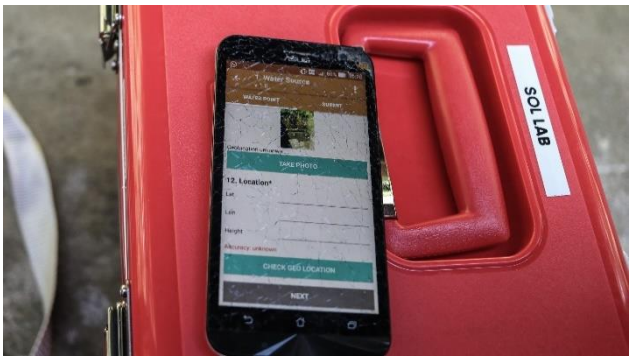

MEASURING E. COLI IN WATER: SMARTPHONE-BASED TESTING KIT PHASE ONE

EXPERIMENTATION RESULT

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October 2018

KOPERNIK

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Bali has faced serious problems related to deteriorating water quality over recent times, with Escherichia coli (E. coli) contamination frequently reported in natural spring waters[1,2]. Monitoring risk of E. coli in water is difficult as microbiological assessments for water rely on laboratory analysis (ex-situ) which is a complex and time-consuming process.

Akvo Caddisfly offers a practical means to measure E. coli combining the Aquagenx Compartment Bag Test (CBT) method and a data collection tool in the form of a smartphone app. We tested the E. coli level in 30 water samples, including refillable water from a depot, water filtered by a Nazava water filter and spring water using the Akvo test kit. We then compared Akvo's readings with laboratory results of drinking water samples from the same sources to evaluate:

- The accuracy of the Akvo Caddisfly in indicating E. coli levels
- The time required to perform and complete the tests
- The cost of performing the tests

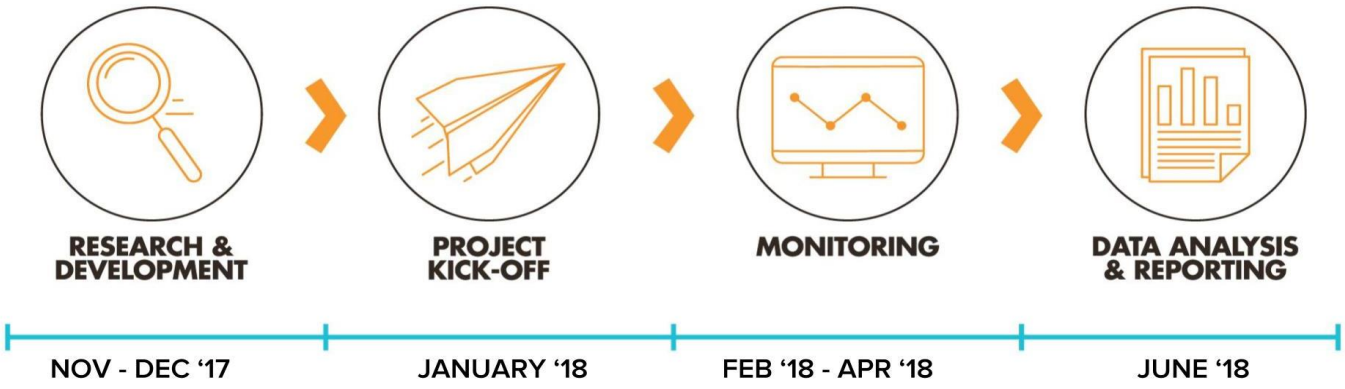
The result from this experiment showed that Akvo Caddisfly kit performs reasonably well in measuring the E. coli levels in drinking water. 80 percent of the time both Akvo's and Laboratory readings fell under the same WHO's risk profile and based on absence-presence cases, both readings were in agreement 83 percent of the time. We also found that the Akvo Caddisfly E. coli test kit gave results four times faster than the laboratory and further reduced the time required to perform a series of E. coli tests by 67 percent. In term of cost, the laboratory analysis is cheaper as compared to Akvo at least until 800 tests are conducted, provided that the laboratory is located within 25km. The app and data collection platform were appropriate for large datasets and comprehensive water quality studies

Special points of interest:

- 80-83 percent of the time Akvo Caddisfly test kit matches the laboratory test results for E. coli level in drinking water samples.
- Akvo Caddisfly E. coli test kit reduces 67% of the time required to perform 1000 tests.

