POWERPLEX® 16 THERMAL CYCLING PROTOCOL FOR THE HYBAID MULTIBLOCK SYSTEM

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DNA typing is a useful and well-established tool in forensic casework but there is still a lot of space for improvements and technological innovations of the process. Under the heading "Technological Innovations for Forensic DNA Testing" we present 3 protocols, which can help the forensic laboratories to set up a better standard of laboratory practice. The protocols described below make use of the instrumentation of the Thermo Electron Bioscience Division – Hybaid MultiBlock System, KingFisher™ magnetic particle processor and Fluoroscan Ascent FL luminometer. We want to demonstrate the usefulness of the presented protocols for the crucial steps of forensic DNA testing – extraction, quantitation and PCR.

MultiBlock System (MBS) is based on the concept of a central computer that executes programs and monitors the performance of a number of satellite blocks. Accurate sample temperature control is achieved by Active Tube Control software. This type of control uses the remote thermistor probe mounted in an appropriate tube. The tube termistor probe monitors the temperature within the dummy sample tube and this information is fed back to precisely control the block temperature to achieve the optimum cycling profile. In order to transfer the PCR protocol for the PowerPlex® 16 System (Promega Corporation, Madison, WI) from the Perkin-Elmer GeneAmp PCR System 9700 Thermal Cycler we used the thermistor lead extension from a block control machine (P-E 9700) to tube control. During the transfer of the protocol, we used the control tube of the MBS as a temperature probe in the block control machine. The final protocol is shown in the table below.

PP16_ MBS	Stage 1	Stage 2	Stage 3			Stage 4			Stage 5	Stage 6
	1 cycle	1 cycle	10 cycles			22 cycles			1 cycle	1 cycle
Temp °C	95,0	96,0	94,0	60,0	70,0	90,0	60,0	70,0	60,0	4,0
Time	00:11:00	00:01:00	00:00:30	00:00:30	00:00:45	00:00:30	00:00:30	00:00:45	00:30:00	HOLD
Gradient	0	0	0	0	0	0	0	0	0	
Ramp °C/sec	MAX	MAX	1,32	0,60	0,25	1,50	0,60	0,25	MAX	
Temp Inc	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	
Time Inc	0	0	0	0	0	0	0	0	0	

Conclusions:
The protocol PP16-MBS is optimized for the use of PowerPlex®16 with the *AmpliTaq* Gold® DNA polymerase accordingly to the manufacturer recommendations.

The protocol PP16-MBS gives on the Hybaid MultiBlock System exactly the same DNA profile, including the peak height, sister peak imbalance and background noise if compared to the data obtained with the block control machine Perkin-Elmer GeneAmp PCR System 9700 Thermal Cycler.