MORLING

EUROPEAN DNA PROFILING GROUP - ROME - 19 APRIL 1991

Report of Meeting

The meeting was held at the Universita del Sacro Cuore, Roma. Chairman: Peter Martin

Vince Pascali welcomed the members to the meeting. Peter Martin thanked Vince Pascali and Ernesto d'Aloja for all their hard work in the preparation of the venue. He also welcomed Sylvie Frakowiak and Corinne Gagnor as the representatives of the French police laboratories.

Steve Rand gave apologies for Bernd Brinkmann who could not attend. Bertrand Ludes gave apologies for Patrice Mangin who was overseas.

2nd EDNAP Collaborative Exercise

Peter Gill outlined the salient features of the document circulated prior to the meeting.

The aim of the 2nd exercise was to see if we could achieve results which matched within much smaller limits. Having all used a standard protocol the results were compared, being aware that not all factors could be standardised.

He described the necessary criteria for determination of the allele size and then carried out a simulation exercise to determine whether matches had been achieved.

For the determination of a match he used the 2.8% guideline. Having determined that there is a match it is then necessary to calculate the probability that the stain originated from the suspect/victim. For this determination to be made each laboratory must have:-

- (a) reproducible day-to-day methodology
- (b) population frequency distributions
- (c) genomic control which falls within a 'window'

For YNH 24

achieved > 99% agreement at a 2.8% match criterion achieved 100% agreement at a 3.6% match criterion

For MS 43A

The match criterion was much greater due to the size difference in alleles. (The higher the molecular size of the allele, the greater the degree of error.)

Peter Gill then described the Bayesian approach recommended by Evett et al with reference to the data held on the Met Lab Index.

Comparing results from duplicate blood samples he asked the question "what is the strength of evidence provided knowing that the samples match". The results indicated that very strong evidence of match can be obtained using only two probes (YNH 24 and MS 43A) and a Bayesian analysis.

He then went on to see how often evidence of a false match might be obtained by using a computer to compare every sample on the Met Lab index with every other (knowing that none should match). Only 1 in 100,000 comparisons would provide strong evidence of a match which was not real.

If information is to be exchanged a Bayesian approach will give a priority rating to the matches and help to ensure all matches are identified.

On a European level the following appear to be important:-

- (a) Common enzyme
- (b) Common probes
- (c) Marker ladder
- (d) Genomic control
- (e) Electrophoretic system

The method of calculating the allelic (fragment) size is also important.

A uniform system has advantages:-

- (a) Band sizes, determined in different labs, are directly comparable.
- (b) The same statistical methodology can be used for interpretation.

Local Protocols

Each laboratory was asked to report on the comparison of local protocols with a uniform system.

Münster:

Differences in molecular sizes were obtained when different marker

ladders were used.

Zurich:

Although the electrophoretic separations were performed for a long period at low voltage no significant differences were seen between acetate or borate buffers. This laboratory will now employ borate

buffer for routine use.

Copenhagen:

The local protocol includes ethidium bromide and a different agarose.

Only small differences were observed.

Oslo:

The local protocol employed a different buffer and agarose and significant differences were seen. At 14kb there was a difference of

700 bases.

Rome:

A different marker 'ladder' was used for routine work and differences of approximately 1% were observed. This could be due to the system of band measurement.

of band measurement.

Strasbourg:

No differences were seen with YNH 24. however MS 43A with acetate buffer showed a maximum of 300 bp difference.

Wiesbaden:

The local protocol used is similar to the uniform system apart from the marker 'ladders'. It was noted that different scientists obtained different measurements.

Met Lab:

There was no significant difference as the local protocol is similar to the uniform system with the exception of the use of ethidium bromide.

Santiago:

This lab also reported no significant difference as protocols are similar.

London Hospital

) Neither laboratory had sufficient time to perform the exercise.

Mainz

After discussion it was suggested that laboratories who had experienced difficulties could exchange information to resolve problems.

Effect of Temperature on Electrophoretic Separations

Steven Rand and Matthew Greenhalgh presented a collaborative exercise.

Observations of the results from Munster and the MPFSL in previous exercises seemed to indicate a consistent difference although the protocols used in the two labs were very similar. The main difference being that at Munster the gels were run at 4°C whilst at the MPFSL they were run at room temperature. To test this observation the same DNA sample was run repeatedly at both 4°C and room temperature in both laboratories.

