

Short Report

Prepared by Dr. rer. nat. Katrin Luden (scientific consultant)

Introduction

This report summarizes the topics and discussions of the evaluation workshop for the 3rd microbiological PT round held in Winhoek, Namibia. It is meant to inform all interested laboratories and help with corrective actions. A more detailed report will be published on <http://www.SADCMET.org>.

Workshop

All participants and the German experts were welcomed by Namwater representative Dr. Shivute (CEO) the SADCMET regional coordinator Mr. Masuku the PTB representative Ms Wunderlich and SADC WaterLab Association Chair Mrs. Mwambo. Afterwards the workshop split into two groups chemistry and microbiology to evaluate the respective PT schemes. 16 participants representing laboratories from 14 different countries participated in this evaluation workshop of the 3rd Microbiology Proficiency Testing round.

Report of the PT provider UNBS

Mrs Jacqueline Kwesiga from Uganda Bureau of Standards (UNBS) described trial runs and several packaging simulations that were conducted in preparation for the 3rd PT. DHL was used as a courier as previously. With 33 laboratories participating in the 2010 PT a major increase in participants was achieved although some difficulties with email-communication were encountered. These were mainly due to unsatisfactory performance of hardware and server-provider. This has been addressed by UNBS by switching to a different provider. Samples were dispatched on the announced date and mostly received within a few workdays. Unfortunately some of the samples were frozen when arriving at the participating laboratories. This might be due to the very cold temperatures the ice bricks had been stored at before use in packaging. The packaging will have to be further improved. The only packages held up in customs were the ones to Mauritius. 9 out of 33 samples reached their destination within the optimum 2 day period and 12 samples showed temperatures in the desired range at reception.

Evaluation of the PT

General aspects: Analysis of the reported data revealed that only 17 of 33 laboratories (52 %) managed to start analysis of the PT samples at the day of reception. In one case analysis was even delayed for 6 days. The PT samples contain live organisms and therefore have limited stability. Hence it is crucial to start analysis right away. As the PT samples were announced well in advance and there is only once a year a chance to participate in this performance testing the lack of suitable arrangements for analysis is not easy to comprehend.

Unfortunately the participants' results did not show a consensus value that could be used as assigned value to judge the reported results by statistical means. Next to

reaching a number of approximately 30 participants in order to have a sound basis for statistical evaluation it is necessary to come up with a trustworthy assigned value.

It was decided that the laboratory of the scientific consultant NLGA (Germany) and at least one or two more laboratories should receive samples and be dealt with as expert laboratories. These data will be compared to the UNBS Quality control data. The consensus value of these expert laboratories will then be used for statistical evaluation of performance according to ISO 13528.

Methods: All participants had been asked to give detailed information on the methods used for analysis of the PT samples with their results. Sometimes some ISO standard was cited but the method described did not match the ISO standard.

If changes are made in medium used e.g. due to availability problems or other things changed the method stated can only be "modeled after ISO..." or a similar description. The method has then to be validated at laboratory level.

Participants compared within small working groups the methods for E. coli/Coliforms used in different laboratories for PT analysis. Microbiological analysis of water samples is greatly operationally defined. E.g. methods for detection and enumeration of total coliforms use anything from lactose fermentation (gas and acid production from lactose) to enzyme activity (β -galactosidase) to describe this group. It is not surprising that this leads to a very different set of species detected by various methods. Use of many different methods as used by the participants of the PT complicates and in the worst case prevents comparison of results within the PT scheme but also comparison of results from regular analysis in routine laboratory work. This problem was discussed in detail and a working group installed to come up with a recommendation on standardized methods best suitable for analysis of potable water in the SADC and EAC region. The recommendation is published on the SADCMET website.

Training on method validation

Method validation was discussed using **ISO/TR 13843:2000 Water quality - Guidance on validation of microbiological methods**. The necessity to clearly define the purpose/scope of the method to be validated was stressed. Regulatory limits have to be taken into account as well as the matrix the method is supposed to be used with. For example a method for drinking water analysis of E. coli with a limit of 0 in 100 ml is not fit for purpose as long as a sample volume of less than 100 ml is used. After defining the purpose very clearly a basic description of target organisms and the method can be written. Experimental data has to be gathered from pure culture experiments as well as from natural samples. Numbers for sensitivity, selectivity, specificity, efficacy, rate of false positives and false negatives have to be calculated. Uncertainty of counts and the robustness of the method at the limits have to be checked. For quantitative methods it is also important to know the linearity range. To save a lot of work compared to a full scale method validation it is recommended to use an established standard (e.g. ISO) because this only has to be verified which adds to the benefit of comparability.

A method described in an established standard only has to be verified to prove that it works in the hand of the user according to its characteristic values (establishes that the method performs to its specifications).

Training on measurement uncertainty

The topic of measurement uncertainty was shortly addressed during the workshop. Unfortunately there is no gold standard for calculating measurement uncertainty for microbiological methods. A few approaches were shortly introduced: the Top-down approach (GUM/EURACHEM/CITAC) and the Bottom-up approach (VAM/NORDTEST) used in chemistry. Moreover there is the ISO CD 29201 as a standard under development that deals with the question of MU in microbiology. This ISO describes two ways the step-by step (GUM) and the global approach.

The workshop ended with a visit at Namwater laboratories.

Conclusions

- There is a strong need to improve the methods used for analysis in the PT.
- Evaluation of the PT gave valuable leads for improvement to all laboratories even though a statistical evaluation of the PT was not possible.
- Evaluation for the next PT will have to be based on an assigned value. Therefore NLGA and other laboratories in Germany will be approached to serve as expert laboratories. At least one laboratory from the southern African region should be recruited as expert laboratory as well.
- Participants of the workshop felt it important to recommend and use comparable methods for the analysis of E. coli and Total coliforms in potable water that are fit for purpose.
- The PT provider UNBS did a good job in sample preparation but still there is need for improvement on the logistics.
- After detailed discussions of the total plate count methods in the workshop 2009 this workshop focused on the methods of E. coli/Coliform detection.
- Quite a few participants need to adjust their methods to the standards cited or validate their used method which would be much more laborious.
- The workshop was filled with lively discussions and left enough room for networking and sharing experiences.