside 20 negative specimens.

In addition, to supplement the results of the prospective clinical studies, a total of 750 preselected archived frozen (retrospective) specimens were also evaluated. These specimens served to increase the sample size for analytes that showed low prevalence in the clinical prospective study or that were less represented in a particular sample type (Para-Pak C&S or FecalSwab). The same Comparator Methods detailed in Table 19 were used as confirmatory testing for the presence of the nucleic acids from the expected analytes. In total 2808 specimens (1939 prospective, 119 prospective archived and 750 retrospective) were evaluated in the clinical study. These specimens were collected using Para-Pak C&S (1217) or FecalSwab (1591).

The positive percentage agreement (PPA) and the negative percentage agreement (NPA) were calculated for the prospective and retrospective studies and for each sample type (Para-Pak C&S and FecalSwab) separately.

The PPA was calculated as $100\% \times (TP / (TP + FN))$. True positive (TP) indicates that both the QIAstat-Dx Gastrointestinal Panel 2 and comparator method showed a positive result for this specific target, and false negative (FN) indicates that the QIAstat-Dx Gastrointestinal Panel 2 result was negative while the comparator method result was positive. The NPA was calculated as $100\% \times (TN / (TN + FP))$. True negative (TN) indicates that both the QIAstat-Dx Gastrointestinal Panel 2 and the comparator method showed negative results, and a false positive (FP) indicates that the QIAstat-Dx Gastrointestinal Panel 2 result was positive, but the comparator method result was negative. The exact binomial two-sided 95% confidence intervals for PPA and NPA were calculated.

Where a composite comparator was used (Table 19), the result was determined as the majority of the three individual test results (i.e., a positive composite comparator result is based on positive results for at least two comparator tests and a negative composite comparator result is based on negative results for at least two comparator tests). If insufficient pathogen positive sample was available to obtain a majority test result a worst-case model was applied in the PPA calculation. In this model the PPA was calculated including all observed true positive and false negative samples between QIAstat-Dx and the composite comparator while for the samples where it was not possible to conduct testing with the complete comparator due to insufficient sample volume, the following was done:

- Samples that were negative in QIAstat-Dx and positive for one comparator assay, negative (or insufficient volume) for a second comparator and insufficient volume for a third comparator were included in the calculations as worst-case false negatives.
- Samples that were positive in QIAstat-Dx and positive in one comparator test, negative (or insufficient volume) for a second comparator and insufficient volume for the third comparator, were considered as worst-case false positives and therefore, excluded in the PPA calculations.

The results of the clinical performance of the prospective, prospective archived and retrospective studies are summarized in Table 20, Table 21, and Table 22 respectively.

Discrepancies between the QIAstat-Dx Gastrointestinal Panel 2 and the comparator methods were investigated for the analytes that the QIAstat-Dx Gastrointestinal Panel 2 test result was compared to one FDA-cleared method. Discrepancies analyses are footnoted on each clinical performance summary Table below (Table 20 and Table 21).

Table 20. Clinical Performance in the Prospective study

Analyte	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
Viruses							
Adenovirus F40/F41	FecalSwab	5/6 a	83.3	43.7-97.0	1216/1216	100.0	99.7-100.0
	Para-Pak	1/2 b	50.0	9.5-90.6	703/704 b	99.9	99.2-100.0
	C&S						
Astrovirus	FecalSwab	3/3	100.0	43.9-100.0	1219/1219	100.0	99.7-100.0
	Para-Pak	6/6	100.0	61.0-100.0	700/700	100.0	99.5-100.0
	C&S						
Norovirus GI/GII	FecalSwab	31/33 c	93.9	80.4-98.3	493/495 c	99.6	98.6-100.0
	Para-Pak	14/18 d	77.8	54.8-91.0	399/399 d	100.0	99.1-100.0
	C&S						
Rotavirus A	FecalSwab	21/23 e	91.3	73.2-97.6	1197/1199e	99.8	99.4-100.0
	Para-Pak	3/3	100.0	43.9-100.0	702/703 f	99.9	99.2-100.0
	C&S						
Campylobacter	FecalSwab	65/67 g	97.0	89.8-99.2	1151/1155g	99.7	99.1-99.9
	Para-Pak	30/31 h	96.8	83.8-99.4	675/677 h	99.7	98.9-99.9
	C&S						
Plesiomonas shigelloides	FecalSwab	0/0	N/A	N/A	1220/1222 i	99.8	99.4-100.0
	Para-Pak	5/6 j	83.3	43.7-97.0	698/700 j	99.7	99.0-99.9
	C&S						
Salmonella	FecalSwab	14/16 k	87.5	64.0-96.5	1206/1206	100.0	99.7-100.0
	Para-Pak	19/20 l	95.0	76.4-99.1	688/688	100.0	99.4-100.0
	C&S						

Table 20. Clinical Performance in the Prospective study (continued)

