

**U.S. Poultry Species Coordination Activities**  
*Supported by Allotments of Regional Research Funds, Hatch Act*  
*For the Period 1/1/03-12/31/03*

**Overview:** Coordination of Poultry Genome Mapping under the National Animal Genome Research Program (NAGRP) is a joint effort of Michigan State University (MSU) and the USDA, ARS, Avian Disease and Oncology Laboratory (ADOL). CSREES support is allocated via NRSP-8. The NAGRP is made up of the membership of the Animal Genome Technical Committee, including the Poultry Species Subcommittee.

**FACILITIES AND PERSONNEL:** Jerry Dodgson, Department of Microbiology & Molecular Genetics, MSU, serves as Coordinator with Hans Cheng of ADOL as Co-Coordinator. Both MSU and ADOL provide facilities and support.

**OBJECTIVES:** 1. Develop high resolution comparative genome maps aligned across species that link agricultural animal maps to those of the human and mouse genomes, 2. Increase the marker density of existing linkage maps used in QTL mapping and integrate them with physical maps of animal chromosomes, and 3. Expand and enhance internationally shared species genome databases and provide other common resources that facilitate genome mapping.

PROGRESS TOWARD OBJECTIVE 1. High resolution poultry genome maps.

**The Reference Linkage Map(s).** Numerous labs have cooperated in mapping DNA-based polymorphic markers by genotyping samples from the same two international reference crosses, the Compton population (Bumstead and Palyga, *Genomics* 13, 690-697, 1992), and the East Lansing population (Crittenden *et al.*, *Poultry Science* 72, 334-348, 1993). This map has been enhanced by genotyping of a third cross, the Wageningen population, by Martien Groenen and colleagues (Groenen *et al.*, *Genomics* 49, 265-274, 1998). A consensus map based on all three map populations has been published (Groenen *et al.*, *Genome Res.* 10:137-147, 2000). Updates (Schmid *et al.*, *Cytogenet. & Cell Genet.* 90:169-218, 2000) bring the number of markers on the consensus map to 1965, placed into 50 linkage groups, covering around 4000 cM. The East Lansing map has expanded to 1249 markers on 43 linkage groups (other evidence places E46 on GGA2 and E66 on GGA5, but there is not enough statistical support in our map alone to establish these linkages). Over 320 genes have been mapped in the East Lansing map. These provide evidence that gene order is conserved between the human and chicken genomes to a remarkable extent. Progress on the linkage map has slowed as work has shifted towards physical maps and preparing for the genome sequence, as described below.

PROGRESS TOWARD OBJECTIVE 2. Physical maps and map integration.

Zhang, Dodgson and colleagues have generated a library of over 115,000 BACs (~15X) at Texas A&M and have fingerprinted 65,000 of these (Lee *et al.*, *Animal Genetics* 34:151-152, 2003). A first generation physical map of the chicken genome containing 2331 contigs has been published (Ren *et al.*, *Genome Research* 13:2754-2758, 2003). P. de Jong (Childrens Hospital of Oakland Research Institute, CHORI) has generated a ~10X BAC library (CHORI-261) with extra large inserts using DNA we provided from the same bird used for the Texas A&M BACs. CHORI has also prepared a large insert turkey BAC library (CHORI-260) in collaboration with Kent Reed and Dave Harry. The Washington U. Genome Sequencing Center (WUGSC) has been provided copies of the Texas A&M and CHORI-261 chicken BAC libraries and has fingerprinted 188,369 of them, generating over 133,000 useable BAC

fingerprints. These have been used to generate a second generation BAC contig map comprised of about 280 contigs, two-thirds of which have been anchored to the genetic linkage/chromosome map by techniques described below. The map should be submitted for publication and become publically available in January. The Dodgson lab has been integrating the BAC contigs with the linkage map primarily by screening the libraries for genetic markers using overgo hybridization (Romanov *et al.*, *Cytogenetics and Genome Research*, in press). Their efforts, plus those of cooperating scientists who've been provided BAC filters (see below), have led to over 5700 BAC assignments to over 720 distinct markers or genes. These linkages are an essential part of the contig and sequence assembly processes. Crooijmans and Groenen have also fingerprinted their BAC library (Crooijmans *et al.*, *Mamm. Genome* 11:360-363, 2000) and building contigs.

Burnside, Cogburn, Morgan, Cheng, Reed and Neiman in the U.S. and several European groups have generated EST collections and applied them to microarray analysis. Boardman *et al.* (*Current Biology* 12:1965-19-69, 2002) announced the sequencing of over 300,000 chicken ESTs from a wide variety of tissues and developmental stages. Data are posted in Genbank and in a searchable format at [www.chick.umist.ac.uk](http://www.chick.umist.ac.uk). A joint project between the U. of Manchester (UMIST, **Simon Hubbard**, **Stuart Wilson**, and **Paul Boardman**) and the Sanger Institute (**Jane Rogers**) is sequencing full length chicken cDNA clones (goal of 10,000) using both UMIST and other libraries.

Radiation hybrid (RH) panels have been constructed by Vignal and colleagues at INRA (Morisson *et al.*, *Genet. Sel. Evol.* 34:521-533, 2002), and a framework RH map is in the process of being constructed. This should be done by early 2004.

The Washington U. Genome Sequencing Center (WUGSC) has completed ~6X sequencing of the chicken genome (primarily whole genome shotgun) and this is now being assembled. The full assembly should be done by year's end. A paper will appear in the first half of 2004 describing the sequence. A test 5.2X sequence assembly covered ~1 Gbp (~97% coverage). As of Dec. 12, the NCBI Trace Archive (<http://www.ncbi.nlm.nih.gov/Traces/trace.cgi?>) contains 11,524,114 chicken sequences or over 3.8% of the total Archive.

