A Field Evaluation of Cholera Rapid Diagnostic Tests

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Surveillance with RDTs is now a possibility

Public health surveillance for cholera

Guidance Document 2024



Gavi 🚷

cholera

PROGRAMMES & IMPACT INVESTING IN GAVE OUR ALLIANCE

Diagnosis for all



Global deployment of

rapid diagnostic tests to boost fight against

More than 1.2 million cholera rapid diagnostic tests will be shipped to 14 countries in largest-ever global deployment, with the first shipment landing today in Malawi.

This first official deployment of tests through Gavi, the Vaccine Alliance, will improve timely detection and monitoring of outbreaks, effectiveness of vaccination campaigns in response to current outbreaks, and targeting of future preventive vaccination efforts.

The global cholera rapid diagnostic test procurement programme is a collaboration between Gavi, WHO, UNICEF, FIND, and other partners.

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But the test performance of RDTs is unknown

Muzembo et al, 2021; PMID: 34506946

Study, index test, year		SENSITIVITY (95% CI)	Study, index test, year		SPECIFICITY (95% CI)
George, Crystal VC, 2014	-	0.66 [0.53 - 0.77]	George, Crystal VC, 2014	-	0.92 [0.82 - 0.97]
Islam, Crystal VC, 2019	*	0.72 [0.65 - 0.79]	Islam, Crysta/VC, 2019		0.77 [0.76 - 0.78]
Islam, Crystal VC, 2019		0.72 [0.51 - 0.88]	Islam, Cholkit, 2019	i w	0.87 (0.83 - 0.90)
Islam, Cholkit, 2019		0.76 (0.55 - 0.91)	Islam, Cholkit, 2019		0.90 [0.87 - 0.93]
Wang, IP, 2006		0.77 (0.46 - 0.95)	Wang, IP, 2006		0.78 (0.65 - 0.88)
Page, Crystal VC, 2012	*!	0.79 (0.72 - 0.85)	Page, Crystal VC, 2012		0.69 [0.59 - 0.78]
Islam, Cholkit, 2019	-	0.79 [0.62 - 0.91]	Islam, Cholkit, 2019		0.87 [0.86 - 0.89]
Matias, SD Bioline, 2017	-	0.81 [0.76 - 0.86]	Matias, SD Bioline, 2017	1.	0.93 [0.88 - 0.96]
Page, Crystal VC, 2012	-	0.88 [0.83 - 0.92]	Page, Crystal VC, 2012	*	0.89 (0.79 - 0.95)
Ley, Crystal VC, 2012	*	0.90 (0.85 - 0.93)	Ley, Crystal VC, 2012	* i	0.47 (0.43 - 0.52)
Mwaba, SD Bioline, 2018	-*	0.91 [0.81 - 0.97]	Mwabe, SD Bioline, 2018		0.95 [0.89 - 0.98]
Page, Crystal VC, 2012	*	0.92 [0.87 - 0.96]	Page, Crystal VC, 2012	-=-	0.71 [0.61 + 0.79]
Mukherjee, Crystal VC, 2010	*	0.92 [0.83 - 0.97]	Mukherjee, Crystal VC, 2010	-#1	0.73 (0.65 - 0.80)
Page, Crystal VC, 2012	+	0.92 [0.87 - 0.95]	Page, Crystal VC, 2012		0.83 (0.72 - 0.91)
Chibwe, Cholkit, 2020	-#	0.93 (0.83 - 0.96)	Chibwe, Cholkit, 2020	i-#	0.96 [0.78 - 1.00]
Ontweka, Crystal VC, 2016	-	0.94 [0.81 - 0.99]	Ontweka, Crysta/VC, 2016		0.69 [0.56 - 0.80]
Denue, Crystal VC, 2018	100	0.95 (0.89 - 0.98)	Denue, Crystal VC, 2018	-*-i	0.59 [0.45 - 0.72]
Boncy, Crystal VC, 2013	1	0.95 [0.93 - 0.97]	Boncy, Crystal VC, 2013	*	0.80 [0.75 - 0.85]
Harris, Crystal VC, 2009		0.97 [0.90 - 1.00]	Harris, Crystal VC, 2009		0.75 [0.57 - 0.89]
Wang, IP, 2006	18	0.97 (0.88 - 1.00)	Wang, IP, 2006		0.76 [0.60 - 0.88]
Matias, Artron, 2017		0.99 (0.93 - 1.00)	Matias, Artron, 2017		0.64 (0.50 + 0.76)
Matias, Crystal VC, 2017	im	0.99 (0.97 - 1.00)	Matias, Crystal VC, 2017	* i	0.60 [0.53 - 0.66]
Sayeed, Crystal VC, 2018		1.00 [0.82 - 1.00]	Sayeed, Crystal VC, 2018	*	0.81 [0.68 - 0.90]
Sayeed, Cholkit, 2018		1.00 [0.82 - 1.00]	Sayeed, Choikit, 2018	*	0.81 [0.68 - 0.90]
Pooled		0.91 [0.87 - 0.94]	Pooled	\$	0.80 [0.74 - 0.84]
		Q = 250.64, df = 23.00, p = 0.00		1	Q = 518.00, df = 23.00, p = 0.00
		12 = 90.82 [88.08 - 93.56]			12 = 95.56 [94.50 - 96.62]
	0.5 1.0			0.4 1.0	

