HOW MEANINGFUL IS A QUANT RESULT?

Kate Stevenson, Institute of Environmental Science and Research Ltd (ESR)

When a laboratory considers an upgrade of a consumable or chemistry it can be difficult to select one specific kit for use. Suitability of use, published research, cost and familiarisation with supplier products are just some considerations for selecting a kit for validation.
When an upgrade of quantitation kits in the Forensic Biology laboratory at the Institute of Environmental science and Research Ltd was being considered, we undertook a lab evaluation of three different kits from different suppliers to make an informed decision about the best kit for use in our laboratory. This "battle of quant" kits included a comparison of performance of the Quantifiler™ Trio kit (ThermoFisher), the Powerquant® kit (Promega) and the Investigator Quantiplex HYres kit (Qiagen).

Our study included standard validation requirements for sensitivity, concordance and accuracy yet the validation was also extended to include comparing the effectiveness of the inhibition markers (both IPC and passive reference dye) and/or degradation indexes incorporated into the kits, the stability of the standards and how the targets within each kit performed to wide range of mixture ratios.

Even after a kit was selected for use in the laboratory, further experimental work was required in response to variable results being generated for low template DNA samples. This could be partly attributed to the lower limit of detection using the kit and new HID software on the 7500 instruments. The previous limit for detection was 0.001ng/µL for the Quantifiler™ human kit and the Quantifiler™ Y kit whilst for the new kits the limit of detection was 0.00001ng/µL (0.01pg/µL). The effect of preparation method (automated versus manual processing), extraction methods but more importantly time delay between extraction and quantitation were also deemed to be contributing factors. This resulted in downstream procedural changes both within the laboratory and reporting guidelines.

The data specific to the comparison of the kits as well as the contributing factors to variance in results obtained from the kit implemented are presented.