DIRECT AMPLIFICATION OF BLOOD ON CRIME SCENE SUBSTRATES USING THE QIAGEN INVESTIGATOR® 24PLEX GO!

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Autosomal Short Tandem Repeat (STR) Analysis is a molecular technique performed widely in forensic laboratories. In violent personal crime cases, blood is routinely encountered on various substrates. Direct amplification of these bloodstains without time consuming, labor intensive extraction and quantitation steps have proven to be useful in forensic science laboratories. The QIAGEN 6-dye amplification system, Investigator® 24plex GO! allows the identification of 22 polymorphic STR loci and includes two innovative internal PCR controls (Quality Sensors). The quality sensors, QS1 and QS2, provide information about whether there is degradation and/or inhibition. These also indicate the absence of DNA or a failed PCR reaction. The current research utilizes this kit to directly amplify minute amounts of blood deposited on various simulated crime scene substrates while the substrates remained in the reaction during amplification. Substrates chosen for deposition of blood included various types of fabric, denim jeans, leather, and also woodchip, straw, grass and other objects.

Blood from four deceased donors were diluted in a 1:1 ratio and 0.2 µL of each diluted sample was deposited on 1.2 mm punch or cutting of each substrate. After bloodstains were dried overnight, they were transferred to individual tubes. In the next step, 5 µL of Investigator GO! Lysis Buffer was added to each punched substrate and left at room temperature for 20 minutes with occasional mixing. Then, 20 µL reaction mixture was added to each tube and amplification was performed following recommended protocol. During the amplification step, all substrates remained in the reaction mixture. Amplified products were injected into the 3130xl capillary electrophoresis (CE) system. GeneMarker® HID analysis software v 2.9 from SoftGenetics® was used for fragment analysis.

All bloodstains created from the four blood samples were amplified successfully even when the substrates remained in the reaction mixture during amplification steps. Consistent and concordant profiles were obtained from all of the bloodstained substrates. The S peak on QS2 locus occasionally dropped out indicating inhibition in the sample, even when a complete profile was obtained.

The results of this research indicate that the Investigator® 24plex GO! is a valuable tool which can be easily incorporated in the analysis of blood in forensic laboratories. Since there is no extraction and quantitation involved in the procedure described above, the results can be obtained within a very short period of time.