Content

TIMELINE



[1] *E.coli found in Bali temple water has Gianyar regency focusing on water quality*. Coconuts Bali. 5 July 2017.
[2] *E coli level in Tukad unda is high, people use it for brushing teeth*. Tribun News. 1 december 2015.

CONTEXT

Access to clean water and sanitation is still a major challenge for people in both rural and urban areas in Indonesia. This has been confirmed by a recent water quality survey in Yogyakarta by BPS and BAPPENAS, showing that nearly 90 percent of households in the province are consuming water contaminated by E. coli bacteria[3]. While the majority of drinking water is boiled, the other option of purchasing ready-to-drink water from refillable water depots *(Depot air minum isi ulang)* such as shown in Figure 1, also does not guarantee E.coli-free water[4]. High contamination of E. coli in water is associated with an increased risk of diarrhea, which is primarily responsible for causing death in children under five years of age in Indonesia[5].

In Bali, high E. coli contamination has also been reported in spring water, highlighting the deteriorating quality of natural water supplies. Although springs are not the main source of drinking water, Balinese people have a strong tradition of drinking water directly from the spring because they believe it is holy water and contains healing and spiritual powers (Figure 2). Despite the possible dangers of drinking this water, the local environmental agency has not performed water quality testing for all water points in Bali due to a lack of staff and equipment to conduct appropriate water quality testing throughout the region[6].

Microbiological assessments for water quality is often a tedious and time-consuming process. The survey team has to take water samples from the source and at the point of consumption in the house and then deliver the samples to the laboratory within four to six hours. It is therefore costly to do regular E. coli test in areas that are far away from laboratory facilities. Laboratory analysis also typically requires 1-2 weeks to provide results.

In this experiment, Kopernik will test a simple E. coli test kit developed by Akvo for field water testing (in-situ measurements). By comparing Akvo's measurements against the laboratory results, we will understand whether Akvo's kit can be a substitute for laboratory tests to perform E. coli measurements in remote areas.

Figure 1. Typical refill water depot with ozone treatment in Bali

Figure 2. Sayan resident took spring water for drinking purpose

[3] *Yogyakarta survey reveals challenges and opportunities for ensuring access to clean water and sanitation*. UNICEF Indonesia. 5 January 2017

[4] *YLKI: Refill water depot is not all hygienic*. BBC news Indonesia. 15 Februari 2013.

[5] *Diarrhea Data and Health Information*. Indonesia's ministry of Health. Quarter II - 2011.

[6] *Hundreds of water points has not yet tested*. Tribun Bali. 17 April 2018.

HYPOTHESIS

We hypothesized that:

- the AKVO Caddisfly water testing system can give E. coli readings four times faster than the comparative laboratory analysis;
- the AKVO Caddisfly water testing system can indicate E. coli levels within a 95 percent accuracy rate in comparison to laboratory results.

Figure 3. Akvo Caddisfly E. coli test kit (cooler box is optional)

METHODOLOGY

Over two months, we tested 30 different water sources in Bali, of which 10 were samples of refillable water from depots, 10 were samples of water filtered by Nazava water filters, and 10 were samples of spring water. We measured E. coli in each sample using two methods concurrently. This resulted in a total of 60 measurements or 30 pairs of datasets.

Figure 4. Sample water containing E. coli changes its colour after 40 hours

Akvo Caddisfly E. coli kit (Figure 3) combines the Aquagenx compartment bag test (CBT) method with an app to measure E. coli levels on the spot, so that it is not necessary to bring the water sample to the laboratory. Akvo Caddisfly measures the E. coli level as the "Most Probable Number" (MPN) per 100ml. Akvo's app calculates the level and displays it as a value between 0 to 100, along with the risk profile according to WHO standards[7]. The app is also connected to a data collection platform and monitoring tool that captures timely and geo-referenced information, enabling data to be immediately digitized, accessed, and verified online. Figure 4 shows colored-change sample before it is captured by Caddisfly app. 5 below shows the Akvo kit and the steps performed to use it for E. coli testing.