RDT clinical sensitivity and specificity is highly variable

Muzembo et a	, 2021; PMID:	34506946
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Islam, Crystal VC, 2019	*	0.72 (0.65 - 0.79)	Islam, Crysta/VC, 2019		6.77 (0.76 - 0.78)
Islam, Crystal VC, 2019		0.72 [0.51 - 0.88]	Islam, Cholkit, 2019	18	0.87 (0.83 - 0.90)
Islam, Chokit, 2019		0.76 (0.55 - 0.91)	Islam, Cholkil, 2019		0.90 (0.87 - 0.93)
Wang, IP, 2006		0.77 (0.46 - 0.95)	Wang, IP, 2006		0.78 (0.65 - 0.86)
Page, Crystal VC, 2012	-	0.79 (0.72 - 0.86)	Page, Crystal VC, 2012	-	0.69 (0.59 - 0.78)
Islam, Cholkit, 2019	-8-	0.79 [0.42 - 0.91]	Islam, Chokil, 2019		0.87 (0.86 - 0.89)
Matian, SD Bioline, 2017	-	0.81 (0.76 - 0.86)	Mattan, SD Bioline, 2017		0.85 (0.86 - 0.96)
Page, Crystal VC, 2012		0.88 (0.83 - 0.92)	Page, Crystal VC, 2012		0.89 (0.79-0.95)
Ley, Crystal VC, 2012		0.90 (0.85 - 0.90)	Ley, Crystal VC, 2012	* i	0.47 (0.43 - 0.52)
Meaba, SD Boline, 2018	+	0.91 (0.81 - 0.97)	Mwaba, SD Bioline, 2018		0.95 (0.96)
Page, Crystal VC, 2012	+	0.92 (0.87 - 0.94)	Page, Crystal VC. 2012	-	6.71 (0.61 - 0.79)
Akherine, Crystal VC, 2010	+	0.82 (0.83 - 0.97)	Mukherjee, Crystal VC, 2010	-#1	0.73 (0.65 - 0.60)
Page, Crystal VC, 2012	+	0.92 (0.87 - 0.96)	Page, Crystal VC, 2012	-	8.83 (0.72 - 0.91)
Chiltwe, Chulkit, 2020	-	0.93 (0.80 - 0.94)	Chiltere, Cholkil, 2020		0.96 (0.78 - 1.00)
Ontwelka, Crystal VC, 2016	-	0.94 (0.81 - 0.99)	Ontwelka, CrystalVC, 2016		0.69 (0.56 - 0.60)
Denue, Crystal VC, 2018		0.95 (0.89 - 0.96)	Denue, Crystal VC, 2018	-#-i	0.59 (0.45 - 0.72)
Boncy, Crystal VC, 2013		0.95 [0.93 - 0.97]	Boncy, Crystal VC, 2013	+	0.80 (0.75 - 0.85)
Harris, Crystal VC, 2009		0.87 [0.90 - 1.00]	Hamia, Crystal VC, 2009		0.75 (0.57 - 0.89)
Wang.IP, 2006		0.97 (0.88 - 1.00)	Wang, IP; 2006		0.76 (0.60 - 0.66)
Mattas, Artron, 2017		0.99 (0.93 - 1.00)	Matian, Artron, 2017		0.64 (0.50 - 0.76)
Matian, Crystal VC, 2017	im	0.99 (0.97 - 1.00)	Mattan, Crystal VC, 2017	* 1	0.00 (0.53 - 0.66)
Sayeed, Crystal VC, 2018	-1=	1.00 [0.82 - 1.00]	Sayweet, Crystal VC, 2018	-+-	0.81 (0.68 - 0.90)
Sayned, Chokit, 2018		1.00 (0.82 - 1.00)	Sayeed, Choke, 2018	+	0.81 (0.0-98.0)
Pooled	4	0.91 (0.87 - 0.94)	Pooled	4	0.80 (0.74 - 0.84)
	- i -	G = 250.64, df = 23.00, p = 0.00			Q=518.00, df=23.00, p= 0.00
		12 × 90.82 (88.08 - 93.56)			12 = 95.56 [94.50 - 96.62]
	0.5 1.0			0.4 1.0 BPECIFICITY	

Variability may be attributable to differences in:

- Test type/manufacturer
- Pre-treatment of stool
- Sample type (stool/rectal swab)
- Reader errors
- Phage and antibiotics
- Age groups
- Gold standard used