Small differences in allele size (approximately 1%) were noted in each lab between the different temperatures. however the magnitude and direction of change was not the same when comparing the two laboratories. It is thought that temperature does have a small effect on band size measurement but that other factors such as choice of marker ladder can have a greater effect. Further work is now in progress.

In the ensuing discussion Vince Pascali suggested that a change in temperature should affect the marker 'ladders' to the same extent as the samples. (There was some disagreement on this point.)

He further suggested that we should investigate the different methods employed for measuring band sizes. Laboratories could exchange autoradiographs and make local measurements and a subsequent comparison could be made.

It was agreed that CRSE and the Met Lab would co-ordinate an exercise to circulate autoradiographs from each laboratory.

Quality Management

Peter Gill outlined his views on the paper which had been previously circulated.

Testing can be on 2 levels:-

(a) the ability to obtain the right answer (proficiency testing)

(b) a complete management system which allows for advice and assistance.

To achieve (b) it is necessary to have a DNA Working Group and this would provide for an interactive system rather than a simple QA exercise. It would allow for:-

(a) trouble shooting

- (b) production of a master protocol which would
 - i) achieve standards
 - ii) become the property of the EDNAP group, which would make all the decisions.

Progress could be achieved by co-ordination and collaboration between member laboratories.

Viv Emerson stated that we should only be interested in the fact that laboratories attain a standard which has been set by EDNAP.

CRSE would provide a Quality Assurance programme which could either be used as proficiency test or as part of a Quality Management system.

For the first exercises laboratories would be given the results with no comments on performance.

In the discussion which followed Steve Rand suggested that some laboratories might join the scheme solely to avoid adverse criticism. He said that he could see many problems with the QA programme.

Viv Emerson said that CRSE would contact each laboratory after the meeting.

Replies to Viv Emerson by mid June.

Future of EDNAP

Vince Pascali presented his thoughts on the future of the group. He stated the aims and the achievements which had been made.

At present we are a group of 8 university laboratories and 5 state police laboratories and he asked the members why we should expand to include other labs.

As it had been agreed that we should encompass other laboratories in Europe he suggested that the present membership should form an executive committee and invite representatives from other countries to attend meetings.

He felt that there were issues to be addressed beyond the integration process and that we should select objectives which are compatible with the EDNAP aims. Initially the group could become a European repository for population data and this would have the immediate advantages of

- (a) an extended array of frequencies
- (b) a deeper insight into population genetics

He went on to discuss the problem of funding which is a major consideration for some of the member laboratories. As most of the EEC programmes are running we cannot get funding for 1991 but we must apply for 1992 as soon as possible.

Viv Emerson has already sent a letter requesting funding for 1992 and has probably been successful in funding the TREVI meeting from the "Human Genome Project". He said that there were other schemes which we could try, eg. common collaboration of measurement within the Europe Biomedical and Health Project. In order to get funding for the mobility and interchange of researchers within Europe we need to establish centres of excellence with some from of training facility.

The discussion then returned to the future of EDNAP and Peter Martin was asked to make an application for EDNAP to become a Working Group within the International Society for Forensic Haemogenetics. (See attached proposal.)

The next meeting will be on Saturday afternoon 21 September 1991 following the ISFH meeting in Mainz. This will be a short meeting designed to inform others who wish to take part in future meetings.

Proposal to International Society for Forensic Haemogenetics

There was a unanimous decision at the meeting in Rome on 19 April, to make an application to the International Society for Forensic Haematologics to become a working group called EDNAP.

Aims:

To meet the original aims of EDNAP

- (a) To determine accurately alleles using single locus probing
- (b) To facilitate the exchange of data between countries.
- (c) The group will only study DNA extracted from stained material.

Further aims:

- (i) Set and maintain standards within DNA profiling for European laboratories engaged in stain work.
- (ii) To provide a forum for the exchange of population frequency databases and collaboration in research.

Composition:

(i) Executive committee

One member from each laboratory comprising the existing EDNAP group, plus one representatives from each of the following countries:

Greece

Ireland

Belgium

Portugal

Sweden

Austria

Finland

Iceland

Malta

who wish to join and have a commitment to attain the standard already achieved.

(The composition of the Executive can be further discussed in Mainz.)

(ii) All scientists wishing to participate in the aims of the working group.

We intend to hold open meetings in conjunction with the Society.

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On Wednesday, 15 May 1991 the above delegates will join D Lingard's group for the CRSE presentations.