2008 Clone-based Functional Genomics Resources (ORFeomics) Subcommittee Report

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Significant progress continues to be made towards achieving the long-term community goal of obtaining full length (FL)-cDNAs and open reading frame (ORF) clones for all annotated *Arabidopsis thaliana* protein coding genes. In addition, many other clone-based resources are being produced for gene functional analysis such as those useful for protein:protein interaction mapping. A detailed description of many of these clone resources and their utility is available (Bleys, A., Karimi, M., and Hilson, P. (2008) Cloned-based functional genomics. Methods Mol. Biol. In press). This brief update focuses on highlights from this past year in the area of ORF and FL-cDNA clone production. Running totals from all the major projects are included in Table 3.

1) Yale group (Kumar/Snyder and colleagues)

This group has produced 5,090 open-end ORF clones (ORFs containing a start but no stop codon) that have been deposited with ABRC (Table 3). Information about these ORF clones (sequence validation, vector-type, Atxgxxx name etc.) can be found at this website (http://plants.gersteinlab.org/) under 'clone status' subheading. Their goal for the coming year is to add 5,000 more open-end ORF clones into the database.

2) Salk/Invitrogen (Ecker and colleagues)

During the past year this group has produced 12,107 Gateway ORF clones (ORFs containing both native start and stop codons) that have been fully validated and deposited with ABRC (Table 3). Information about these ORF clones: sequence validation, vector-type, gene (Atxgxxx) name, etc, can be found at this website: http://methylome.salk.edu/cgi-bin/clones.cgi

3) DFCI/Salk (Vidal and colleagues)

During the past year this group has produced 9,036 activation domain (AD) yeast two-hybrid Gateway clones and corresponding transformed yeast strains and 8,916 DNA binding domain yeast two-hybrid Gateway clones and corresponding transformed yeast strains. Glycerol stocks for the ~18,000 clones/strains are being prepared and will be deposited in the ABRC by September, 2008. Information about these clones: sequence validation, vector-type, gene (Atxgxxx) name, etc, will be available at this website: http://methylome.salk.edu/cgi-bin/clones.cgi

4) ATOME (Lurin and colleagues)

Over the past several years, this group has constructed ~5,000 ORF entry clones (with and without stop codons.) These clones can be ordered from CNRGV, a French stock center in Toulouse: (http://cnrgv.toulouse.inra.fr/en). They are currently constructing a database which will be open to the public and will present their clone information. Until this is completed, it is suggested that investigators view available information on their website (http://urgv.evry.inra.fr/orfeome/) and to order clones by sending an email to infocnrgv@toulouse.inra.fr. In the coming year, as part of the of Agron-omics project, this group plans to clone 500 ORFs (ORFs with and without stop codons) which are not currently available in other public ORF collections.

5) RIKEN (Shinozaki and colleagues)

- cDNA clone project: RIKEN PSC and BRC groups have determined full-length sequences of 7,766 RAFL cDNAs in collaboration with National Institute of Genetics (PI: Yuji Kohara), which was supported by National Bioresource Project in Japan. Contact Persons for this project: Motoaki Seki (email: mseki@psc.riken.jp), Kazuo Shinozaki (e-mail: sinozaki@rtc.riken.jp) and Masatomo Kobayashi (e-mail: kobayasi@rtc.riken.jp). Therefore, the total number of RAFL cDNA clones whose full-length sequences have been determined is 21,005 as of March 24, 2008. The number includes the results of the collaboration with the SSP groups. Some of the RAFL clones are mapped to the same AGI code according to TAIR7 gene model. The RAFL cDNA clones are available from RIKEN BRC.

- ORF clone project: RIKEN PSC groups have collected ORF clones for 443 Arabidopsis transcription factors that are not found in the RAFL cDNA collection, and determined their sequences in collaboration with National Institute of Advanced Industrial Science & Technology (PI: Masaru Ohme-Takagi). Contact Persons for this project: Kazuo Shinozaki (e-mail: sinozaki@rtc.riken.jp) and Masaru Ohme-Takagi (e-mail: m-takagi@aist.go.jp). The ORF clones will be available from RIKEN BRC.

Table 3. Arabidopsis ORF and cDNA clone repertoires*

Creator	Format	Focus	Validation	Scale	URL	Stock center†
ORF clones						
SSP consortium & Salk Institute	Univector pUNI51		Full sequence	14,214	signal.salk.edu/cdnastatus.html http://methylome.salk.edu/cgi-bin/clones.cgi	ABRC
Salk/Invitrogen	Gateway entry		Full sequence	12,114	signal.salk.edu/cdnastatus.html http://methylome.salk.edu/cgi-bin/clones.cgi	ABRC
TIGR	Gateway entry	Hypothetical genes	Full sequence	3,041	www.tigr.org/tdb/hypos/	ABRC
Peking-Yale Joint Center	Gateway entry	Transcription factors	5' and 3' end seq.	1,282		ABRC
Dinesh-Kumar et al.	Gateway expression	TAP-tagged transcription factor	5' and 3' end seq.	1,281		ABRC
REGIA	Gateway entry	Transcription factors	5' and 3' end seq.	962	gabi.rzpd.de/materials/	GABI/ RZPD
Dinesh-Kumar et al.	Gateway entry, no stop pLIC-CTAP	Plant protein chips	5' and 3' end seq.	1,527 (5,299)	plants.gersteinlab.org/	ABRC
ATOME 1	Gateway entry		5' and 3' end seq.	1,809	urgv.evry.inra.fr/orfeome/	CNRGV
ATOME 2	Gateway entry, no stop	Originates from SSP	5' and 3' end seq.	3,476	same	CNRGV
Doonan et al.	Gateway Expression	GFP fusion for subcellular location		155		ABRC
Callis et al.	Gateway entry	Protein ubiquitination	Full sequence	111	plantsubq.genomics.purdue.edu	ABRC
Sheen et al.	Expression	Epitope tagged MAPK	Full sequence	100	genetics.mgh.harvard.edu/ sheenweb/category_genes.html	ABRC
cDNA clones						
RIKEN/SSP/ Salk Insitute	λ ZAP or λ PS		Full sequence	21,508	www.brc.riken.go.jp/lab/ epd/Eng/order/order.shtml	BRC
MPI-MG	Gateway expression		5' end seq.	4,500	gabi.rzpd.de/materials/	GABI/RZPD
Génoscope/LTI	Gateway entry		Full single pass seq.	28,866	www.genoscope.cns.fr/ Arabidopsis	CNRGV

*Modified from P. Hilson, personal communication

†Stock centers distributing Arabidopsis clone repertoires:

- Arabidopsis Biological Resource Center, (ABRC, USA), http://www.biosci.ohio-state.edu/pcmb/Facilities/abrc/abrchome.htm
- RIKEN BioResource Center (BRC, Japan), http://www.brc.riken.jp/lab/epd/Eng/catalog/pDNA.shtml
- GABI Primary Database (GABI/RZPD, Germany), http://gabi.rzpd.de/
- National Resources Centre for Plant Genomics (CNRGV, France), http://cnrgv.toulouse.inra.fr/ENG/index.html
- European Arabidopsis Stock Centre (NASC, UK), http://arabidopsis.info/
- BCCM/LMBP Plasmid and DNA library collection (BCCM/LMBP, Belgium), http://bccm.belspo.be/db/lmbp_gst_clones/
- Open Biosystems Inc., USA, www.openbiosystems.com/