Analyte	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
<i>Yersinia enterocolitica</i>	FecalSwab	15/16 m	93.8	71.7-99.0	1199/1206m	99.4	98.8-99.7
	Para-Pak	3/3	100.0	43.9-100.0	698/703 n	99.3	98.4-99.7
	C&S						
Diarrheagenic E. coli/Shigella							
Enteropathogenic E. coli (EPEC)	Para-Pak	57/65	87.7	77.6-93.6	632/632	100.0	99.4-100.0
	C&S						
Enterotoxigenic E. coli (ETEC) lt/st	FecalSwab	9/10 o	90.0	59.6-99.2	427/430 o	99.3	98.0-99.8
	Para-Pak	9/10 p	90.0	59.6-99.2	390/395 p	98.7	97.1-99.5
	C&S						
Shiga-like toxin E. coli (STEC) stx1/stx2	Para-Pak	5/6 q	83.3	43.7-97.0	397/400 q	99.3	97.8-99.7
	C&S						
E. coli O157	FecalSwab	10/10	100.0	72.3-100.0	1212/1212	100.0	99.7-100.0
	Para-Pak	2/2	100.0	34.2-100.0	703/704 r	99.9	99.2-100.0
	C&S						
Cryptosporidium	FecalSwab	2/4	50.0	15.0-85.0	1218/1218	100.0	99.7-100.0
	Para-Pak	6/6	100.0	61.0-100.0	699/700 s	99.9	99.2-100.0
	C&S						
Cyclospora cayetanensis	FecalSwab	3/3	100.00	43.9-100.0	1219/1219	100.0	99.7-100.0
	Para-Pak	18/19 t	94.7	75.4-99.1	687/687	100.0	99.4-100.0
	C&S						

Table 20. Clinical Performance in the Prospective study (continued)

Analyte	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
<i>Entamoeba histolytica</i>	FecalSwab	0/0	N/A	N/A	1222/1222	100.0	99.7-
	Para-Pak	0/0	N/A	N/A	706/706	100.0	100.0
	C&S						99.5- 100.0
<i>Giardia lamblia</i>	FecalSwab	6/8 ^u	75.0	40.9-	434/441 ^u	98.4	96.8-
	Para-Pak	1/1	100.0	92.9	406/406 ^v	100.0	99.2
	C&S			20.7- 100.0			99.1- 100.0

a Adenovirus F40/41 was not detected in the single false negative specimen (0/1) in FecalSwab using a different FDA-cleared test method

b Adenovirus F40/41 was not detected in the single false negative specimen (0/1) and in the single false positive specimen (0/1) in Para-Pak C&S using a different FDA-cleared test method

c ten (10) FecalSwab samples positive for Norovirus GI/GII in both QIAstat-Dx and one FDA-cleared comparator were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. The sample size for NPA is smaller for Norovirus GI/GII as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the prospective study.

d two (2) Para-Pak C&S samples positive for Norovirus GI/GII in both QIAstat-Dx and one FDA-cleared comparator were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. One (1) Para-Pak C&S sample negative in QIAstat-Dx and positive with one FDA-cleared comparator with insufficient volume for complete composite comparator testing were classed as false negative in the PPA calculations. The sample size for NPA is smaller for Norovirus GI/GII as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the prospective study.

e Rotavirus A was detected in one of the two false negative specimens (1/2) and was not detected in the two false positive specimens (0/2) in FecalSwab using a different FDA-cleared test method.

f Rotavirus A was not detected in the single false positive specimen (0/1) in Para-Pak C&S using a different FDA-cleared test method.

Table 20. Clinical Performance in the Prospective study (continued)

Analyte	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
g <i>Campylobacter</i> was not detected in the two false negative specimens (0/2) and was detected in three of the four false positive specimens (3/4) in FecalSwab using a different FDA-cleared test method.							
h <i>Campylobacter</i> was not detected in the single false negative specimens (0/1) and was detected in one of the two false positive specimens (1/2) in Para-Pak C&S using a different FDA-cleared test method.							
i <i>Plesiomonas shigelloides</i> was not detected in the two false positive specimens (0/2) in FecalSwab using a different FDA-cleared test method.							
j <i>Plesiomonas shigelloides</i> was not detected in the single false negative specimen (0/1) and was not detected in the two false positive specimens (0/2) in Para-Pak C&S using a different FDA-cleared test method.							
k <i>Salmonella</i> was not detected in the two false negative specimens (0/2) in FecalSwab using a different FDA-cleared test method.							
l <i>Salmonella</i> was not detected in the single false negative specimen (0/1) in Para-Pak C&S using a different FDA-cleared test method.							
m <i>Yersinia enterocolitica</i> was not detected in the single false negative specimen (0/1) and was not detected in the seven false positive specimens (0/7) in FecalSwab using a different FDA-cleared test method.							
n <i>Yersinia enterocolitica</i> was not detected in the five false positive specimens (0/5) using a different FDA-cleared test method.							
o six (6) FecalSwab samples positive for ETEC in both QIAstat-Dx and the primary FDA-cleared comparator were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. The sample size for NPA is smaller for ETEC as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the prospective study.							
p three (3) Para-Pak C&S samples positive for ETEC in both QIAstat-Dx and one FDA-cleared comparator were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. The sample size for NPA is smaller for ETEC as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the prospective study.							

Table 20. Clinical Performance in the Prospective study (continued)

Analyte	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
q one (1) Para-Pak C&S sample positive for STEC in both QIAstat-Dx and one FDA-cleared comparator was excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. The sample size for NPA is smaller for STEC as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the prospective study.							
r Shigella/ EIEC was detected in the single false positive specimen (1/1) in Para-Pak C&S using a different FDA-cleared test method.							
s Cryptosporidium was not detected in the single false positive specimen (0/1) in Para-Pak C&S using PCR followed by bi-directional sequence analysis.							
t For Cyclospora cayetanensis there was one (1) false negative specimen in Para-Pak C&S that was not further investigated by discrepant analyses.							
u two (2) FecalSwab samples positive for Giardia lamblia in both QIAstat-Dx and one FDA-cleared comparator were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. Two (2) FecalSwab samples negative in QIAstat-Dx and positive with one FDA-cleared comparator with insufficient volume for complete composite comparator testing were classed as false negative in the PPA calculations. The sample size for NPA is smaller for Giardia lamblia as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the prospective study.							
v the sample size for NPA is smaller for Giardia lamblia as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the prospective study.							

