CIDT Technical Workgroup Report
March 2023

I. Charge

The Vp Illness Response Committee requests the CIDT Technical Workgroup reconvene to provide the committee with information regarding:

1. current data on what percentage of CIDT+ are being culture confirmed
2. if there are states or regions where culture confirmation is lacking,
3. now that CIDT has been in use for 5 years, updated data regarding false positives and false negatives
4. are different CIDT tests comparable
5. what are the frequency of co-detections and the frequency of accurate reporting of co-detection
6. what are the predominant CIDT tests being conducted
7. what percentage are Vp specific (such as Diatherix).

II. Members

1. Jessica Jones (Chair) – FDA
2. Robert Schuster – New Jersey
3. AJ Erskine – Bevans Oyster
4. Stephen Combes – Maine
5. Laurie Stewart – Washington
6. Christina Grant – California
7. Kirk Wiles – Texas
8. Elisa Elliot – FDA
9. Shannara Lynn – NOAA
10. Michael Hughes – CDC

III. Response to Charge

The CIDT Technical Workgroup met on multiple occasions to discuss the charge and identify sources of data appropriate to use for response to the specific questions in the charge. There were discussions about appropriateness of data as many of the data sources, including ones used here, are either very limited (in time or in area of the country) or are from passive surveillance. Passive surveillance data is potentially limited by underreporting and/or bias (not representative) based on the nature of data collection. The Workgroup has attempted to provide the most complete responses to the specific questions of the charge, but it is important to note that there are limitations to collecting and interpreting laboratory data reported to state and national public health authorities. Provided below are scenarios that can lead to gaps in knowledge regarding CIDT testing and should be considered when interpreting the data included in this report (examples below from one state, but most likely similar in other states):

- Clinical labs may perform reflex cultures on CIDT-positive specimens and if the culture is positive, the lab may only report the culture result to so the original CIDT-positive result is unknown to public health.

- Clinical labs may perform reflex cultures on CIDT-positive specimens and if the culture result is negative, the lab may only report the positive CIDT result so the negative clinical lab culture result is unknown to public health.
- Clinical labs may perform reflex cultures on CIDT-positive specimens and if the culture result is negative, the lab may not report the case at all to public health assuming that the case has been ruled out. In this scenario, the original CIDT result would be totally unknown to public health.
- Clinical settings (medical offices) and some clinical labs lack the ability to perform reflex culture on CIDT-positive specimens. If the clinical lab does not submit the stool specimen to a public health lab for culture then any potential culture results (positive or negative) are unknown.
- Clinical labs that perform culture in-house are expected to perform reflex culture more often. If such labs test a high volume of specimens, this could skew the data.
- A higher CIDT culture positive rate from clinical labs located closer to the PHL could skew the data, especially if they test a high volume of specimens.
- There is a *potential impact of seasonality*, with higher culture-confirmation observed in summer.

Table 1. CIDT-positive/ Culture-confirmed by Month (Washington State, 2021-2022).

Month	CIDT-positive/ culture not attempted	CIDT-positive/ culture-negative	CIDT-positive/ culture-confirmed	Percent Culture Confirmed*
January	1	2	0	0
February	0	1	0	0
March	2	2	1	33.3
April	1	1	1	50.0
May	1	0	0	0
June	3	4	0	0
July	12	13	38	74.5
August	9	10	29	74.4
September	2	6	8	57.1
October	2	2	4	66.7
November	2	1	0	0
December	1	3	2	40.0
TOTAL	**36**	**45**	**83**	**64.8**

*Of cases with culture attempted.

1. *Current data on what percentage of CIDT+ are being culture confirmed:*

- It is important to note that there are a variety of reasons that a CIDT-positive result may not be culture confirmed. Culture confirmation is expected to be less than 70% for other pathogens, such as *Salmonella* (Voetsch et al., 2004), and *Vibrio* spp. are known to be more difficult to culture.

Table 2. CIDT-positive cases and culture outcome.

