



MINISTÈRE DE L'INTÉRIEUR,
DE L'OUTRE-MER
ET DES COLLECTIVITÉS TERRITORIALES

NATIONAL FORENSIC SCIENCE INSTITUTE

LYON FORENSIC SCIENCE LABORATORY

AUTOMATED GENOTYPING UNIT

The Automated Genotyping Unit of the Lyon Forensic Science Laboratory enables the analysis of 10 000 reference samples per month. From the samples taken by police officers in France and French overseas territories, this unit supplies the national DNA database (FNAEG) with the analysed profiles. It meets the highest requirement standards in terms of reliability.

Each profile being obtained from two samples, after various technical controls; it means that 300 000 samples are processed each year, in order to determine 10 000 profiles per month.

Supplementary DNA amplification and analytical devices have been added to increase the analytical capacity up to 20 000 profiles per month for 2009, without changing the premises.

Concerning organization, two automated lines are set up in two clean rooms, linked together by an airlock, to enable the transfer of microplates. In these rooms, elements are rigorously controlled : the rate of particles in suspension, air pressure, temperature and hygrometry.

It is the first time in Europe that forensic genetic analyses have been made both in clean rooms and with such an automated process.

This unit has been operational since September 2006 and it represented a global investment of 2.6 million euros.

Technical description of the process

Sampling on individuals

On a national level, Police and Gendarmerie officers collect buccal swabs from individuals on a specific FTA card (Fast Technology for Analysis). Due to the chemical formula on FTA cards, cells are lysed on contact with FTAs and DNA is bound to the matrix (cellulose fibres).

DNA samples can be stored or transported at room temperature, and the captured DNA is protected from all form of damage. Bar-codes ensure traceability of FTA cards and their corresponding forms, both tightly linked to the judicial procedure.



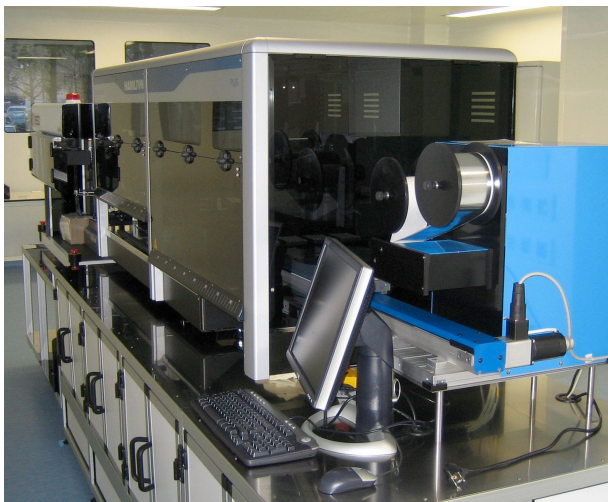
FTA card with the buccal swabs sampling kit

Preparation of samples for DNA amplification

In the first clean room, 1.2 mm discs are removed from FTA papers with an automated puncher coupled up to a robot.

Those discs are automatically placed in 96 wells microplates and will undergo a washing process.

A digital camera fixed on robots controls operations, so that they go smoothly.



One of the automated lines designed for the preparation of samples for amplification

Then, different reagents are added to enable the visibility of the peaks along with the amplification of the 16 non-coding DNA zones, which bear no genetic information.

When the process is over, microplates are automatically sealed and transferred to the adjacent clean room, through an airlock.

The pipetting system of each robot enables fluid transfer operations of around one microliter (one millionth of a liter), with a 5% accuracy.

DNA amplification and separation of the obtained fragments

In this second clean room takes place DNA amplification, which is called PCR (Polymerase Chain reaction). At this stage, the number of newly-synthesized molecules being really high, the room is low-pressured compared to the outside air. Indeed, a DNA contamination of the premises could disturb future analyses.

The staff working in this room is thus fully

equipped and gets into it through a low-pressure airlock.

Amplification is made with automated thermocyclers. During the process, the double-stranded DNA molecule is opened by rising temperature which enables the access to amplification enzymes. The double helix is then closed up by lowering temperature. The closing/opening cycles of the double DNA helix are repeated 27 times.

After amplification, samples are divided up on 384 wells microplates, and are mixed with a denaturing agent before being analysed on electrophoresis devices.

Electrophoresis consists in separating small DNA fragments in an electric field, which have been synthesized during the amplification stage, from the 16 zones studied.



One of the capillary electrophoresis device equipped for the analysis of DNA fragments

Determination of genetic profiles

Depending on people and according to the zones studied, DNA fragments are obtained. They are more or less long and correspond to a varied number of repetitions of the same pattern.

Many computers are placed in the same room and are linked to the electrophoresis devices through the computing network of the laboratory. Thus, analytical data is automatically retrieved.

From these data, which are analysed by different softwares, engineers and technicians establish the final genetic profile. This one is designed as an alphanumeric chart which can be sent to the national genetic fingerprints database (FNAEG).

Quality standards accreditation

The Lyon Automated Genotyping Unit was designed in order to obtain the ISO 17025/2005 international standard accreditation.

Therefore, all the standard requirements were complied with, from the very design of the building, to the setting up of the automated lines.

In order to ensure the traceability and reliability of analyses, two samples of each FTA are made and analysed on two separated

and identical automated lines. Results are validated and sent to the automated national genetic fingerprints database (FNAEG) only after perfect matching of the two results obtained from the 2 distinct lines.

Particular care was brought to suppressing static electricity, which can be a very disturbing parameter for such analyses. To this end, antistatic materials were chosen for the floor coverings of the clean rooms. Besides, hygrometry and temperature are carefully regulated.

Laboratoire de Police Scientifique de Lyon
31 Avenue Franklin Roosevelt
69134 Ecully CEDEX- FRANCE
Tel : 04.72.86.89.70
Fax : 04.72.86.85.85