Primary Objective

To estimate the **sensitivity** and **specificity** of commercially available lateral flow RDTs for *V. cholerae* compared to PCR as the reference standard among medically attended symptomatic suspected cholera cases *(under optimal testing conditions)*

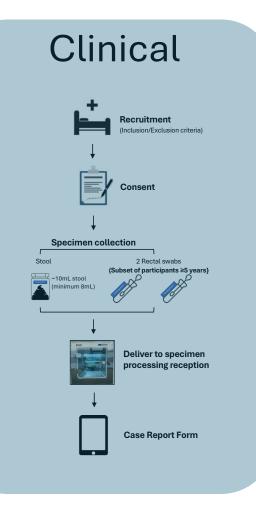
Secondary Objectives

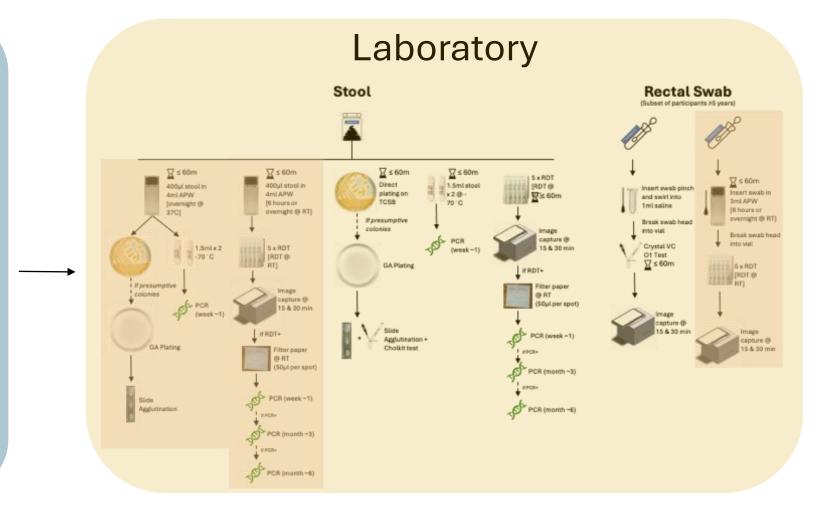
- 1. To estimate how test performance varies as a function of (a) gold standard, (b) enrichment, (c) sample type, (d) antibiotic use, (e) disease severity, and (f) age and season
- 2. To quantify the color intensity of the RDT control and test bands through an image processing algorithm and compare to technician classification
- 3. To quantify the loss in PCR test sensitivity over time from dry filter paper samples left in ambient conditions
- 4. To compare the use of RDTs for testing suspected colonies to slide agglutination

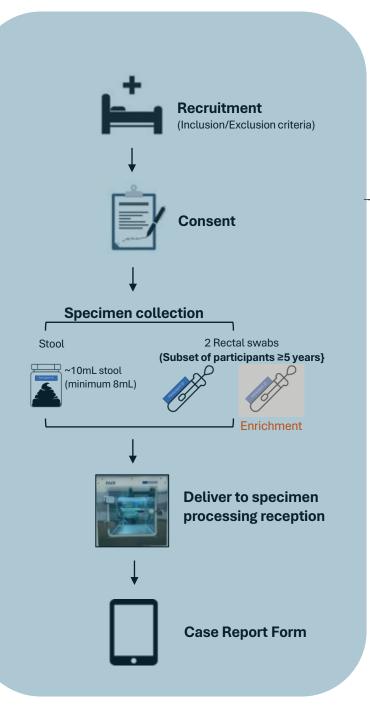
Study Design

- Setting: 'Ideal' setting of <u>icddr,b Dhaka hospital</u> to ensure rapid and highquality processing of samples
- Sample population: patients seeking care at icddr,b hospital with acute watery diarrhea
- Sample size: stratified sample by age group (1-4 and 5+) and season (pre-monsoon, monsoon, post-monsoon and winter) with 3,292 AWD cases being recruited
- *Tests:* 5 commercially available lateral flow RDTs - *Crystal VC 01, Crystal VC 01/0139, Artron, Abbott, Cholkit*