Table 21. Clinical Performance in the Prospective Archived study

Analyte	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
Norovirus	FecalSwab	48/49*	98.0	89.3 -	2/4*	50.0	15.0-85.0
GI/GII	Para-Pak	26/28*†	92.9	99.6	37/38*	97.4	86.5-99.5
	C&S			77.4 - 98.0			

Table 21. Clinical Performance in the Prospective Archived study (continued)

Analyte	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
Shiga-like toxin E. coli (STEC) stx1/stx2	Para-Pak C&S	12/13†§	92.3	66.7 - 98.6	51/52‡	98.1	89.9-99.7

* For Norovirus GI/GII four out of the eighty-one (4/81) prospectively archived samples (positive by standard of care) were negative by the composite comparator and therefore included as negative samples in the NPA calculations

† One (1) Para-Pak C&S sample negative in QIAstat-Dx and positive for Norovirus GI/GII with one FDA-cleared comparator with insufficient volume for complete composite comparator testing was classed as false negative in the PPA calculations.

‡ For STEC five out of the eighteen (5/18) prospectively archived samples (positive by standard of care) were negative by the composite comparator and therefore included as negative samples in the NPA calculations.

§ One (1) Para-Pak C&S sample positive for STEC in both QIAstat-Dx and one FDA-cleared comparator was excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing.

Table 22. Clinical Performance in the Retrospective study

	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
Viruses							
Adenovirus F40/F41	FecalSwab	23/26 a	88.5	71.0-96.0	203/203	100.0	98.1-100.0
	Para-Pak C&S	29/29	100.0	88.3-100.0	39/39	100.0	91.0-100.0
Astrovirus	FecalSwab	2/3 b	66.7	20.8-93.9	191/191	100.0	98.0-100.0
	Para-Pak C&S	0/0	N/A	N/A	14/14	100.0	78.5-100.0

Table 22. Clinical Performance in the Retrospective study (continued)

	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
Norovirus GI/GII	FecalSwab	28/32 c	87.5	71.9-95.0	74/75 c	98.7	92.8-99.8
	Para-Pak C&S	27/29	93.1	78.0-98.1	86/86 d	100.0	95.7-100.0
Rotavirus A	FecalSwab	8/9 e	88.9	56.5-98.0	185/185	100.0	98.0-100.0
	Para-Pak C&S	2/2	100.0	34.2-100.0	12/12	100.0	75.8-100.0
Bacteria							
Campylobacter	FecalSwab	31/31	100.0	89.0-100.0	161/163 e	98.8	95.6-99.7
	Para-Pak C&S	3/3	100.0	43.9-100.0	11/11	100.0	74.1-100.0
Plesiomonas shigelloides	FecalSwab	2/2	100.0	34.2-100.0	192/192	100.0	98.0-100.0
	Para-Pak C&S	33/36 g	91.7	78.2-97.1	117/117	100.0	96.8-100.0
Salmonella	FecalSwab	30/31 h	96.8	83.8-99.4	161/163 h	98.8	95.6-99.7
	Para-Pak C&S	1/1	100.0	20.7-100.0	13/13	100.0	77.2-100.0
Yersinia enterocolitica	FecalSwab	32/34 i	94.1	80.9-98.4	160/160	100.0	97.7-100.0
	Para-Pak C&S	1/1	100.0	20.7-100.0	14/14	100.0	78.5-100.0
Diarrheagenic E. coli/Shigella							
Enteropathogenic E. coli (EPEC)	Para-Pak C&S	60/65 j	92.3	83.2-96.7	42/42	100.0	91.6-100.0

Table 22. Clinical Performance in the Retrospective study (continued)

	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
Enterotoxigenic <i>E. coli</i> (ETEC) lt/st	FecalSwab	22/24 k	91.7	74.2-97.7	85/86 k	98.8	93.7-99.8
	Para-Pak	23/24	95.8		61/61 l	100.0	94.1-100.0
	C&S			79.8-99.3			
Shiga-like toxin <i>E. coli</i> (STEC) stx1/stx2	Para-Pak C&S	60/64	93.8	85.0-97.5	44/44 m	100.0	92.0-100.0
<i>E. coli</i> O157	Para-Pak C&S	39/42 n	92.9%	80.1-99.4	16/16	100.0	80.6-100.0
<i>Shigella</i>/Enteroinvasive <i>E. coli</i> (EIEC)	FecalSwab	22/24 o	91.7	74.2-97.7	170/170	100.0	97.8-100.0
	Para-Pak	0/0	N/A		14/14	100.0	78.5-100.0
	C&S			N/A			
Parasites							
<i>Cryptosporidium</i>	FecalSwab	6/6	100.0	61-100.0	186/188 p	98.9	96.2-99.7
	Para-Pak	26/26	100.0	87.1-100.0	117/117	100.0	96.8-100.0
	C&S						
<i>Cyclospora cayentensis</i>	FecalSwab	1/1	100.0	20.7-100.0	193/193	100.0	98.1-100.0
	Para-Pak	1/1	100.0		13/13	100.0	77.2-100.0
	C&S			20.7-100.0			
<i>Entamoeba histolytica</i>	FecalSwab	0/0	N/A	N/A	194/194	100.0	98.1-100.0
	Para-Pak	0/0	N/A	N/A	14/14	100.0	76.5-100.0
	C&S						

