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Microsoft® Excel® spreadsheet-based animal colony management for genetically altered animals

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Animal research has significantly advanced our understanding of mammalian systems and disease. The need for genetically altered animal models, particularly mice, is expected to increase due to the Human and Murine Genome Projects that plan to identify all genes, and explore their role in relation to other genes. It has been these authors's experience, that despite the anticipated value of gene targeted and transgenic animal studies toward this goal, many institutions do not maintain a transgenic mouse colony, in part due to the high costs of genotyping (and associated housing of animals during the genotyping delay) and extensive record keeping required with these colonies. Federally mandated regulatory oversight of animal colonies also dictates a high quality of animal care, including health monitoring and record keeping. Thus, these facilities are a major fiscal burden to every institution that maintains research animals.

In addition to routine animal care expenses, genetically altered animal colonies incur additional costs associated with the need for rapid and accurate assessment of the genotype for every animal produced. A tissue sample (tail snip or blood sample) is obtained from the progeny, the chromosomal DNA is isolated, and genotyping data is generated to identify progeny harboring the perti-

nent genetic alterations. The genotyping information is essential to identify mice that will be designated as experimental or breeding stock. Breeding strategies can be very complex, depending on the genotype of the breeding stock and the desired genetic make up of the progeny. Genotype considerations are not limited to the induced chromosomal changes, but can also include the genetic background strain of the line that harbors the genetic alterations. Thus, essential to the efficient management (and financial survival) of a transgenic colony is the ability to rapidly generate and communicate genotyping data, breeding instructions, and breeding productivity between the end user(s), the animal care staff, and the genotyping staff.

Animal colony management has been an issue of concern for many years (1-8). There are a number of animal colony management software programs available, each with their own advantages and special adaptations. Colony management software costs can range from gratis [CoPE (a collaborative pedigree drawing environment) Software. Retrieved December 8, 2003 from <http://www.nslj-genetics.org/soft/cope.pdf>] to greater than \$1 million for one license (Progeny Software, LLC. <https://www.progeny2000.com/products.html>; Southwest Foundation for Biomedical Research.

Genetic Analysis and Pedigree Management Software. <http://www.sfb.org/sfbr/public/software/software.html>; colony management databases. <http://maillists.uci.edu/mailman/public/uci-mouseusers/2001-December/000007.html>). In each case, employees must be trained in the software operation, and if a problem arises, the facility is solely dependent on the technical support of the originator. A nightmare scenario is that the researcher or colony manager is unable to access the database after they are dependent on it. Cost-effectiveness and ease of operation are also important factors when deciding on animal colony management software, especially in view of potentially high turn over of animal care staff and limited training resources. The process described here is a very basic system that is inexpensive, well documented, and readily modified to meet the special needs of any colony/end user. The only software requirement is Microsoft® Excel®, and the only hardware requirement (in addition to a personal computer) is a mechanism to allow the same Excel file to be accessed by multiple users (animal care staff, genotyping facility, and end user staff). In the Excel workbook, related information is stored in one database (also called a file) that can contain many spreadsheets, which organize, list, and analyze related information. Group access to the database is easily accomplished on any local area network.

Figure 1 describes the overall process, illustrating the interactions and the information flow between all staff and the two databases (breeding and genotyping) that are maintained as two separate Excel workbooks. Entries into the genotyping database are initiated by the animal care staff when a litter is weaned, ear-tagged, and a tissue sample is sent to the genotyping facility (which might also be the end users laboratory). The animal care staff enters the weanling data [date of birth (DOB), unique identifier or tag number, sex, parents] for all progeny (this data set can be modified to meet the needs of each facility and can include other information deemed essential, such as cage #, room #, rack #, name of animal care staff entering data, etc.). See Figure 2A for sample genotyping database entries. Results of genotyping assays (PCR or Southern blot analysis) are entered by the genotyping facility and should

include citations that allow the end user to find the primary data (notebooks or other databases). The genotyping facil-

ity adds the genotyping results and notifies the end user that the results are in the genotyping database. The end user is

then able to access this information and assess several factors: (i) productivity of breeders; (ii) ratio of transgene passed from generation to generation; (iii) sex; (iv) age; and (v) number of mice available for breeding and experimental use. This information allows the end user to predict (and request) the number of breeder stock necessary to maintain colony production at the desired level and to schedule experiments utilizing the animals produced. With this information, the end user can then modify the breeding database accordingly.