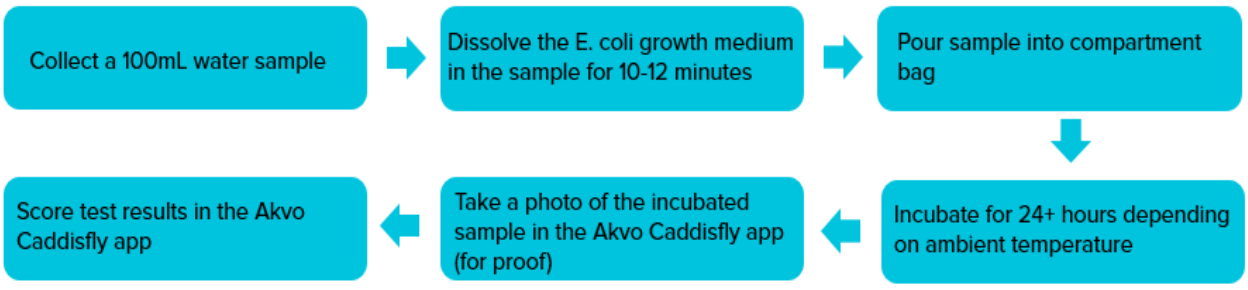


Figure 5. The E. coli test kit from Akvo is based on the compartment bag test AquaGenx

[7] Guidelines for drinking water quality Volume 3. WHO. 1997.

Laboratory tests were performed concurrently in a public laboratory facility *(UPT Balai Laboratorium Kesehatan)* in Denpasar. Their bacteriological test also measured the MPN of E. coli. Their results are stated according to the national standard for drinking water, which stipulates that no E. coli (zero MPN per 100ml) should be present in any drinking water[8]. For the laboratory tests we collected 150 ml of water from each point. We put this sample directly into the sterile bottles provided and stored them in a cooler box with ice. We drove the sample to the laboratory and submitted it on the same day for microbiological analysis. Figure 6 and 7 shows the water sample and results obtained from the laboratory.

Figure 6. Water sample prepared for bacteriological test in the Laboratory

FINDINGS

Measurements from Akvo Caddisfly were compared against the laboratory results, after applying WHO's E. coli risk guidelines for each water source tested. Figure 8 below illustrates this comparison in bar charts. This figure demonstrates that both Akvo and the laboratory tests showed 100 percent of low risk for E. coli in refillable water from depots. Both methods gave a slightly different result for Nazava filtered water, where Akvo indicated a 20 percent intermediate risk as compared to 10 percent indicated in the laboratory results. These risk incidents were identified on two Nazava filtration units where the filter had not been cleaned properly. The difference became more prominent in the spring water, where in most cases Akvo revealed a higher risk of E. coli than the laboratory results. The reason behind these discrepancies will be further discussed at the end of this section. Based on WHO standards, measurements from the two methods fell under the same risk category 80 percent of the time.