Table 22. Clinical Performance in the Retrospective study (continued)

	Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
		TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI
<i>Giardia lamblia</i>	FecalSwab	29/31 q	93.6	79.3-98.2	46/48 q	95.8	86.0-
	Para-Pak	27/28 r	96.4		92/92 r	100.0	98.9
	C&S			82.3-99.4			96.0- 100.0

d Adenovirus F40/41 was detected in one of the three false negatives (1/3) in FecalSwab using a different FDA-cleared test method

b Astrovirus was detected in the single false negative specimen (1/1) in FecalSwab using a different FDA-cleared test method.

c two (2) FecalSwab samples positive for Norovirus GI/GII in both QIAstat-Dx and one FDA-cleared comparator were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. The sample size for NPA is smaller for Norovirus GI/GII as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the retrospective study.

d the sample size for NPA is smaller for Norovirus GI/GII in Para-Pak C&S as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the retrospective study

e Rotavirus A was detected in the single false negative specimen (1/1) in FecalSwab using a different FDA-cleared test method.

f Campylobacter was detected in one of the two false positive specimens (1/2) in FecalSwab using a different FDA-cleared test method.

g Plesiomonas shigelloides was detected in one of the three false negative specimens (1/3) in Para-Pak C&S using a different FDA-cleared test method.

h Salmonella was not detected in the single false negative specimen (0/1) and was not detected in the two false positive specimens (0/2) in FecalSwab using a different FDA-cleared test method.

i Yersinia enterocolitica was not detected in the two false negative specimens (0/2) in FecalSwab using a different FDA-cleared test method.

Table 22. Clinical Performance in the Retrospective study (continued)

Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
	TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI

j Enteropathogenic E. coli (EPEC) was detected in all three false negative specimens (3/3) in Para-Pak C&S using PCR followed by bi-directional sequence analysis. There were two (2) other false negative specimens that were not further investigated by discrepant analyses.

k ten (10) FecalSwab samples positive for ETEC in both QIAstat-Dx and one FDA-cleared comparator were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. One (1) FecalSwab sample negative in QIAstat-Dx and positive with one FDA-cleared comparator with insufficient volume for complete composite comparator testing was classed as false negative in the PPA calculations. The sample size for NPA is smaller for ETEC as only a portion of the samples with a negative result in QIAstat-Dx and one FDA-cleared comparator was tested with the complete composite comparator in the retrospective study.

l The sample size for NPA is smaller for ETEC in Para-Pak CS&S as only a portion of the samples with a negative result in QIAstat-Dx and one FDA-cleared comparator was tested with the complete composite comparator in the retrospective study).

m The sample size for NPA is smaller for STEC in Para-Pak C&S as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the retrospective study

n E. coli O157 was not detected in two false negative specimens (0/2) in Para-Pak C&S using a different FDA-cleared test method. There was one (1) false negative specimen in Para-Pak C&S that was not further investigated by discrepant analyses.

o Shigella/Enteroinvasive E. coli (EIEC) was detected in one of the two false negative specimens (1/2) in FecalSwab using a different FDA-cleared test method.

p Cryptosporidium was not detected in the two false positive specimens (0/2) in FecalSwab using PCR followed by bi-directional sequence analysis

q four (4) samples positive for Giardia lamblia in both QIAstat-Dx and one FDA-cleared comparator were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. Two (2) FecalSwab samples negative in QIAstat-Dx and positive with one FDA-cleared comparator with insufficient volume for complete composite comparator testing was classed as false negative in the PPA calculations. The sample size for NPA is smaller for Giardia lamblia as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the retrospective study.

Table 22. Clinical Performance in the Retrospective study (continued)

Medium Brand	Positive Percent Agreement			Negative Percent Agreement		
	TP/TP+FN	%	95% CI	TN/TN+FP	%	95% CI

r One (1) Para-Pak C&S samples positive for Giardia lamblia in both QIAstat-Dx and primary FDA-cleared comparator (were excluded from the PPA calculations because the samples did not have sufficient volume for complete composite comparator testing. One (1) Para-Pak C&S sample negative in QIAstat-Dx and positive with one FDA-cleared comparator with insufficient volume for complete composite comparator testing was classed as false negative in the PPA calculations. The sample size for NPA is smaller for Giardia lamblia as only a portion of the samples with a negative result in QIAstat-Dx and in one FDA-cleared comparator was tested with the complete composite comparator in the retrospective study.

The proportion of failed runs on initial attempt, and following repeats are summarized in Table 23. The error breakdown due to instrument, invalid results, ‘sample too concentrated’ failures and other run failures are summarized in Table 24.

Table 23. Failure Rates Summary

Transport media	Study	Initial Runs			Final Runs		
		N/Total	%	95% CI	N/Total	%	95% CI
FecalSwab	Prospective	16/1227	1.3	0.7 - 2.1	3/1227	0.2	0.1 - 0.7
	Prospective Archived	0/53	0.0	0.0 - 6.7	0/53	0.0	0.0 - 6.7
	Retrospective	11/366	3.0	1.5 - 5.3	5/366	1.4	0.4 - 3.2
	Total	27/1646	1.6	1.1 - 2.4	8/1646	0.5	0.2 - 1.0
Para-Pak C&S	Prospective	66/740	8.9	7.0 - 11.2	21/740	2.8	1.8 - 4.3
	Prospective Archived	3/66	4.5	0.9 - 12.7	0/66	0.0	0.0 - 5.4
	Retrospective	46/454	10.1	7.5 - 13.3	25/454	5.5	3.6 - 8.0
	Total	115/1260	9.1	7.6 - 10.9	46/1260	3.7	2.7 - 4.8