PROGRESS TOWARD OBJECTIVE 3: Database and other map resources.

**ChickGBASE:** The latest version of ChickGBASE is constructed in the comparative mapping Arkdb format. Arkdb was primarily developed by Andy Law, Dave Burt, Alan Archibald, and others at the Roslin Institute. ChickGBASE is available in the Arkdb format at <http://www.thearkdb.org/browser?species=chicken>. A mirror site for the poultry database has been mounted at the Iowa State database site, <http://www.genome.iastate.edu/>. James Reecy has taken over direction of all bioinformatics efforts for the NAGRP, including chicken.

**WWW Homepage:** We maintain a WWW homepage for the Poultry Genome which links to ChickGBASE, the Roslin Institute homepage, and a variety of other genome mapping resources. The Homepage provides the latest EL maps and mapping data, an updated list of published microsatellites, descriptions of available resources, the latest cytogenetic map, and access to a host of other information relating to both genetic and physical maps. It can be accessed at <http://poultry.mph.msu.edu>.

**Reference Panel DNA:** DNA from the East Lansing international reference population has been sent to many laboratories throughout the world.

**Primer Kits:** Several kits of microsatellite primer pairs have been made available for free distribution. The first of these is the **Population Tester Kit**. This contains 9 primer pairs which define microsatellites with high polymorphic information content (numerous alleles widely distributed in several populations). A new version of a framework primer kit (with 147 well-spaced microsatellite marker primer pairs) called the "**Comprehensive Mapping Kit #7**" was made available last year (thanks in part to a generous donation from Hy-Line International). One or more kits have now been provided ~120 different labs, worldwide. Two **Chicken Gene Primer Pair** kits have been made available containing a total of 300 primer pairs to sequenced chicken genes for use in EST mapping and expression analysis.

**Physical Mapping Resources:** At least three public **BAC libraries** for chicken and one for turkey are now available. These are described above under Objective 2 progress. **Filter arrays of robot-spotted BAC clones** are being distributed for both the Texas A&M and CHORI-261 chicken BAC libraries (so far, 46 requests have been filled). **PCR-screenable BAC DNA pools** from row, column and superplate pools of the first 30,000 clones of the *Bam*HI library have been supported, in part, by Coordination funding and are now available. Once a clone(s) of interest has been identified, it (they) can be obtained at nominal cost from the Texas A&M GENefinder BAC Center or CHORI.

**Chicken Microarray:** A 13K chicken spotted cDNA glass slide array is now available from the Array Facility at the Fred Hutchinson Cancer Research Center, FHCRC. This is the result of combined efforts of FHCRC (Jeff Delrow and Paul Neiman), the U. of Delaware (Joan Burnside), GSF, Munich (Jean-Marie Buerstedde) and the Roslin Institute (Dave Burt) with partial support from Coordination funds. FHCRC arrays are available at \$150 per array. NAGRP Coordination funds have been used to make a small number of free test arrays available to NAGRP members, in hopes of getting as many users involved as possible.

**Newsletter:** The Poultry Genome Newsletter is published quarterly and is distributed through our WWW Homepage, electronically on the angenmap email discussion group and via hard copy to scientists worldwide.

**Meetings:** Over 1800 scientists attended the joint Plant and Animal Genome XI meeting held last January, held jointly with the annual NAGRP meeting. Coordination funds helped support attendance at PAG-XI and will do so again for the upcoming PAG-XII in January, 2003. A meeting, organized by AviGenics, Inc. and the U. of Georgia, was held Nov. 13-14, 2003 in Atlanta, GA on the "Chicken Genome Sequence: Impact and Applications". A large turnout of both industry and academic scientists attended. Another meeting on "Livestock Genomes: Sequence Annotation and Informatics Challenges" was sponsored by Texas A&M University and the Baylor Genome Sequencing Center in October that discussed plans for and applications of genome sequences of all major farm animals. The Coordinator also gave an invited presentation on the chicken genome at the Advances in Genome Biology & Technology Meeting, Marco Island, FL in February, 2003.

## PLANS FOR THE FUTURE.

### OBJECTIVE 1. High resolution poultry genome maps.

Efforts will be made to improve overall map marker density as a result of the upcoming genome sequence. Comparative chicken/human map development will also continue and QTL analysis should expand. New primer panels based either on SNPs and/or microsatellites will be developed.

### OBJECTIVE 2. Physical maps and map integration.

Additional efforts are underway to fill gaps between contigs, improve map resolution and integrate these physical maps with existing linkage maps. All sequence contigs will need to be aligned with the linkage map and the cytogenetic maps. EST and array approaches will continue to expand for both chicken and turkey in the U.S. and in Europe. RH panel framework mapping should finish in 2004. Assembly, editing, and annotation of the sequence will be major tasks for the upcoming year.

### OBJECTIVE 3: Database and other map resources.

The chicken ChickGBASE database will continue to be updated and improved at the Roslin Institute and the Iowa State mirror site. Discussions are underway about what types of bioinformatic resources will be required to handle the full genome sequence and related data. At least part of this role will be filled by the Ensembl group in Europe, the National Center for Biotechnology Information at NIH, and the U. of California Santa Cruz sites in the same way they now serve the human and model organism genomes. These and other locations will greatly enhance the comparative map between the chicken and other vertebrate genomes. Poultry Genome Newsletter and homepage information will be continue to be distributed and enhanced. We also will continue to distribute reference panel DNAs, microsatellite primer panels, BAC library resources (library, clones, filter sets, PCR screening pools) and Type I gene-based primers. Chicken microarrays are now available through the Fred Hutchinson Cancer Research Center Genome Facility with initial development support from coordination and other funds. Discussions have begun with Affymetrix and other companies interested in providing new array resources based on the genome sequence.

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