Study Design







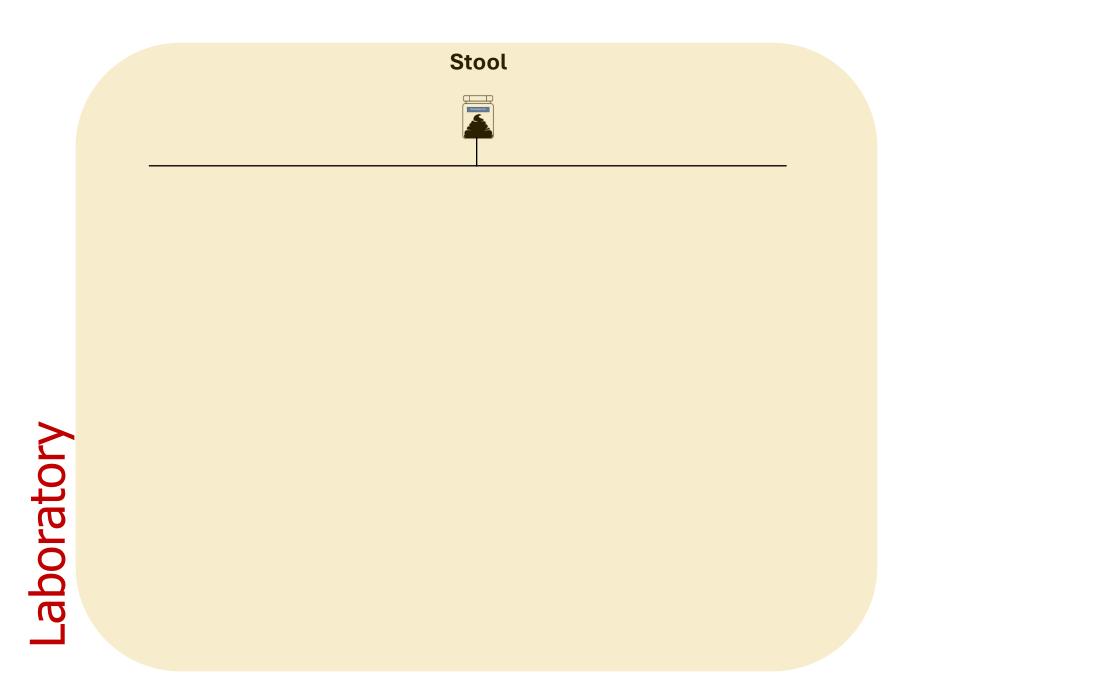
Clinical

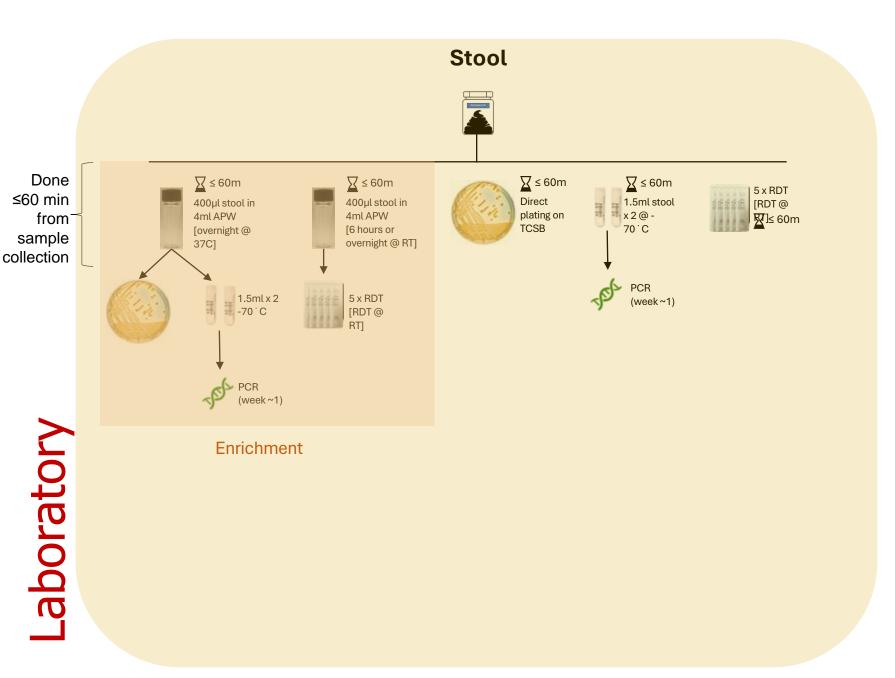
Inclusion criteria

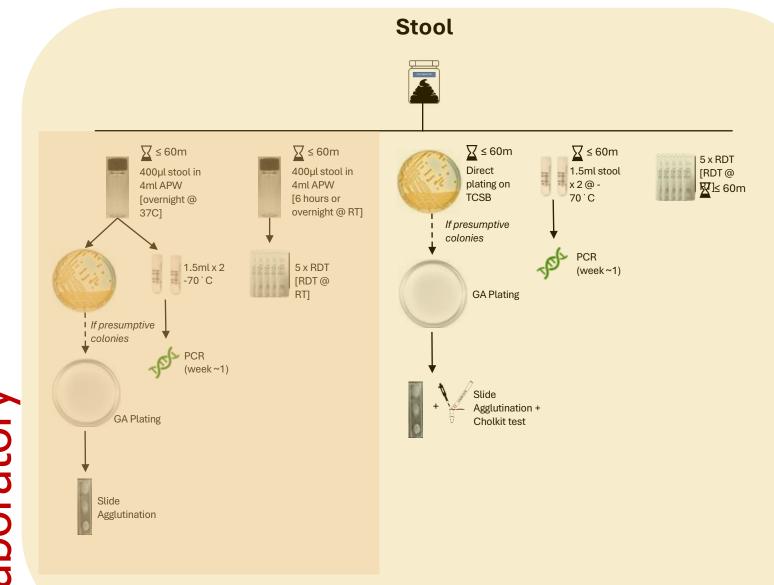
- AWD (≥3 or more loose stools within 24-hour period) and symptoms in last 24 hours (having lasted <8 days before hospital visit)
- Non-bloody stool
- Purged stool in 3 hours before study enrollment
- Willing and able to provide a stool sample and rectal swab