Figure 7. Public laboratory facility in Denpasar, Bali

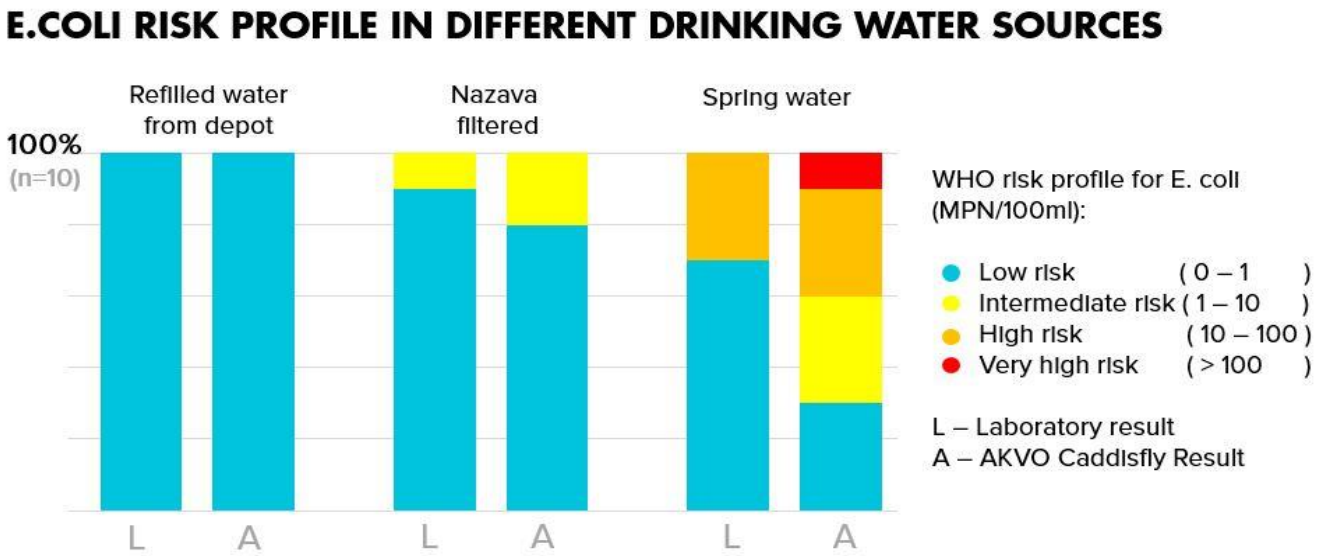


Figure 8. E. coli risk profile in different drinking water sources as measured with Akvo and Laboratory.

[8] Drinking water quality requirement. Ministry of health regulation no.492/MENKES/PER/IV/2010.

A comparison of the results can also be presented in terms of an absence or presence of E. coli in the water samples according to the national standard for the quality of drinking water. Absence is when E. coli measurement indicates 0 MPN level, and presence is when the E. coli measurement indicates more than 0 MPN level. The outcome from this categorization are four possible outcomes, summarized in figure 9 below. Based on an absence-presence classification, 83.3 percent of the time the Akvo results were in line with the laboratory results.

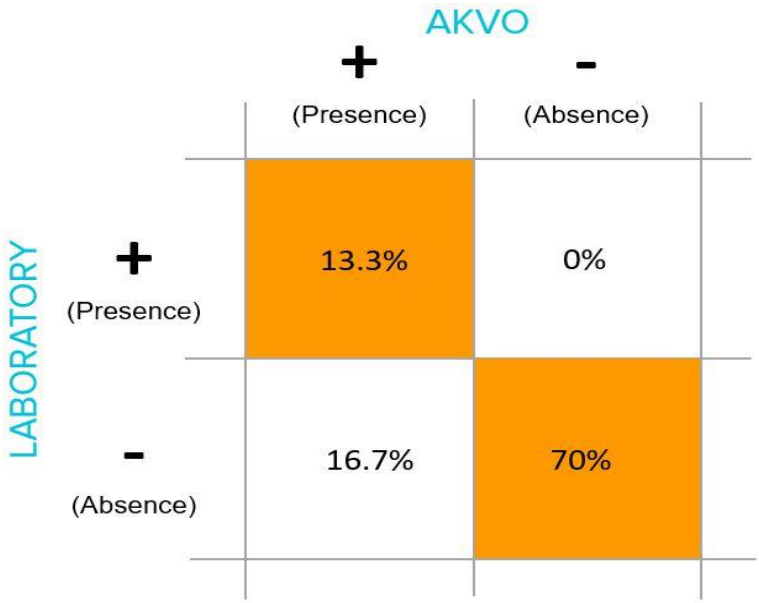


Figure 9. E. coli risk profile in different drinking water sources as measured with Akvo and Laboratory

