

## Ship-borne measurements of microbial enzymatic activity: A rapid biochemical indicator for microbial water quality monitoring

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Contamination of aquatic ecosystems by human and animal wastes is a global concern for water quality. Disclosing fate and transport processes of fecal indicator organism (FIO) in large water bodies is a big challenge due to material intensive and time consuming methods used in microbiological water quality monitoring. In respect of utilization of large surface water resources there is a dearth of rapid microbiological methods that allow a near-real time health related water quality monitoring to be implemented into early warning systems. The detection of enzymatic activities has been proposed as a rapid surrogate for microbiological pollution monitoring of water and water resources (Cabral, 2010; Farnleitner et al., 2001, 2002). Methods such as the beta-D-Glucuronidase assay (GLUC), targeting FIO such as E. coli, were established. New automated enzymatic assays have been implemented during the last years into on-site monitoring stations, ranging from ground- to surface waters (Ryzinska-Paier et al., 2014; Stadler et al., 2017, 2016). While these automated enzymatic methods cannot completely replace assays for culture-based FIO enumeration, they yielded significant information on pollution events and temporal dynamics on a catchment specific basis, but were restricted to stationary measurements.

For the first time we conducted ship-borne and automated measurements of enzymatic GLUC activity on large fresh water bodies, including the Columbia River, the Mississippi River and Lake Mendota. Not only are automated enzymatic assays technically feasible from a mobile vessel, but also can be used to localize point sources of potential microbial fecal contamination, such as tributaries or storm drainages. Spatial and temporal patterns of enzymatic activity were disclosed and the habitat specific correlation with microbiological standard assays for FIO determined due to reference samples. The integration of rapid and automated enzymatic assays into well-established systems for ship-borne measurements of physico-chemical parameters, such as the FLAMe (Crawford et al., 2015), paves new ground for data interpretation and process understanding.

Cabral, J.P.S., 2010. Water Microbiology. Bacterial Pathogens and Water. Int. J. Environ. Res. Public. Health 7, 3657–3703.

doi:10.3390/ijerph7103657

Crawford, J.T., Loken, L.C., Casson, N.J., Smith, C., Stone, A.G., Winslow, L.A., 2015. High-speed limnology: using advanced sensors to investigate spatial variability in biogeochemistry and hydrology. Environ. Sci. Technol. 49, 442–450. doi:10.1021/es504773x

Farnleitner, A. h., Hocke, L., Beiwl, C., Kavka, G. c., Zechmeister, T., Kirschner, A. k. t., Mach, R. l., 2001. Rapid enzymatic detection of Escherichia coli contamination in polluted river water. Lett. Appl. Microbiol. 33, 246–250. doi:10.1046/j.1472-765x.2001.00990.x

Farnleitner, A.H., Hocke, L., Beiwl, C., Kavka, G.G., Mach, R.L., 2002. Hydrolysis of 4-methylumbelliferyl- $\beta$ -

d-glucuronide in differing sample fractions of river waters and its implication for the detection of fecal pollution. Water Res. 36, 975–981. doi:10.1016/S0043-1354(01)00288-3

Ryzinska-Paier, G., Lendenfeld, T., Correa, K., Stadler, P., Blaschke, A.P., Mach, R.L., Stadler, H., Kirschner, A.K.T., Farnleitner, A.H., 2014. A sensitive and robust method for automated on-line monitoring of enzymatic activities in water and water resources. Water Sci. Technol. J. Int. Assoc. Water Pollut. Res. 69, 1349–1358. doi:10.2166/wst.2014.032

Stadler, P., Blöschl, G., Vogl, W., Koschelnik, J., Epp, M., Lackner, M., Oismüller, M., Kumpan, M., Nemeth, L., Strauss, P., Sommer, R., Ryzinska-Paier, G., Farnleitner, A.H., Zessner, M., 2016. Real-time monitoring of beta-d-glucuronidase activity in sediment laden streams: A comparison of prototypes. Water Res. 101, 252–261. doi:10.1016/j.watres.2016.05.072

Stadler, P., Farnleitner, A.H., Zessner, M., 2017. Development and evaluation of a self-cleaning custom-built auto sampler controlled by a low-cost RaspberryPi microcomputer for online enzymatic activity measurements. Talanta 162, 390–397. doi:10.1016/j.talanta.2016.10.031