Exclusion criteria

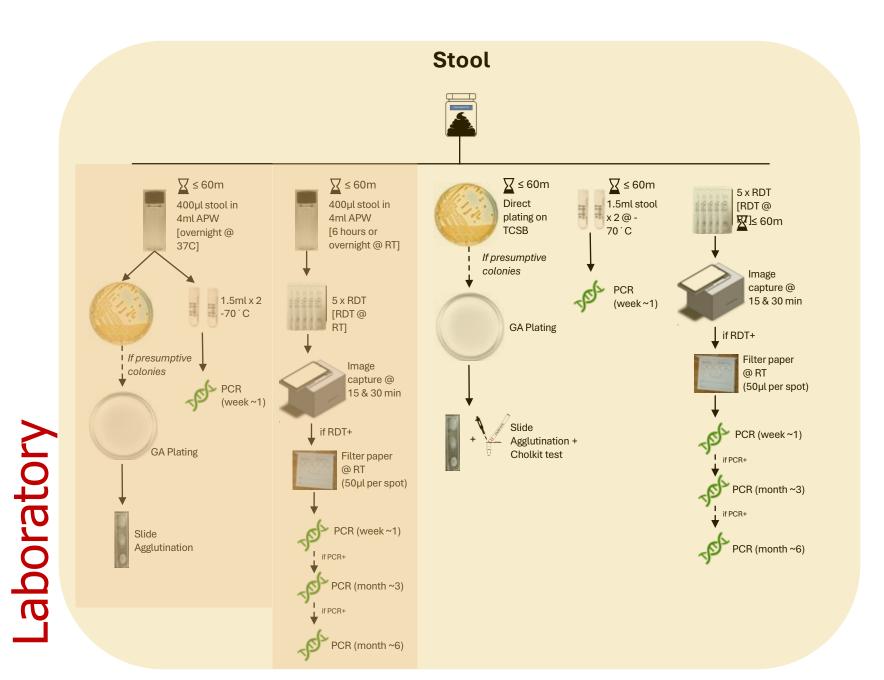
• Administered antibiotics for treatment after entering icddr,b