Akvo displayed higher readings and was more conservative than the laboratory results across 6 samples, primarily in the spring water samples. This is likely due to the different way the two tests work with incubation and storing temperatures. The laboratory required us to store samples in a cooler box filled with ice, rendering an environment below 5 degrees celsius inside the box. While there is no ice storing requirement found in the similar test procedure (multiple tube fermentation technique) as documented by the US Environmental Protection Agency[9], this temperature likely limits the growth of E. coli and may prevent inactive E. coli from being detected during the actual test[10]. Contrary to the laboratory procedure, Akvo's sample was stored in a cooler box without ice as instructed. Akvo's incubation process was also performed in the same cooler box as the samples were stored, which was placed in a room with a temperature between 25 - 28 degrees celsius.

[9] Total Coliform: Multiple Tube Fermentation Technique. US Environmental Protection Agency. 2015
[10] The effect of temperature on the growth of the bacteria E. coli. Saint Martin University Biology Journal. May 2006

TIme and cost effectiveness

Performing a test with AKVO Caddisfly took an average of 15-20 minutes and required an incubation period of around 24 - 48 hours. We noticed with most of our water samples that were incubated at room temperature, that they did not start changing color until after 40-48 hours. Obtaining the result for E. Coli measurements with AKVO was thus four times faster than sending the samples to the laboratory to analyze, which requires 7-9 days of waiting time. Moreover in Bali, the laboratory only receives sample submissions until 12.30pm so the number of tests that can be performed and submitted for laboratory analysis within the same day is limited.

Based on our findings, we also calculated the time and the cost required to complete the E. coli tests projected up to 1000 tests. This is presented in figure 10. Performing and completing the E. coli test using Akvo reduced the working time by 56 percent when running 1,000 tests to 67 percent when running only 1 test. In terms of cost, the microbiological analysis in the government laboratory was approximately IDR115,000 (US$8) per test, while tests using the Aquagenx kit cost IDR125,000 (US$8.60) per test. The other cost elements are transport costs for the laboratory and shipping costs for the Aquagenx kit. Transport costs to the laboratory was calculated based on fuel costs covering a 50km return trip, which is around IDR60,000 (US$4.28). Shipping the kits from the United States cost between US$150 (for a minimum of 50 tests) to US$227 (for up to 1000 kits).