Table 24. Failure Types Breakdown

Transport Media	Study	Failure Reason	Initial Runs		Final Runs	
			N/Total	%	N/Total	%
FecalSwab	Prospective	Instrument	0/1227	0.0	0/1227	0.0
		Invalid*	0/1227	0.0	0/1227	0.0
		Sample too Concentrated†	5/1227	0.4	0/1227	0.0
		Other‡	11/1227	0.9	3/1227	0.2
	Prospective Archived	Instrument	0/53	0.0	0/53	0.0
		Invalid	0/53	0.0	0/53	0.0
		Sample too Concentrated	0/53	0.0	0/53	0.0
		Other	0/53	0.0	0/53	0.0
	Retrospective	Instrument	1/366	0.3	0/366	0.0
		Invalid	1/366	0.3	0/366	0.0
		Sample too Concentrated	0/366	0.0	0/366	0.0
		Other	9/366	2.5	5/366	1.4

Table 24. Failure Types Breakdown (continued)

Transport Media	Study	Failure Reason	Initial Runs		Final Runs	
			N/Total	%	N/Total	%
Para-Pak C&S	Prospective	Instrument	9/740	1.2	2/740	0.3
		Invalid	5/740	0.7	5/740	0.7
		Sample too Concentrated	35/740	4.7	7/740	0.9
		Other	17/740	2.3	7/740	0.9
	Prospective Archived	Instrument	0/66	0.0	0/66	0.0
		Invalid	1/66	1.5	0/66	0.0
		Sample too Concentrated	1/66	1.5	0/66	0.0
		Other	1/66	1.5	0/66	0.0
	Retrospective	Instrument	1/454	0.2	0/454	0.0
		Invalid	10/454	2.2	6/454	1.3
		Sample too Concentrated	10/454	2.2	2/454	0.4
		Other	25/454	5.5	17/454	3.7

* Internal Control failures with at least one analyte detected and the other analytes reported as 'invalid'

† Run failures related to 'sample concentration too high'. These specimens were repeated with 100 microliters as detailed in Appendix C.

‡ Run failures related to workflow checkpoints.

Co-infections

The QIAstat-Dx Gastrointestinal Panel 2 reported multiple organism detections (i.e., co-infections) for a total of 15 and 29 prospective specimens in FecalSwab and Para-Pak C&S, respectively. This represents 7.0% of positive specimens (15/213) in FecalSwab and 17.0%

of positive specimens (29/171) in Para-Pak C&S. Most multiple detections in FecalSwab specimens (14/15; 93%) contained two organisms, while 6.7% (1/15) contained three organisms. In Para-Pak C&S specimens, most multiple detections (22/29; 75.9%) contained two organisms, while 24.1% (7/29) contained three organisms. The most common multiple infections are shown in Table 25 and Table 26 below.

Table 25. Most Prevalent Multiple Detection Combinations (≥2 instances) as Determined by the QIAstat-Dx Gastrointestinal Panel 2 in the Prospective Clinical study in FecalSwab specimens

Multiple Detection Combination	Number of FecalSwab Specimens
<i>Campylobacter</i> + Norovirus GI/GII	2
Enterotoxigenic <i>E. coli</i> (ETEC) It/st + Norovirus GI/GII	3
<i>Campylobacter</i> + Rotavirus A	4

Table 26. Most Prevalent Multiple Detection Combinations (≥2 instances) as Determined by the QIAstat-Dx Gastrointestinal Panel 2 in the Prospective Clinical study in Para-Pak C&S specimens

Multiple Detection Combination	Number of Para-Pak C&S Specimens
<i>Campylobacter</i> + Enteropathogenic <i>E. coli</i> (EPEC)	3
Enteropathogenic <i>E. coli</i> (EPEC) + Salmonella	3
Enteropathogenic <i>E. coli</i> (EPEC) + Enterotoxigenic <i>E. coli</i> (ETEC) It/st	4

The analytes most commonly found in mixed infections in the FecalSwab specimens were *Campylobacter* (9), Norovirus GI/GII (7), Rotavirus (4) and ETEC (3) as shown in Table 27, while the analytes most commonly found in mixed infections in the Para-Pak C&S specimens were EPEC (17), ETEC (8), *Campylobacter* (7), Norovirus GI/GII (5), Rotavirus (4) and STEC (5) as shown in Table 28.

Table 27. Prevalence of Analytes in Mixed Infections in FecalSwab specimens as determined by the QIAstat-Dx Gastrointestinal Panel 2

Analyte	N	%
Adenovirus F40/F41	1	3.2
Astrovirus	1	3.2
<i>Campylobacter</i>	9	29.0
Enterotoxigenic <i>E. coli</i> (ETEC) lt/st	3	9.7
<i>Giardia lamblia</i>	2	6.5
Norovirus GI/GII	7	22.6
<i>Plesiomonas shigelloides</i>	1	3.2
Rotavirus A	4	12.9
<i>Shigella</i> /Enteroinvasive <i>E. coli</i> (EIEC)	2	6.5
<i>Yersinia enterocolitica</i>	1	3.2

Table 28. Prevalence of Analytes in Mixed Infections in Para-Pak C&S specimens as determined by the QIAstat-Dx Gastrointestinal Panel 2