Laboratory



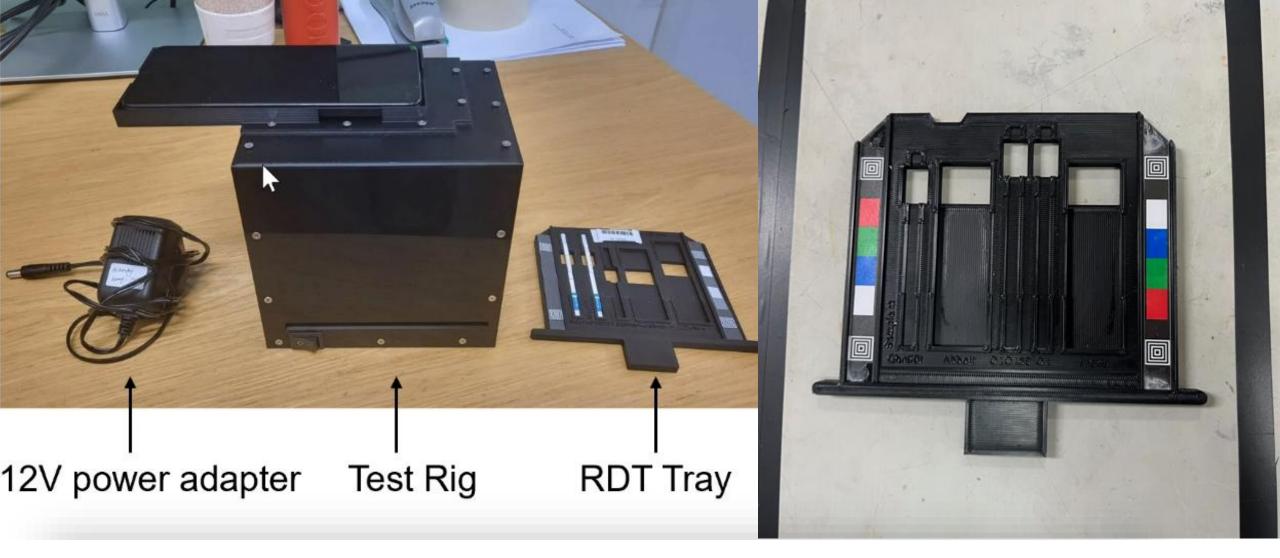
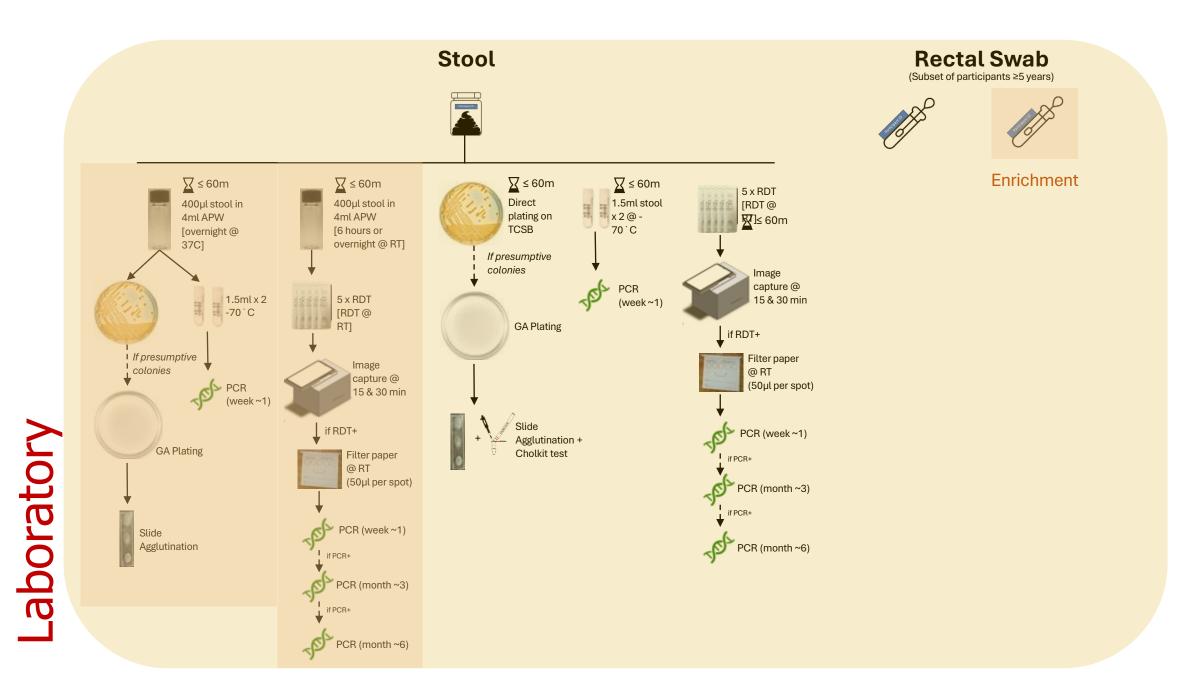
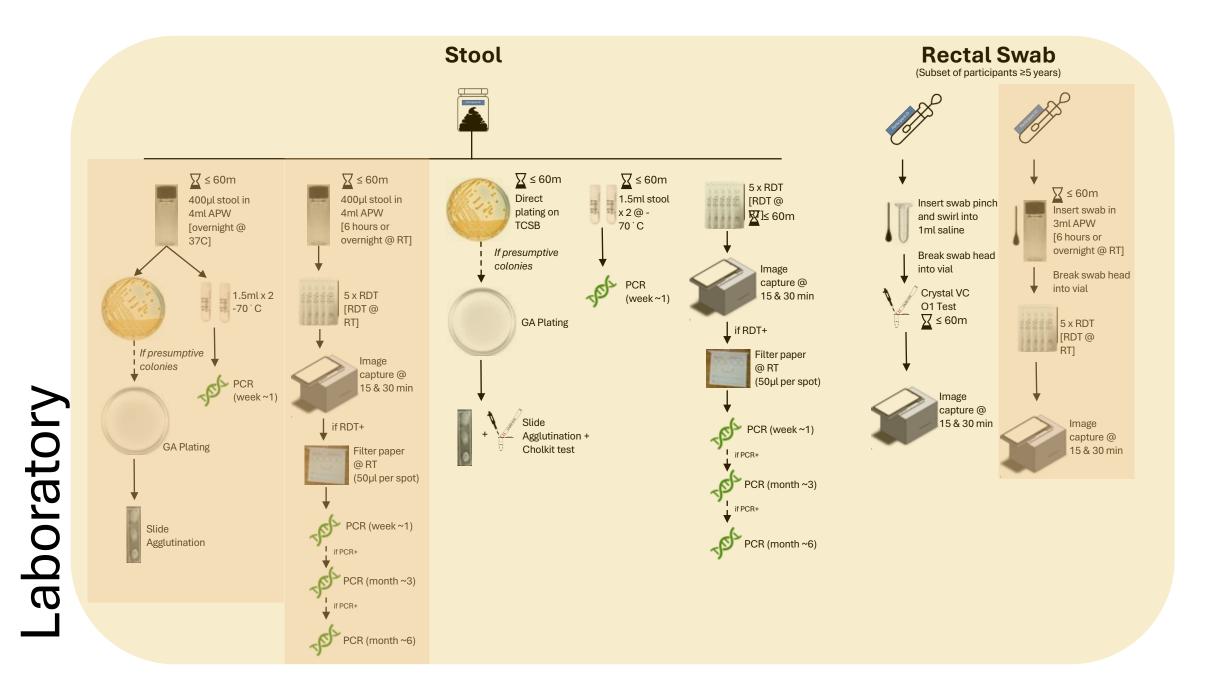


