

## Human Identification Kit, for multiplex amplification of X - Chromosome specific STR loci.









This Kit contains 15 markers which are X- chromosome specific, an autosomal STR marker (D21S11) to rule out sample mix-up and AMXY as a sex determination marker. The GT XDetector Kit is intended for molecular biology applications in forensic, human identification, and kinship issues.

## Storage conditions

- Prevent exposure of primer mix to direct light. This may have an impact on the intensity of the fluorescent dye.
- Store all components at -20°C.
- Avoid repeated freezing-thawing cycles to maintain the good quality of the kit. We recommend to aliquot the components if necessary.

## GT XDetector components

**Table 1: Provided with the Kit are Box A and Box B. They should be kept separately.**

BOX-A			BOX-B		
	Tube Label	Tube cap colour		Tube Label	Tube cap colour
1	PCR Mix		1	GT500 Size Standard	
2	Primer Mix		2	GT XDetector AL	
3	GT HSTaq		3	GTM5 v2 (Optional)	
4	GT QCDF150 (Control DNA-50ng/μl)				
5	GT QCW (H2O)				

## Instructions

1. Bring all components to room temperature.
2. Vortex Primer Mix and PCR Mix and spin down briefly to remove all residues from the lid. Gently mix the enzyme by inverting or pipetting.
3. Prepare a Master Mix for your reaction according to the following recipe. Every preparation can be done at room temperature.

**Table 2: PCR reaction set-up**

Component	Volume for 1 reaction[μl]
GT QCW (H <sub>2</sub> O)	10
PCR Mix	7
Primer Mix	1
GT HSTaq	1

4. Vortex Master Mix briefly.
5. Transfer 19μl of Master Mix to each 0.2ml PCR tube for each sample you want to analyze.
6. Add 1μl of DNA template (1-5 ng per reaction) to each PCR tube.
7. Vortex and spin down each PCR tube. Make sure that no drops are left at the tube wall or lid.
8. Place tubes into thermal cycler.

9. Please use the following PCR program for the amplification of all markers.
10. Store the PCR products at 2-6°C until analysis with Genetic Analyzer.

**Table 3: PCR program**

Initial step	Cycling			Final Extension	Storing in Cycler
	Denaturation	Annealing	Extension		
95 °C	95 °C	62 °C	70 °C	70 °C	4 °C
20 min	1 min	90 sec	2 min	17-20 min	∞
27-30 Cycles					

**Note**

- We recommend storing PCR product at 2-6°C in a dark place (fluorescent dyes!)
- The quality of the results will reduce with increased time gap between PCR amplification and capillary electrophoresis.
- A quality control (provided in the kit) and a negative control should be run in each Multiplex PCR to verify successful amplification of each marker.
- Varying quantity of DNA template may require different numbers of cycles in PCR program. Please see “GT XDetector User Manual” for further information.

**WARNING**

After PCR is complete, tubes should never be opened in the PCR setup area or beside kit components. Risk of contamination!

**How to analyze data from GT XDetector Kit**

- GT XDetector Kit is optimized for usage on ABI PRISM Genetic Analyzer like ABI 3130/xl or ABI3500/xL. Make sure your ABI Data Collection Software supports 5-dye fragment analysis.
- Calibrate the instrument spectrally with the GTM5 v2 Matrix Standard Spectral Calibration Kit. For further information write us ([support@genetek.de](mailto:support@genetek.de)).
- We recommend verifying a successful Multiplex PCR by gel electrophoresis before analyzing it on Genetic Analyzer.
- Prepare PCR products for capillary electrophoresis according the ABI protocol. Analyze the samples using the GT XDetector GeneMapper Panel provided on our website.

**Note**

- For further information regarding GT XDetector Kit please see “GT XDetector User Manual”. It includes recommendations for different DNA amounts per reaction, table containing the names and sizes of all amplified markers as well as troubleshooting.
- To simplify the analysis of your samples, we provide a panel.
- Please find all documents regarding GT XDetector Kit on our website: [www.genetek-biopharma.com](http://www.genetek-biopharma.com).
- It may arise that alleles fall outside their size range and overlap with the size range of another locus. This appears at low frequencies in populations.
- For any further clarification, please contact our technical service via email: ([support@genetek.de](mailto:support@genetek.de)).