Analyte	N	%
Adenovirus F40/F41	1	1.5
Astrovirus	1	1.5
<i>Campylobacter</i>	7	10.8
<i>Cryptosporidium</i>	2	3.1
<i>Cyclospora cayetanensis</i>	2	3.1
Enteropathogenic <i>E. coli</i> (EPEC)	17	26.2
Enterotoxigenic <i>E. coli</i> (ETEC) lt/st	8	12.3
<i>Giardia lamblia</i>	1	1.5
Norovirus GI/GII	5	7.7

Table 28. Prevalence of Analytes in Mixed Infections in Para-Pak C&S specimens as determined by the QIAstat-Dx Gastrointestinal Panel 2 (continued)

Analyte	N	%
<i>Plesiomonas shigelloides</i>	7	10.8
Rotavirus A	1	1.5
<i>Salmonella</i>	4	6.2
Shiga-like toxin <i>E. coli</i> (STEC) stx1/stx2	5	7.7
<i>Shigella</i> /Enteroinvasive <i>E. coli</i> (EIEC)	3	4.6
<i>Yersinia enterocolitica</i>	1	1.5

Contrived specimen testing

Several analytes, such as *Entamoeba histolytica* are so rare that both prospective and retrospective testing efforts were insufficient to demonstrate system performance. To supplement the prospective and retrospective specimens' test results, an evaluation of contrived specimens was performed. Contrived specimens were prepared using negative residual specimens that had previously tested negative by QIAstat-Dx Gastrointestinal Panel 2 and comparator methods. At least, 50% of these specimens were spiked at concentrations slightly above the Limit of Detection (2x LoD) and the rest at 5x and 10x LoD, using quantified strains for each pathogen. A minimum of 50 contrived specimens were tested for each evaluated analyte. The analyte status of each contrived specimen was blinded to the users analyzing the specimens. Results are summarized in Table 29 below.

Table 29. Test Results Summary for Contrived Specimens

QIAstat-Dx GI2 Target	Positive Percent Agreement (PPA)			
	Medium Brand	Fraction	Percentage	95% CI
Astrovirus	FecalSwab	33/34	97.1	85.1-99.5
	Para-Pak C&S	34/34	100.0	89.8-100.0

Table 29. Test Results Summary for Contrived Specimens (continued)

QIAstat-Dx GI2 Target	Positive Percent Agreement (PPA)			
	Medium Brand	Fraction	Percentage	95% CI
Rotavirus A	FecalSwab	35/35	100.0	90.1-100.0
	Para-Pak C&S	34/35	97.1	85.5-99.5
<i>Plesiomonas shigelloides</i>	FecalSwab	33/33	100.0	89.6-100.0
	Para-Pak C&S	34/35	97.1	85.5-99.5
<i>Yersinia enterocolitica</i>	FecalSwab	34/34	100.0	89.8-100.0
	Para-Pak C&S	34/35	97.1	85.5-99.5
<i>Shigella</i> /EIEC	FecalSwab	35/35	100.0	90.1-100.0
	Para-Pak C&S	34/34	100.0	89.8-100.0
<i>Cryptosporidium</i>	FecalSwab	27/27	100.0	87.5-100.0
	Para-Pak C&S	31/31	100.0	89.0-100.0
<i>Cyclospora cayetanensis</i>	FecalSwab	26/26	100.0	87.1-100.0
	Para-Pak C&S	30/30	100.0	88.6-100.0
Entamoeba histolytica	FecalSwab	35/35	100.0	90.1-100.0
	Para-Pak C&S	34/35	97.1	85.5-99.5

Troubleshooting Guide

This troubleshooting guide may be helpful in solving any problems that may arise. For more information, see also the Frequently Asked Questions page at our Technical Support Center: www.qiagen.com/FAQ/FAQList.aspx (for contact information, visit www.qiagen.com).

Additional information about specific QIAstat-Dx Gastrointestinal Panel 2 error codes and messages can be found in Table 30:

Table 30. Information about specific QIAstat-Dx Gastrointestinal Panel 2 error codes and messages

Error Code	Error message displayed
0x02C9	Cartridge execution failure: Sample concentration too high.
0x032D	Please repeat by loading 100 microliters of the sample in a new cartridge (as per Appendix C
0x0459	explanation)
0x045A	
0x04BF	
0x0524	
0x058B	
0x05E9	
0x0778	
0x077D	
0x14023	


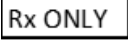








When the sample concentration is too high and the test must be repeated by loading 100 µL, follow the workflow detailed in the Appendix C of this document.











Contact Information

For technical assistance and more information, please see our Technical Support Center at www.qiagen.com/Support, call 00800-22-44-6000, or contact one of the QIAGEN Technical Service Departments or local distributors (see back cover or visit www.qiagen.com).

Symbols

The following symbols appear in the instructions for use or on the packaging and labeling:

Symbol	Symbol definition
	Contains reagents sufficient for <N> reactions
	Prescription Use only
	Use by
	In vitro diagnostic medical device
	Catalog number
	Lot number
	Material number (i.e., component labeling)
	Global Trade Item Number
	Unique Device Identifier
	Contains

Symbol	Symbol definition
	Component
	Number
	Gastrointestinal application
Rn	R is for revision of the Instructions for Use and n is the revision number
	Temperature limitation
	Manufacturer
	Consult instructions for use downloadable from resources.qiagen.com/674623
	Protect from light
	Do not reuse
	Caution, consult accompanying documents
	Do not use if package is damaged

Symbol

Symbol definition



Flammable, risk of fire



Corrosive, risk of chemical burn



Health Hazard, risk of sensitization, carcinogenicity



Risk of harm

Appendices

Appendix A: Installing the Assay Definition File

The Assay Definition File (ADF) of the QIAstat-Dx Gastrointestinal Panel 2 must be installed on the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 prior to testing with QIAstat-Dx Gastrointestinal Panel 2 Cartridges.