Image capture rig



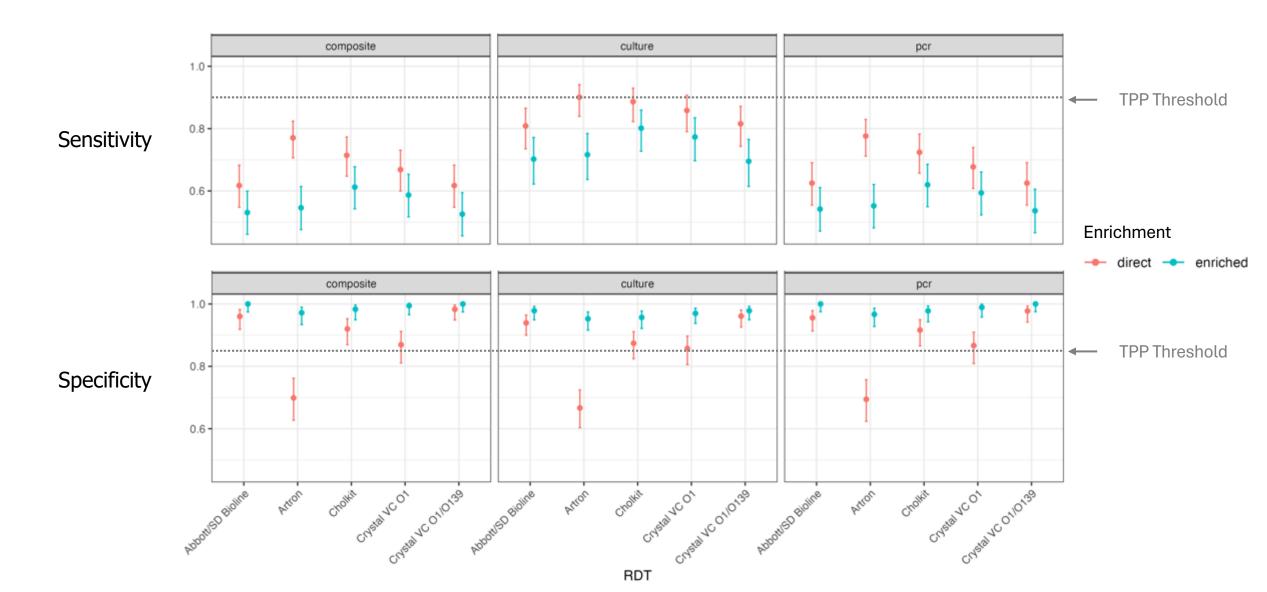




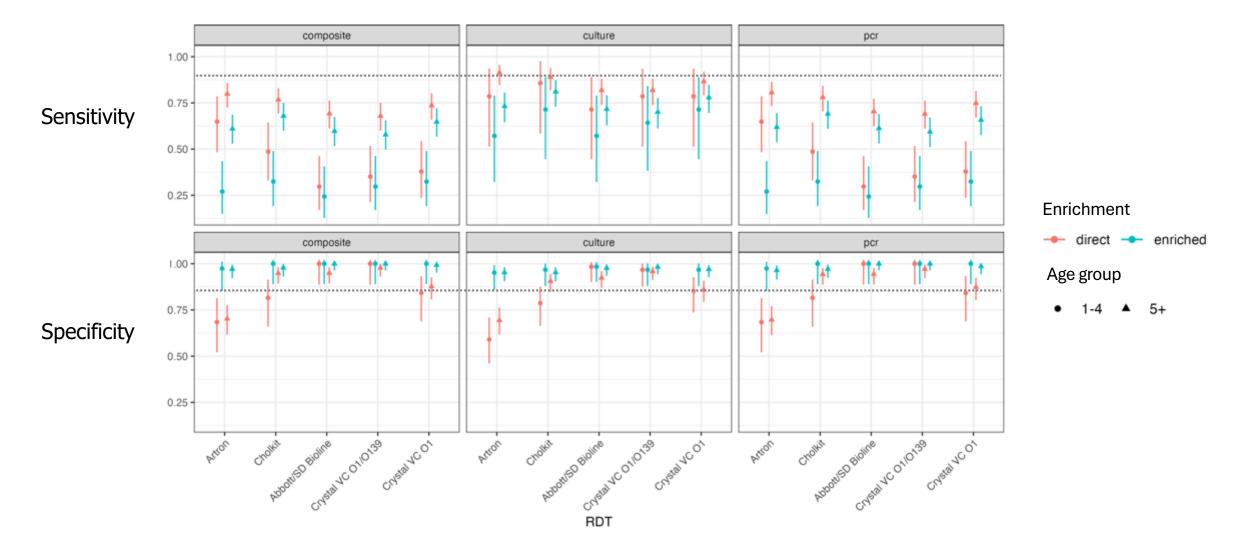
Enrollment to date

	Winter (November – February)	Pre-monsoon (March – June)	Monsoon (July – August)	Post-monsoon (September – October)	Total
1-4 years target	0 suspected cases (0 culture confirmed)	1891 suspected cases (104 culture confirmed)	0 suspected cases (0 culture confirmed)	0 suspected cases (0 culture confirmed)	1891 suspected cases (104 culture confirmed)
1-4 years actual	0 suspected (0 culture confirmed)	102 suspected cases (20 confirmed)			102 suspected cases (20 confirmed)
≥5 years target	296 suspected cases (52 culture confirmed)	206 suspected cases (52 culture confirmed)	532 suspected cases (52 culture confirmed)	367 suspected cases (52 culture confirmed)	1401 suspected cases (208 culture confirmed)
≥5 years actual	98 suspected cases (32 culture confirmed)	199 suspected cases (95 culture confirmed)			297 suspected cases (127 culture confirmed)

RDT test performance (all ages)



RDT test performance (by age group)

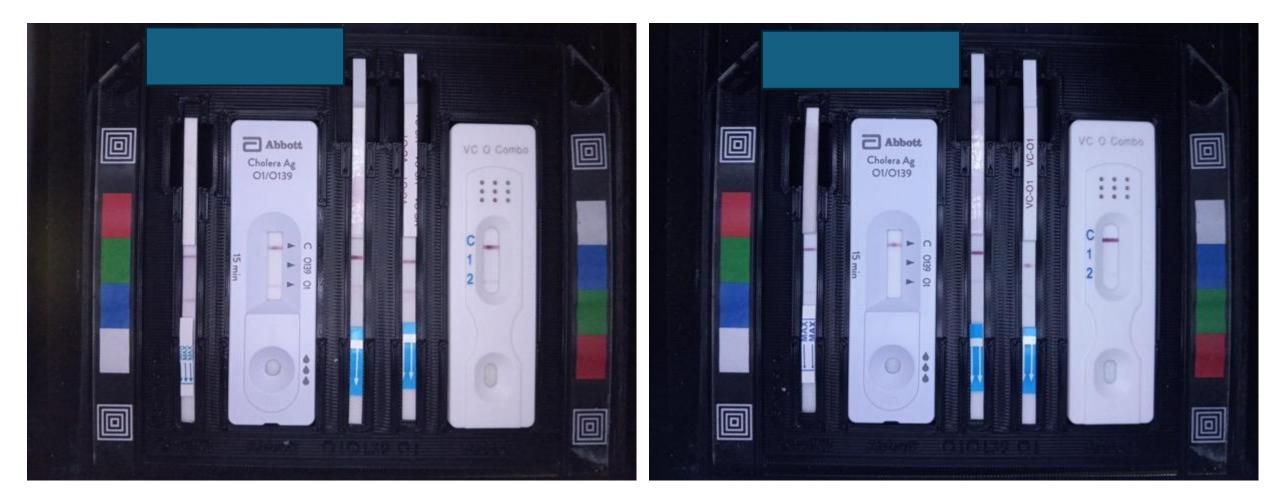


Observation of false positive VC 0139 results

	Direct	Enriched
Abbott/SD	1.3% (5/372)	0.0% (0/382)
Crystal VC 01/0139	11.0% (42/372)	1.3% (5/382)

*compared to PCR and culture

Standardized photos provide more nuanced insights into test performance





Interim Conclusions (interpret with caution)

- Sensitivity
 - Three tests, when used directly on stool, demonstrate performance consistent with TPP minimum of 90% sensitivity using culture as gold standard
 - No tests, direct or enriched, when compared to PCR meet TPP sensitivity minimum
- Specificity
 - All tests but one, when used directly or with enrichment, meet or exceed the TPP 80% specificity threshold.
- Age
 - Sensitivity of all tests appears reduced in young children 1-4 years old compared to others with larger effects when PCR is used
- Enrichment
 - Enrichment decreases sensitivity of all tests consistently
 - Enrichment increases specificity though variable across tests

Next Steps

- Complete enrollment
 Procure RDTs for remainder of study
 Continue to monitor results
 - Latent class analyses taking into account lack of gold standard, antibiotics, age, season

Acknowledgements

icddr,b

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Government of the People's Republic of Bangladesh



