A COST-EFFECTIVE WORKFLOW FOR MASSIVELY PARALLEL SEQUENCING OF DRUG METABOLIZING ENZYMES

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Mutations in drug metabolizing enzymes can lead to varying responses to similar doses of pharmaceutical compounds. These mutations can have serious implications ranging from requiring an adjustment of drug regimens to adverse reactions that otherwise would be unanticipated by health care practitioners. In some instances, these adverse reactions can lead to temporary disorientation or even death. In forensic settings, this could play a role in molecular autopsies.

In this study a robust method was developed for the use of massively parallel sequencing to identify polymorphisms in drug metabolizing enzymes. This method successfully sequenced the *CES1* gene of 172 juvenile saliva samples. Automated sample extraction in conjunction with long amplicon polymerase chain reaction (LAPCR) was used for target enrichment. The use of LAPCR for target enrichment is a cost-effective means of avoiding the use of costly manufacturer made primer panels often used for similar MPS sequencing applications. Library preparation for MPS was accomplished with Illumina Nextera XT™ library preparation kits. Sequencing was completed on an Illumina MiSeq FGx™ in research use only (RUO) mode. Data processing was completed using the standard installed MiSeq Control Software, Real Time Analysis software., and MiSeq Reporter. The generated variant call files were then examined, combined and filtered through free to use software including Integrated Genomics Viewer and VCFtools. Several single nucleotide and insertion/deletion polymorphisms were sequenced in the sample population.