1. Welcome

Angel Carracedo welcomed members to Santiago de Compostela. Mr Profilis, EU, was unable to attend the meeting and there was no representation from Athens, Münster, and Paris/Lyon/Toulouse.

2. Update on STADNAP exercises

2.1 mtDNA from head hair samples 1  Gillian Tully

A manuscript for this exercise is now in preparation. Members were requested to provide Gillian Tully with the methods used and names of authors as soon as possible.

2.2 mtDNA from head hair samples 2  Gillian Tully

To date, results have only been received from one laboratory that reported heteroplasmacy at one base. Some laboratories have also reported a difficulty with obtaining PCR products.

Final results should be sent to Gillian Tully by the end of September 2000

2.3 Response to the publication of Grzybowski  Gillian Tully

A response was prepared and sent to Electrophoresis that received a subsequent reply from Grzybowski. Gillian will address the issue with a further communication and it is intended that all of the correspondence will be published in the next edition of Electrophoresis.

2.4 Manuscript on the presentation of mtDNA evidence  Gillian Tully

It was suggested that the manuscript should either be sent to Bruce Weir for comments prior to submission or to request that the journal use him as a referee. There was general agreement that the manuscript should be submitted to Forensic Science International.
2.5 Y chromosome pentaplex  
Angel Carracedo

A manuscript has been submitted to Forensic Science International. The names of all authors and the name of the EDNAP group are listed on the first page, while the names of the laboratories, addresses, etc. are presented in an annex. This was considered to be a good compromise following previous discussions.

2.6 Analysis of mixtures  
Peter Gill

So far, there have been insufficient returns to make an effective analysis possible. Members were requested to send their results to Peter Gill by the end of September 2000. It seems to be enough to report results on only one sample with results that seem satisfactory to the laboratory.

2.7 Degraded DNA  
Klaus Bender/Peter Schneider

The degraded material was prepared by subjection to proteinase K and phenol/chloroform treatment followed by RNAse digestion, sonication and DNase I digestion. The results, following various sonication times and different levels of enzyme digestion, were presented.

The final preparation, which demonstrated a general loss of larger fragments, was used for the exercise. As it was agreed that the exercise would include participants outside of EDNAP, samples were sent to a total of 50 laboratories. So far, 40 have registered and by 27 August, results had been received from 20 laboratories. It is intended that a presentation of the preliminary results will be made to the IALM meeting on 7 September 2000.

There was a considerable spread of different multiplexes used by the various participating laboratories and, to date, the analysis of the returns indicates that there are considerable differences between the results obtained. In general, the larger alleles could not be detected but, as results were determined solely by signal strength, it does not follow that the correct results were achieved. Common problems encountered included ‘pull-up’ peaks, ‘stutters’ due to over amplification, loss of information above 150/250 bp and allelic dropout that could be either the larger or smaller allele.

Hermann Schmitter showed some results on 'short STRs' that had been obtained from primers binding very closely to the target alleles. While these singleplex reactions showed greater sensitivity some dropouts were also experienced.

Peter Gill requested that duplicate sample analyses be submitted in order to make an assessment of the level of consensus results. It was appreciated that there would be a necessity for the production of strict guidelines for reporting results.

The intention is to produce 2 publications:

1. Preparation of the degraded material
2. Results of the exercise.

The deadline for the return of results to Peter Schneider is 31 October 2000 and the original coloured printouts should be sent.
It is the intention to try to produce larger batches of degraded material for future experiments. Peter Schneider and Klaus Bender will explore the possibility of producing degraded DNA:

1. a new, larger batch of the previously used cell line, and
2. from 1 - 2 new cell lines.

In the future, the preparation could be passed to an organisation that routinely produces standard reference material. It seems as if also commercial companies could use this as reference material. A strategy will be decided at the next EDNAP meeting.

2.8 Short STRs on telogen hairs

Hermann Schmitter

So far, only 5 sets of results have been received.

While more work was needed on the amplification procedure, some success has been achieved with 2 singleplex reactions for THO1 and TPOX loci. Work is also continuing with a multiplex system using 9 loci and a solid phase amplification approach. Hermann Schmitter will produce a technical note regarding the use of primers that anneal close to the target loci.

