Genomic Arabidopsis Resource Network

Issue 1
21st Dec 2000 **The First GARNet Functional Genomics Meeting, October 2-3, 2000.**

Availability of tools for functional genomics can change the way plant science is approached. Isolating one gene, characterising it, and moving on to the next is in Arabidopsis is now only one of many ways to approach research. Completion of the Arabidopsis sequence has opened a wealth of information. To get maximum benefit from this information, access to functional genomics tools is essential.

The BBSRC IGF initiative (Investigating Gene Function) was started not only to allow the best possible access to functional genomics tools and resources for UK researchers, but also to develop and improve these. Seven different IGF programs have started since January 2000, for Drosophila, microbial eukaryotes, Streptomyces, farm animals, cereals, Brassica and Arabidopsis. The different IGF programs will be funded for 3 years after which many of the services will continue on a cost recovery basis.

The Arabidopsis IGF project is called GARNet (Genomic Arabidopsis Resource Network). GARNet will be organising three annual functional genomics meeting during the time it receives BBSRC sponsorship as part of its remit to make the availability of these tools and the kind of results that can be achieved using them widely known to UK plant scientists. The first of these meetings took place at the University of York, on October 2 and 3, 2000. It was a combined event for the Arabidopsis and Brassica IGF projects.

Nearly 250 UK plant scientists met for 2 days, to hear about the functional genomics resources and services that will form part of GARNet and to discuss possibilities for their experiments with the service providers. Several international speakers were invited to demonstrate the potential of using functional genomics tools by discussing their data. Others introduced international efforts in functional genomics. The meeting program is still available at the GARNet website http://garnet.arabidopsis.org.uk.

- The Proteome Analysis Service provided by Paul Dupree at the Cambridge Centre for Proteomics will become available for the plant research community early in 2001. Paul Dupree also provides the proteomics service for the Drosophilla IGF project. 2D differential PAGE analysis will form one of the cornerstones of the service. Paul reported great increase in the sensitivity of proteome comparison with the use of fluorescent Cye dye labelling of the proteins. This way, up or down regulated proteins can be detected with much increased sensitivity and reproducibility in complex protein mixtures. MS polypeptide identification from selected spots will lead to gene identification. It will also be possible for users to provide samples of purified proteins for identification. Justin Roberts (University of California, Riverside) talked about affinity chromatographic techniques for selective purification and concentration of proteins not accessible with conventional purification techniques. This facilitates Mass Spectrometry and 2D electrophoresis and helps in assigning biochemical properties to predicted ORFs.

- The Transcriptome Analysis Service was introduced by Sean May from the Nottingham Arabidopsis Stock Centre (http://arabidopsis.org.uk). The service will consist of a complete labelling, hybridisation, analysis and data mining service Transcriptome arrays in the form of macro and micro arrays will also be provided, either of a non redundant set of Arabidopsis ESTs or of a gene sequence tag (GST) set. David Lonsdale (JIC) and Jim Beynon (HRI Wellesbourne) will provide the GST set, as part of a European collaboration. As part of quality control for micro arrays, some standard baselines will be generated, comparing different ecotypes, major tissue types and a simple developmental timeline. To date, we cannot exactly say when the first micro arrays will become available. This information will be available on

  http://garnet.arabidopsis.org.uk, and posted to arab-uk.

- Pam Green showed results from the initial rounds of the AFGC (Arabidopsis Functional Genomics Consortium) micro array service, which uses the MSU (Michigan State University) EST set. An important resource that is being created is the ʻspot historiesʼ . This allows visitors of the website (http://afgc.stanford.edu/) to follow the behaviour of one specific gene through the range of experimental

applications. It will be a very useful tool for predictions of relations between gene activities.

- Mike Beale introduced a gas chromatography - mass spectrometry (GS-MS) and liquid (HPLC) chromatography – mass spectrometry (LC-MS) Metabolite Analysis Service for Arabidopsis that has the aim of analysing specific compound classes and developing the methods to provide a metabolic finger print of plant extracts. A mass spectral library of Arabidopsis metabolites will be created. The service is aimed at screening of mutants, tagged lines, or different treatments for changes in metabolite profiles. A high degree of automation will be established to provide high throughput. Since the GARNet meeting, Mike has also started to use NMR as a metabolomic tool, and this looks promising for pattern matching type analysis. The service started in October 2000. Since then various applications have been received and approved by the steering committee. Applications can be submitted via http://garnet.arabidopsis.org.uk

- Oliver Fiehn from the MPI for Molecular Plant Physiology showed that over 1,000 distinct compounds of Arabidopsis leaf extracts can be quantified using GC or LC – MS. Of these compounds, currently about 200 are identified. Newly identified compounds suggest the presence of metabolic pathways previously not identified in plants. Cluster analysis and statistical analysis are used to formulate hypothesis on plant gene functions in comparisons between wild types and mutants or different ecotypes.