The breeding database (Figure 2B) provides all instructions to the animal care staff that are necessary to maintain the line. The information includes ear tag information, source and genotype of breeding stock, type of genetically altered line (transgenic, knockout, knock-in, etc.), and instructions for interpreting the genotyping data. In addition, the breeding database provides breeding instructions indicating the numbers of breeding

Figure 1. Colony management strategy.

BENCHMARKS

pairs, backup breeders, and experimental mice to maintain. Very important for a large facility, this database also contains information regarding the cage card designations both for breeding stock and

experimental stock. This database can be modified to include any and all information needed by either the animal care staff or the end user. It is very easy to add important information for the evaluation of

breeders' performance, such as the breeders age, when the breeders were set up for breeding, when they had their first litter, how many litters they produced, each litter size, and when tails were expected

A

Ear Tag #	DOB	Sex	Sire	Dam	Genotype	PCR Protocol	PCR #	Agarose Gel #	Southern Blot Autorad#	Comments
NAR 231	1/31/2001	M	NAR 100	C57BL/6	+	BGAL	371	300	NONE	
NAR 232		M			-					
NAR 233		M			+					
NAR 234		F			+					
NAR 235		F			-					
NAR 236		F			+					
NAR 237	2/5/2001	M	NAR 104	C57BL/5	+		382	321	NONE	
NAR 238		M			-					
NAR 239		F			-					
NAR 240		F			+					
NAR 241		F			-					

B

Flag	Strain Name & Line	Ear Tag	Breeding Strategy	TG or KO	Genotyping Conversion	TMOS Desired (default 3)	Breeder Stock	Breeder Back Up Stock	Expertl Stock Desired	Expertl Stock Actual	Breeder Card Data: Line Name; Breeder Info; ACUC Protocol #	Comments	Purpose
	BGL line 4	BGL	BGL + X C57BL/6	TG	BGL + = Breed; BGL - = Cull	3	3 Trios	3+	10+; 10-	4;4	BGL Line 4 (BGL+ x C57BL/6); 174-dmd-mi	Notify DMD When Stock Reaches 10 and 10	Expertl mice
	MUKO X MUKO	MUKO	MUKO +/- X MUKO +/- (from MUKO x C57 cross)	KO	NEO+,MU+ = +/- (heterozygous); NEO-,MU+ = WT,NEO+; MU- = -/- homozygous KO	25	21 Trios	0	50 Each +/-,+/+,-/-	17;4;8	MUKO +/- X MUKO +/-; 174-DMD-MI	Notify DMD When Stock Goals are Reached	Expertl mice
	MUKO x C57	MUKO	MUKO +/- x C57BL/6	KO	NEO+,MU+ = +/- (heterozygous); Neo-,MU+ = WT	3	3 Trios	2 +/- Females	0	0	MUKO +/- X C57BL/6; 174-DMD-MI	No- tify When Reach Back Cross 10	1. Generate breeders for MUKO x MUKO; 2. Back Cross MUKO into C57BL/6
3/11/01 DMD...note increased breeders requested, notify dmd when 8 trios obtained	DAC Line 21	DAC	DAC + (M) X B6SJL (F)	TG	DAC+ = Breed; DAC- = Cull	8	3 Trios	3+	Males only; 12+; 12-	7;4	DAC + X B6SJL; 174-DMD-MI	Tag and Tail Only Males; Cull All females	Expertl mice

Figure 2. Sample spreadsheets used in Excel workbook. (A) Genotyping database spreadsheet for a hemizygous transgenic line. (B) Breeding database spreadsheet. This spreadsheet illustrates entries describing a variety of genetic alterations: hemizygous transgenic lines (BGL, DAC), a line harboring a targeted allele that is solely producing mice for experimental use (MUKO x MUKO), and the same line harboring a targeted allele that is being used to produce breeders and is involved in a backcross (MUKO x C57). Note the sample instructions given in the flag box for DAC line 21.