Further hair samples can be obtained from Hermann Schmitter. Primers must be produced or purchased by the laboratories. Ladders can be produced from commercially available ladders.

Deadline for submission of results on the ongoing exercise is end of December 2000.

2.9 EMPOP

Walther Parson

The mtDNA (EMPOP) database is now on-line and consists of:

- web page: www.empop.org
- database of mtDNA sequences (presently only test data)
- software for communication and searching.

Members were encouraged to check the website to determine the search characteristics and the way in which the results of searches are presented. At present there is only artificial data on database.

A proficiency testing (PT) study is underway involving the laboratories in Birmingham, Copenhagen, Mainz, and Santiago. It was decided to form an EMPOP PT group consisting of these laboratories and the Innsbruck laboratory. The EMPOP PT group will suggest a system for assessing the quality and acceptance of mtDNA results to the EDNAP group. It was appreciated that there will be other quality assurance issues, and it realised that not all scientifically relevant data can be presented at the Internet. However, the holders of the database should have as much information as acceptable by the providers and the local regulations. It is the intention to try to make this information available through the database holders on written request and after written permission by the supplier of the information.

3. Planned Projects
3.1 SNP

Ate Kloosterman

There has been no progress since the meeting in Dublin as there has been no input from the commercial companies. Applied Biosystems (AB) cannot become involved until it is known how many laboratories will participate, how many SNapShot kits are required and who will do the pilot work. The meeting agreed that the way forward is with the use of Y chromosome SNPs. Since STADNAP is coming to an end, it was decided to leave the subject to the EDNAP meeting.

4. Updates from other groups

4.1 ENFSI

Peter Gill/Ate Kloosterman

The population survey using SGM-plus is still on going and it is hoped to have all databases by the end of the year. Fst values etc. will be calculated.

A proficiency testing exercise with 10 samples offered by the FSS has been completed.

An on-going proficiency testing exercise is offered by GEDNAP.

A Low Copy Number exercise using handprints on acetate sheets has shown some variation in results but the analysis has not been completed.

Now that the EU grant is finished, there could be some funding problems with the QA audits for member laboratories.

At the last meeting in Wiesbaden on 16/17 March 2000 there were 54 participants. The next meeting will be in Cracow in September 2000.

4.2 Interpol DNA Monitoring Expert Group

Richard Scheithauer

The group now consists of nine members (specialists) from Australia, Europe, South America and the USA; others will be added in the future. Richard Scheithauer is the EDNAP representative and presently the chairman of the group. The aim is to help member states to support DNA investigations, set up national DNA databases, and establish a linking site in Lyon for accessing all databases as appropriate within a legal framework.

A handbook will be produced which will include experience from EDNAP, ENFSI, SWGDAM etc. The contributions have already been made and Interpol will print the final product. Interpol is collecting wishes and offers for training and education in DNA typing etc.

Next meeting will be in December 2000 in Melbourne. The homepage www.interpol.int is now available.

4.3 PCWG

Richard Scheithauer

The last meeting was on 9 June 2000 and included a topic on the establishment of an Europol DNA database for organised crime. As this could prove impossible within existing legislation,
a paper is being prepared with the objective of finding a way of searching current European databases. The document urge the member states to use the 7 STR systems recommended by ENFSI as the minimum in national DNA databases.

5. Update from companies

5.1 Applied Biosystems (AB) Nicola Oldroyd

A new system has been developed - AmpFSTR Identifier. This is a 16 locus system in which the maximum size range and the primer sequences have not altered. The dye placement, however, has changed and an extra dye has been added. The launch will be in October 2000 via the 310. So far, validation is only for databasing in 25 ul reactions and not for casework. Work is currently taking place with the 377 but the 5-dye system will only work with Windows NT and not Mac.

Validation for the 3700 is complete and a publication is in preparation. This still requires some spectral calibration data.

From 1 August 2000, all kits have been licensed also for paternity testing, but this may not be stated on the label.