- Tools of forward and reverse genetics are being created at the Sainsbury Laboratory, NASC and the John Innes Institute.

- Ian Bancroft and his team at GeTCID (The Gene Transfer Clone Identification and Distribution service) are carrying out screens of large insert binary vector clone libraries. Some possible applications are confirmation of T-DNA tagged lines, complementation analysis, transfer of Arabidopsis genes to other plant species and positional cloning (creation of bi-bac tiling path through region of interest). Four different libraries are available, the binary cosmid library (BC) and the TAC library (K) that were already available, and the JAtY and YAtC libraries that were made as part of the GARNet project. The selection markers are respectively Kanamycin, Hygromycin, Phosphinotricin (Basta) and Kanamycin. This service started in Juli 2000 and a number of screenings has already been carried out. Applications for this service can be submitted to the steering committee, through the GARNet website at http://garnet.arabidopsis.org.uk. More information is available at http://www.jic.bbsrc.ac.uk/staff/ian-bancroft/arabIGF.htm.

- Jonathan Jones is creating a population of transposon mutagenised lines. The SLAT lines that were created earlier (60.000 dSpm insertions in Col-0) are very useful for reverse screens. For forward screens they are a poor tool, because there are too few insertions per plant. For GARNet a new population will be constructed with a high copy number of dSpm insertions. About 5,000 lines with about 30 independent inserts per line will be created. The lines will become available via the Nottingham Arabidopsis Stock Centre.

- Macro arrays for knockout identification will be produced at NASC from existing reverse-genetic DNA sets (the SLAT lines) and if appropriate also from BACs.

- ATIS, the Arabidopsis Transposon Insertion Service (http://www.jic.bbsrc.ac.uk/STAFF/michael-bevan/ATIS/index.htm) will sequence transposon tags from single insertion mutant populations. Jonathan Clarke, who is the service manager for Mike Bevan and Jonathan Jones talked about the three different populations from which SINS (Sequence INsertion Sites) will be sequenced, the SLAT, ACTIVATE, and FGT collections. They form respectively a resource for loss-of-function mutations, gain-of-function mutations and gene expression patterns via translational fusions with the reporter gene GUS. SINS can identify a potentially disrupted gene, and define the exact position of the insertion. The ATI database that is under construction will integrate the SINS with the annotated Arabidopsis genome sequence and make the data publicly accessible (http://formaggio.cshl.org/~h-liu/attdb/index.html). The FGT gene trap lines are being constructed under a different grant. SINS on the FGT lines will only be made available after a way has been found to bulk the seeds.

- Sean May and his staff at the Nottingham Arabidopsis Stock Centre will provide Bioinformatics for GARNet. The existing databases, AGR (Arabidopsis genome database) and UK-cropnet will be extended to suit the needs of GARNet bioinformatics. AGR will contain library-screening data (GeTCID) and possibly primer sequence data. For the transcriptomics, proteomics and metabolomics data a new database will be created called NTPdB (NASC Transcriptomics Project Database). This will be a fully relational MySQL database, seamlessly linked with AGR and integrated with the Stanford Microarray Database.

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- GARNet intends to integrate with other functional genomics projects wherever possible. Currently GARNet participates in collaboration for the exchange of 3,000 primer pairs with AFGC and CSIRO. A European collaboration for GST primer exchange is also starting. One of the partners in this project is GABI (Genome Analysis in the BIological system plant), the German plant genomics project. Thomas Altmann from the MPI for Molecular Plant Physiology in Golm discussed the general background of GABI. Like GARNet, GABI will be developing resource centres, but unlike GARNet, GABI will also carry out a diverse range of research projects. Eight different plant species (Arabidopsis, barley, sugar beet, rapeseed, potato, poplar, maize and rice) will be used in the various GABI projects.

We have started preparations for the 2001 GARNet functional genomics meeting. This meeting will again be in collaboration with the Brassica IGF project. It will take place at the University of York, on **September 27 and 28, 2001**. The format of this meeting will be different from the first meeting. There will be updates about the services, and there will be international speakers again. But mainly, we would like to hear from the users of different resources. We intend to have poster sessions and oral presentations from users from the different services.