Data Source (Years)	Total CIDT positives	CIDT-positive/ culture not attempted or reported	CIDT-positive/ culture-negative	CIDT-positive/ culture-confirmed	% Culture-confirmed (of culture attempted)
COVIS (2017-2019)[1]	3,718	1,281	1,469	968	39.7
Decuir (2016-2018)[2]	100	0	53	47	47.0
Washington (2021-2022)[3]	164	36	45	83	64.8

[1]CIDT-positive *Vibrio* infections reported to Cholera and Other *Vibrio* Illness Surveillance, 2017-2019
[2]Decuir et al., 2021 reported *Vibrio* infections detected by CIDT in Minnesota, 2016-2018
[3]Washington State reported *Vibrio* cases with CIDT and culture data, 2021-2022

Table 3. *Vibrio* species identified among vibriosis cases that were detected by CIDT and culture confirmed.

	Washington (2021-2022)*		COVIS (2017-2019)**	
Species	**Number**	**% of Total**	**Number**	**% of Total**
V. parahaemolyticus	76	92	497	62
V. cholerae (non-O1/non-O139)	3	4	150	19
V. fluvialis	2	2	70	9
V. mimicus	--	--	20	3
Grimontia hollisae	--	--	10	1
Other *Vibrio* species***	--	--	16	2
V. parahaemolyticus and another *Vibrio* sp.	2	2	--	--
Vibrio species not identified	--	--	39	5
Total	**83**	**100**	**802**	**100**

* Washington State data may not be representative of the rest of the country.
**Includes cases reported from Washington State. Cases in which *Vibrio* was not detected in a stool specimen are excluded; cases in which multiple *Vibrio* species were identified are also excluded.
***Includes *V. alginolyticus* (n=7), *V. vulnificus* (n=5), *V. furnisii* (n=3), and *V. ponticus* (n=1).

2. *Are there states or regions where culture confirmation is lacking?*

- Use of culture confirmation is influenced by state regulations to require clinical labs to submit clinical specimens with suspected *Vibrio* to a public health lab for additional testing, including reflex culture.
    - In December 2015, the Association of Public Health Laboratories published a nationwide assessment of state legal requirements for submission of isolates by clinical laboratories. Nearly all states require clinical laboratories to routinely submit isolates and other clinical materials to state public health laboratories. **However, rules vary by state, pathogen, and nature of investigation (such as outbreak or bioterrorism).**

- Data reported to COVIS during 2017-2019, for all non-cholera *Vibrio* infections:
  - Non-coastal states reported the highest total number *Vibrio* infections detected by CIDT (1,256), followed by states on the Atlantic Coast (1,034), Gulf Coast (838), and Pacific Coast (590).
  - On average, **non-coastal states reported a higher percentage of infections detected by CIDT that were culture-negative (53%)** compared to Atlantic, Gulf Coast, and Pacific states (range: 15%-39%).

3. *Now that CIDT has been in use for 5 years, updated data regarding false positives and false negatives:*

- Hitchcock et al., 2019 inferred false positives from reproducibility (testing the same sample multiple times).
  - Biofire FilmArray (tests from 2016-2018) had **20%** (2/10 samples) **false positives** for non-cholera *Vibrio*.
  - These 2 unconfirmed tests appeared "unlikely" to be vibriosis based on clinical presentation.
- Clarke et al., 2017
  - Identified 1 (**7%**) "**false negative**" for *V. parahaemolyticus* of 14 CIDT-positive tests.
  - The isolate cultured was *V. parahaemolyticus* but was identified as *V. cholerae* by CIDT.
- Kosai et al., 2021
  - Of 268 stool samples, **0 false positives** and **1 (0.4%) false negative** were reported for *Vibrio* group.
  - The false negative sample was positive on retest.
- In addition to the peer-reviewed literature referenced above, manufacturer information was considered. However, it was excluded due to lack of real-world data (validations were conducted only on artificially contaminated samples).
  - It is notable that there have been lot-specific recalls on the Verigene EP and Biofire FilmArrayBiofire FilmArray tests for potential false negative results (these recalls were not specific to *Vibrio* results, but potentially affected).
  - A BioMerieux representative acknowledged false positives for *Vibrio* testing with their CIDT assay (Biofire FilmArray). The company asks their customers to provide false positive samples for them to investigate the issue. In technical notes published by BioMerieux, several potential issues are described, including:
    - Cary Blair transport medium has the potential for *Vibrio* spp. contamination, as it is made with a seaweed component.
    - The *V. cholerae* assay (which is designed to detect the *toxR* gene) may react with other *Vibrio* spp. carrying homologs of *toxR*.