The 3100 16 capillary model uses the same technology as the 310 with automated polymer filling, sample injection and analysis. At present, there are 36 and 50-cm capillaries available, and 22 and 80-cm capillaries are being developed. The capillary array-loading header is now more robust. Run time is approximately 46 mins for STR typing and the throughput is 736-samples/24 hrs. (This compares with 48-samples/24 hrs for the 310 and 384 for the 377). The instrument has cooling and heating facilities. Instrument validation is underway and it is hoped for a completion date sometime in December 2000.

A 5 dye Snapshot multiplex kit is being developed which will offer higher throughput and enhanced software integration.

AB Genemapper v1.0 software incorporates a combination of Genescan/Genotyper and GenBase functionalities. It includes a built-in database and is primarily for allele identification and flagging of questionable results. It will not be available for the Mac.

New initiatives include new extraction procedures, a real-time PCR quantitation method and sensitivity evaluations. AB is organising a series of user meetings focused at information to the forensic science community.

5.2 Promega Tom Mozer

Promega have developed the READIT technology with SNP analysis in mind. This methodology can determine base matches via an 'opposite' polymerase reaction with detection using a luciferase mediated light emission. Activity is only achieved with a perfect match and therefore sequence specific results are obtained.

The method can also be used for quantitation of DNA with a low signal to noise ratio. This is possible even with high levels of bacterial contamination. The technology has the capability
of quantitating mtDNA and, following singleplex reactions, it can also be used for determining SNPs. For example can microtitre plates with a suitable light detector be used for reading results.

The product should be ready within the next six months and any interested members should contact Promega for information.

Promega would be interested in collaboration with EDNAP and members concerning e.g.

- DNA kvantitation by the Alu 3 kit
- SNP typing - including the Y-chromosome SNPs with Readit kit.

5.3 Amersham Pharmacia Biotech (APB) Jackie Evans

APB has developed the MegaBACE 1000 sequencing system that is capable of both sequencing and genotyping. The equipment consists of a 96 glass capillary system (in sets of 16 capillaries) in which the gel medium is loaded at one end and the samples at the other. It is possible to load 96 samples, wait for 2 minutes for electrophoresis and then add a further 96 samples. This can be repeated for approximately 10 times. Primers and dyes of choice can be used. The cost is expected to be approximately £140,000.

The Genetic Profiler analysis system requires minimal editing time and can cope with multiple run analyses. Validation has been carried out on sizing precision, reproducibility, resolution and sample/capillary success.

There is a method for SNP analysis (SNuPe), and APB is looking for collaboration on both SNP and STR development.

Website at www.megabace.com

6. Final reports from the work packages

6.1 WP1 State of the art survey Ate Kloosterman

The report will include the present state of STR and SNP development together with initiatives from the various commercial companies. There will also be a discussion of the options for the future.

Any members who have views on the content of this WP report should contact Ate by 20 September 2000.

6.2 WP2 Inter-laboratory exercises Peter Gill

Synopses of the publications regarding inter-laboratory exercises will be included in the report. Status reports of on-going exercises will be prepared as follows:

- Degraded material - Peter Schneider
- mtDNA analysis - Gillian Tully
- STRs from telogen hairs - Hermann Schmitter
These contributions will be prepared by 20 September 2000

6.3 Technology transfer programme

Niels Morling

Any outstanding reports concerning transfers should be sent to Angel Carracedo and Steve Rand as soon as possible. A list of those who have received training, etc. is available.

Steve Rand will complete this report.

6.4 WP4  Population database compilation

Denise Syndercombe Court

So far, approximately 38,000 genotypes typed with reference to an allelic ladder have been collected. Most were typed using a 377 or a 310 but a considerable number had been analysed with silver staining.

The meeting agreed that data should be accepted which had been obtained using unsequenced ladders but results when no ladders were used were unacceptable. The report should include data sets which do not show statistical fits and all results should have identifiers removed (the identifiers should remain in the laboratory). Laboratories who submitted results but do not have a QA programmes should be flagged.

6.5 Cost statements

Angel Carracedo

All outstanding bills must be sent immediately to Angel Carracedo. Cost statements are required by 20 September 2000

7. Any other business

There was no other business.

Enclosures

Annex 1. List of participants