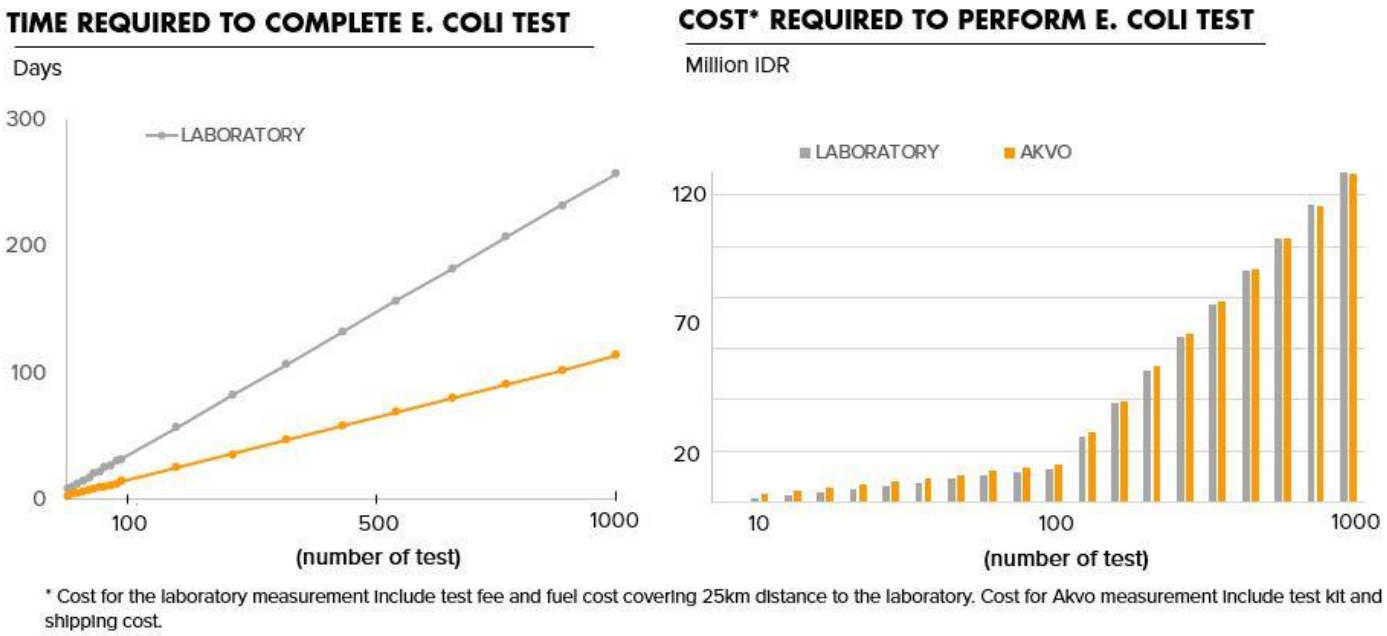


Figure 10. Time and cost effectiveness in performing E. coli test between Laboratory and Akvo measurement

Taking into account all of those elements, the cost projection suggested that the laboratory analysis is cheaper than Akvo's kit, at least until 800 tests are performed. However, it is important to note that the transport cost in this calculation is estimated based on a travel distance of 50km (round trip) to the laboratory to deliver the sample, therefore with a greater distance to the laboratory, it would increase the overall costs for laboratory analysis. In addition to the fact that the Akvo kit reduced the working days required to perform the E. coli test, potentially saving further costs such as staff salaries.

In terms of the software, the app and the online dashboard simplify the data collection process, minimizing human error and providing a way to verify the data at a later stage. The customizable data-capturing platform also allows users to create a comprehensive survey and to build a stronger correlation between a large number of data sets. This is particularly useful to answer questions related to water quality, such as, "How likely is it that E. coli contamination in drinking water will cause diarrhea?"

CONCLUSION

From the experiment's results, 80 percent of the time the Akvo measurements indicated values which fell under a similar risk profile as the laboratory measurements. In terms of the absence-presence classification, the two methods were in agreement 83 percent of the time. A similar evaluation study of E. coli test performance in Ghana defined the 80 - 89 percent range accuracy to be "reasonably well" or "relatively high"[11]. However these statistics did not support our second hypothesis, stating that Akvo would have a 95 percent accuracy rate.

The results from this experiment did prove that the Akvo Caddisfly system can give E. coli readings four times faster than the laboratory results. Furthermore, Akvo will reduce the time taken to perform E. coli tests by 56 - 67 percent. In term of cost, the laboratory analysis is cheaper as compare to Akvo at least until 800 tests, given the laboratory is located within 25 km distance. Overall, the Akvo test kit using the AquaGenx CBT method did prove its worth in being a quick and practical way of detecting E. coli in water.

TESTIMONIAL:

"The smartphone-based E. Coli testing pilot with Kopernik was an exciting experience. The talented team was eager to innovate and open to suggestions. Akvo's reflection for the team would be on improving the understanding of WASH in general and water quality in particular. We look forward to future collaborations with sustainable scaling roadmaps to create bigger impacts. "

Joy Ghosh (Programme and Technical Advisor, Akvo.org)

RECOMMENDATION

Based on the results from this experiment, Kopernik recommends that:

- In collaboration with other organizations, Kopernik searches for and evaluates other rapid E. coli testing methods which could give a result within a maximum of three hours, as specified in the recent target product profile of UNICEF.
- The government, in collaboration with other organizations, looks into the possibility of investing in data collection system like Akvo's for monitoring water quality and other related surveys.
- In collaboration with other organizations, Kopernik promotes the use of an online data collection platform for study and research which requires a large datasets.

[11] Evaluation of microbiological water quality tests test for humanitarian and development setting. Massachusetts Institute of Technology. 2015.