4. *Are different CIDT tests comparable?*

- Based on available clinical (real-world) data, different CIDT assays are not comparable.

- Based on available manufacturer and approval data, available CIDT assays perform similarly. However, as noted above, these data are generated using spiked samples and are not necessarily reflective of performance in the real world.
- Of note, some of the newer CIDT assays (Vikor, Health TrackRX) use the entire specimen in testing, so culture confirmation is not possible.

Table 4. Culture confirmation at Public Health Laboratory by type of CIDT assay.

Data Source	CIDT Assay	CIDT-positive/ culture-negative	CIDT-positive/ culture-confirmed	Percent Culture Confirmed*
Decuir[1] (2016-2018)	Biofire FilmArray	33	13	28.3
	Verigene EP	20	34	63.0
Washington[2] (2021-2022)	Biofire FilmArray	35	60	63.2
	Verigene EP	6	16	72.7
	BD Max Extended	4	7	63.6

[1]Decuir et al., 2021 reported *Vibrio* infections detected by CIDT in Minnesota, 2016-2018

[2]Washington State reported *Vibrio* cases with CIDT and culture data, 2021-2022

*Percent culture confirmed of those for which culture was attempted

5. *What is the frequency of co-detections and the frequency of accurate reporting of co-detection?*
   - Hitchcock et al., 2019 looked at co-detections, but none reported including *Vibrio*.
   - Kosai et al., 2021 reported co-detections using Verigene EP.
     - Of the 11 co-detections reported from 268 stool specimens, 3 (**1.1%**) included *Vibrio*.
     - *Vibrio* was co-detected with *Campylobacter*, *Y. enterocolitica*, and shiga toxin.
     - All 3 specimens were culture-confirmed for *Vibrio*.

Table 5. Identified co-detections at Washington State Public Health Laboratory (2021-2022).

	CIDT-positive Test by Type			Reflex Culture Result for *Vibrio* spp.			
Organisms Detected	**Total**	**Biofire**	**Verigene**	**Positive**	**Negative**	**Not submitted**	**Species**
Vibrio spp. and *Campylobacter* spp.	5	4	1	2 (Biofire)	1	2	*V. parahaemolyticus* and *V. fluvialis*
Vibrio spp. and *Salmonella* spp. and *Cryptosporidium*	1	1	--	--	1	--	--
Vibrio spp. and *Giardia* spp.	1	1	--	--	1	--	--
Vibrio spp. and *Cyclospora* spp.	1	1	--	--	--	1	--

6. *What are the predominant CIDT tests being conducted?*

- In the peer-reviewed literature, Biofire FilmArray is found the most frequently followed by Verigene EP. This is potentially biased by what is published, not necessarily what is actually used, and is likely to vary by region.
- Starting in 2012, FoodNet sites collected standardized data elements about practices from clinical laboratories, including questions about type of CIDT test used to initially detect *Vibrio*. FoodNet is a network of surveillance sites covering 15% of the US population.
    - During 2019-2020, among the yearly average of 131 clinical laboratories that indicated use of a PCR test to detect *Vibrio*, 73% used the Biofire FilmArray Gastrointestinal Panel, 21% used the Luminex Verigene Enteric Pathogens Panel, 5% used the BD Max Extended Enteric Bacterial Panel, and 1% used a laboratory developed test.

7. *What percentage are Vp specific (such as Diatherix)?*

- Vikor claims to differentiate *V. cholerae*, *V. parahaemolyticus*, and *V. vulnificus*.
- The major brands (Biofire FilmArray, Verigene EP) target *Vibrio* spp. but are unable to differentiate species.

IV. References

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V. Additional Reading

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