Note: Whenever a new version of the QIAstat-Dx Gastrointestinal Panel 2 assay is released, the new QIAstat-Dx Gastrointestinal Panel 2 Assay Definition File must be installed prior to testing.

The Assay Definition File (.asy file type) is available at www.qiagen.com. The Assay Definition file (.asy file type) must be saved onto a USB Drive prior to installation on the QIAstat-Dx Analyzer 1.0. This USB Drive must be formatted with a FAT32 file system.

To import an ADF from the USB to the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0, proceed with the following steps:

1. Insert the USB stick containing the Assay Definition File into one of the USB ports on the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0.
2. Press the **Options > Assay Management**. The Assay Management screen appears in the Content area of the display (Figure 23).

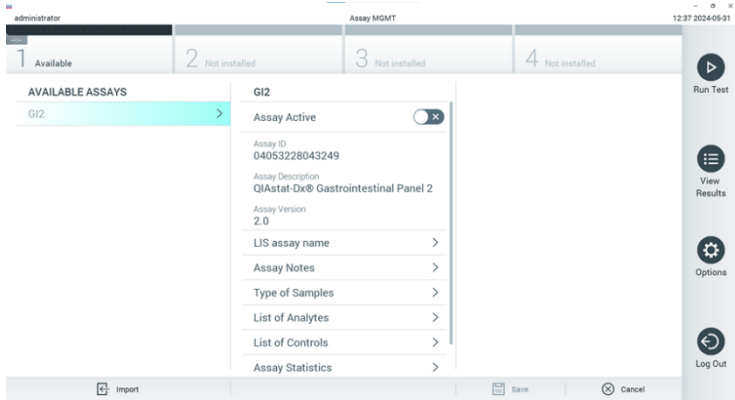


Figure 23. Assay Management screen.

3. Press **Import** in the bottom left of the screen (Figure 23).
4. Select the file corresponding to the assay to be imported from the USB drive.
A dialog box will appear to confirm upload of the file.
5. A dialog box may appear to override the current version by a new one. Press **yes** to override (Figure 24).

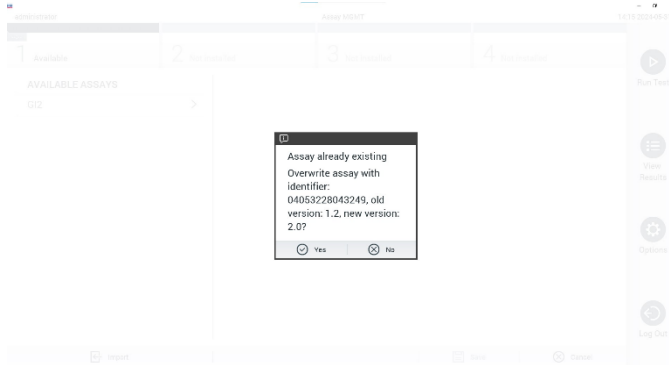


Figure 24. Dialog box that appears when upgrading the ADF version.

6. The assay becomes active by selecting Assay Active (Figure 25).

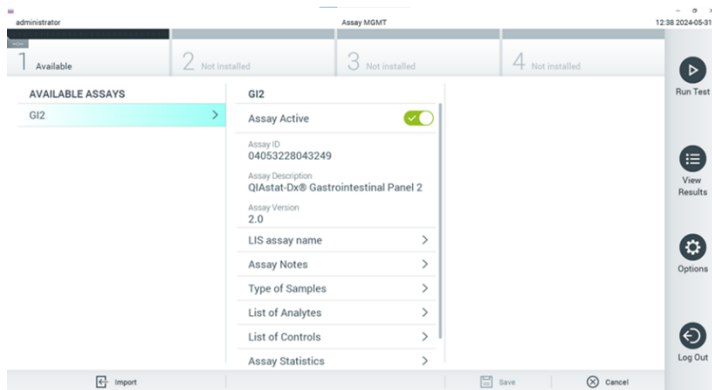


Figure 25. Activating the assay.

7. To assign the active assay to a user, perform these steps:
 - a. Go to **Options > User Management**.
 - b. Select the user who should be allowed to run the assay.

Note: If needed, this step can be repeated for every user created in the system.

- c. Select **Assign Assays** from the User Options tab.
- d. Enable the assay, then press **Save** (Figure 26).

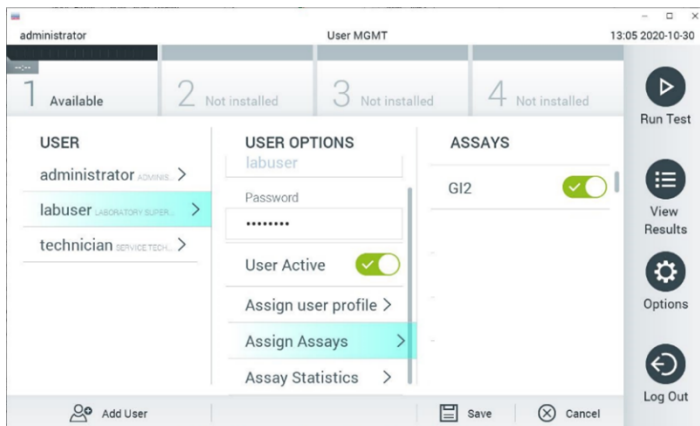


Figure 26. Assigning the active assay.