Figure 3. Sample PCR genotyping photodocumentation. Genomic DNA from recently weaned progeny is isolated, and PCR products specific to the transgene are sized on an agarose gel. Each lane (1–7) represents one animal from which the genomic DNA was derived. The positive control PCR product (A) is produced by primers that recognize an endogenous murine gene (e.g., hemoglobin) and is present in all animals. Transgenic animals are positive (+) for the transgene-specific PCR product (B). Nontransgenic animals lack the B fragment (lanes 1, 2, 3, 6, and 7) and are negative (neg) for the transgene. Other control lanes include lane 8, with no DNA sample, and lane 9, which contains the DNA size markers *PhiX174HaeIII* (Invitrogen, Carlsbad, CA, USA).

for genotyping. Similarly, columns could be added to indicate the number of mice per cage, total number of cages from the same founder (or the same breeders), any phenotypes noted, and whether or not the trio is breeding. Links, such as a picture, can be added. There is also room for other idiosyncratic information or comments from the staff that are deemed essential to the breeding of that particular line. The first column on the breeding database is called the FLAG column. This is used by the end user to indicate to the animal care staff that there is a new instruction. As the end user makes standard work requests to the breeding database, the FLAG box for that strain is manually colored red, indicating to the animal care staff that there is a new instruction (Figure 2B, A5). The date and initials of the scientist/end user making the flagged request are also noted in the FLAG. Any questions regarding the request can usually be clarified quickly in a phone conversation, without the end user needing to go to the animal colony to give instructions in person. When the work is completed, the FLAG column works in reverse, the initials of the animal care staff, pertinent comments, and date of completion are added to the column, and the FLAG column is manually changed to green. The end user and animal care staff managers can then be informed in a timely manner when the work is completed, allowing better predictions of colony productivity

and resource utilization.

The Excel workbook is extremely user-friendly, allowing the user to create separate spreadsheets for individualized needs. Each spreadsheet is accessed by clicking on a tab at the bottom of the screen and can be used for a new line or strain, a new end user, or to archive data. Technical requests such as timed matings, hormone injections, blood sampling, etc., outside of standard work requests, can also be directed to the animal care staff managers. After the manager determines resource availability and enters the request in the breeder database, cop-

ies of the request can be attached or stored in a separate spreadsheet in the Excel workbook. Thus, these separate spreadsheets allow flexibility in the organization of the database.

We have found that this process reduces the need for meetings, verbal and miscellaneous e-mail communications, and trips by the end user to the animal housing rooms. The manipulation of the Excel workbook is easily learned and establishes good “animal care staff/end user communication” with all instructions given in written format so that all managers are well informed and can thus better evaluate performance, while avoiding the problems associated with verbal miscommunications that are often routine in this process. Within a very short time, both the end user and the animal colony staff begin to appreciate the power of the Excel workbook, and staff at all levels provide feedback as to additional information needed in the colony databases for the efficient maintenance of each line. Such additions are usually easily added to the workbook.

As the colony grows, the Excel workbook might become limited in functionality. However, if the process proves successful, then the data from the workbooks can be imported into the more powerful Microsoft Access® or other relational database, which can support much larger data sets and allow the development of a system with great-

er capabilities. Other improvements to this strategy can also be readily established, such as a database that stores photodocumentation (e.g., electronically captured PCR results) (Figure 3), allowing the end user to verify the primary data from the genotyping facility.

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