Appendix B: Glossary

Amplification curve: Graphical representation of the multiplex real-time RT-PCR amplification data.

Analytical Module (AM): The main QIAstat-Dx Analyzer 1.0 or QIAstat-Dx Analyzer 2.0 hardware module, in charge of executing tests on QIAstat-Dx Gastrointestinal Panel 2 Cartridges. It is controlled by the Operational Module. Several Analytical Modules can be connected to one Operational Module.

IFU: Instructions For Use.

Main port: In the QIAstat-Dx Gastrointestinal Panel 2 Cartridge, inlet for transport medium liquid samples.

Nucleic acids: Biopolymers, or small biomolecules composed of nucleotides, which are monomers made of three components: a 5-carbon sugar, a phosphate group and a nitrogenous base.

Operational Module (OM): The dedicated QIAstat-Dx Analyzer 1.0 hardware that provides the user interface for 1–4 Analytical Modules (AM).

Operational Module PRO (OM PRO): The dedicated QIAstat-Dx Analyzer 2.0 hardware that provides the user interface for 1-4 Analytical Modules (AM).

PCR: Polymerase Chain Reaction.

QIAstat-Dx Analyzer 1.0: The QIAstat-Dx Analyzer 1.0 consists of an Operational Module and an Analytical Module. The Operational Module includes elements that provide connectivity to the Analytical Module and enables user interaction with the QIAstat-Dx Analyzer 1.0. The Analytical Module contains the hardware and software for sample testing and analysis.

QIAstat-Dx Analyzer 2.0: The QIAstat-Dx Analyzer 2.0 consists of an Operational Module PRO and an Analytical Module. The Operational Module PRO includes elements that provide connectivity to the Analytical Module and enables user interaction with the QIAstat-Dx Analyzer 2.0. The Analytical Module contains the hardware and software for sample testing and analysis.

QIAstat-Dx Gastrointestinal Panel 2 Cartridge: A self-contained disposable plastic device with all pre-loaded reagents required for the complete execution of fully automated molecular assays for the detection of gastrointestinal pathogens.

RT: Reverse Transcription.

Swab port: In the QIAstat-Dx Gastrointestinal Panel 2 Cartridge, inlet for dry swabs. The swab port is not used for the QIAstat-Dx Gastrointestinal Panel 2 assay.

User: A person who operates the QIAstat-Dx Analyzer 1.0/QIAstat-Dx Analyzer 2.0/QIAstat-Dx Gastrointestinal Panel 2 Cartridge in the intended way.

Appendix C: Additional instructions for use

In case cartridge execution failures corresponding to error codes (0x02C9, 0x032D, 0x0459, 0x045A, 0x04BF, 0x0524, 0x058B, 0x05E9, 0x0778, 0x077D, 0x14023) occur during the testing, the following error message will be displayed in the QIAstat-Dx Analyzer 1.0 or the QIAstat-Dx Analyzer 2.0 screen after the run has finalized:

Cartridge execution failure: Sample concentration too high. Please repeat by loading 100 microliters of the sample in a new cartridge (as per IFU explanation)'.

In this case the test should be repeated using 100 μ L of the same sample following equivalent testing procedures detailed in the "Procedure" Section in the IFU adapted to 100 μ L sample input volume:

1. Open the package of a new QIAstat-Dx Gastrointestinal Panel 2 Cartridge using the tear notches on the sides of the packaging.
2. Remove the QIAstat-Dx Gastrointestinal Panel 2 Cartridge from the packaging.
3. Manually write the sample information, or place a sample information label, on the top of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge. Make sure that the label is properly positioned and does not block the lid opening.
4. Place the QIAstat-Dx Gastrointestinal Panel 2 Cartridge flat on the clean work surface so that the barcode on the label faces upwards. Open the sample lid of the main port on the front of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge.

5. Thoroughly mix the stool in the transport medium, for example, by vigorously agitating the tube 3 times.
6. Open the tube with the sample to be tested. Use the supplied transfer pipette to draw up fluid. Draw the sample to the first fill line on the pipette (i.e., 100 μ L).

Important: Do not draw air, mucus, or particles into the pipette. If air, mucus, or particles are drawn into the pipette, carefully expel the sample fluid in the pipette back into the sample tube and draw up fluid again.

7. Carefully transfer the sample into the main port of the QIAstat-Dx Gastrointestinal Panel 2 Cartridge using the supplied single-use transfer pipette (Figure 7).
8. Firmly close the lid of the main port until it clicks (Figure 8).

From this point proceed following the instructions described in the IFU.

Ordering Information

Product	Contents	Cat. no.
QIAstat-DxGastrointestinal Panel 2	For 6 tests: 6 individually packaged QIAstat-DxGastrointestinal Panel 2 Cartridges and 6 individually packaged transfer pipettes	691421
RELATED PRODUCTS		
QIAstat-Dx Analyzer 1.0	1 QIAstat-Dx Analytical Module, 1 QIAstat-Dx Operational Module and related hardware and software to run molecular diagnostic QIAstat-Dx assay cartridges	9002824
QIAstat-Dx Analyzer 2.0	1 QIAstat-Dx Analytical Module, 1 QIAstat-Dx Operational Module PRO and related hardware and software to run molecular diagnostic QIAstat-Dx assay cartridges	9002828

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit Instructions for Use. QIAGEN kit Instructions for Use are available at www.qiagen.com or can be requested from QIAGEN Technical Services or your local distributor.

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Document Revision History

Revision	Description
Revision 1, June 2024	Initial release
Revision 2, July 2024	Inclusion of QIAstat-Dx Analyzer 2.0

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