GV-SOLAS
Gesellschaft für Versuchstierkunde

Specialist Information

from the Committee for Genetics and

Laboratory Animal Breeding

Objective and Methods of Genetic Monitoring of Isogenic Mouse and Rat Strains

Status: July 2014

Authors:
Kurt Reifenberg, Heidelberg
Hans Hedrich, Hannover
Dirk Wedekind, Hannover
and
Norma Howells, Heidelberg*
(*not a member of the committee; contribution by translating into English language)

Contents

1. Introduction

Isogenic mouse and rat strains [inbred strains, congenic, co-isogenic, consomic, recombinant inbred (RI), and recombinant congenic (RC) strains] are indispensable for biomedical and biological research. The significance of the scientific results that are generated using isogenic animal models depends entirely on the genetic authenticity of the strain being used. Therefore setting up of programs for genetic surveillance of isogenic animal strains is irrevocably indispensable. Genetic surveillance is not only the responsibility of the experimenter but as a consequence of the centralization process observed in most animal facilities also represents a task of the facility management. The manuscript that is presented by the Committee for Genetics and Laboratory Animal Breeding of the Society of Laboratory Animal Science (GV-SOLAS) is aimed to describe the objectives and methods available for genetic monitoring programs.

2. Impact of the genetic authenticity in isogenic experimental animal strains

Genetic authenticity of isogenic animal strains can be affected by genetic contamination, by mutation and by differential fixation of residual heterozygous loci. With inbred strains that have reached a high generation number (F > 60 generations) residual heterozygosity is marginal. Spontaneous mutations will unavoidably occur in the genome of inbred strains and these mutations can lead to a phenotype alteration (Drake et al., 1998). Furthermore, genetic contamination can destroy the authenticity of isogenic animal strains. Principally, genetic contaminations can be avoided by appropriate animal facility management; nevertheless past experiences have indicated that contamination *de facto* frequently occurs (Bitter-Suermann and Lewis, 1980; Kendall et al., 1986; Naggert et al., 1995; Simpson et al., 1997; Sharp, 1981; Threadgill et al., 1997; Benavides 1999; Nitzki et al., 2006).

3. Objective of genetic monitoring

Principally a genetic quality assurance program must fulfill two requirements:

- It must be able to prove genetic authenticity in isogenic strains.
- It must be suitable for genotyping specific differential alleles or to control their expression

4. Methods for control of genetic authenticity

4.1 Polygenically inherited morphological and patho-physiological strain characteristics

4.1.1 Pigmentation

The coat color of mice and rats is a very valuable parameter for authenticity control of inbred strains; basically due to the ease of evaluation. Excellent contributions exist for the complex coat color genetics of mice (Silvers, 1979, Searle, 1968). Coat color variations of the rat have been extensively described by Hedrich (1990).

4.1.2 Physiological reproduction data

The colony index for isogenic strains (number of weaned offspring per breeding female per week); is as a rule due to inbreeding depression significantly lower than the breeding performance of the various outbred and hybrid strains. Significant elevation of the colony index of a reputed isogenic strain is thus a strong indication of genetic contamination.

4.1.3 Behaviour

Appearance of obvious changes in the behaviour of isogenic mice or rats can also be caused by genetic factors. Only appropriately qualified and trained animal care personnel is in the position to observe and recognize behavioral changes in mice and rats.

4.1.4 Patho-physiological strain characteristics

The diversity of isogenic mouse and rat strains is reflected in their different specific phenotypes and abundant information is available, facilitating the selection of specific strains for specified experiments. For example, NOD mice and BB rats develop diabetes mellitus type 1 and SHR rats exhibit hypertension. Loss of such characteristic patho-physiological properties represents a reliable indication of genetic contamination of such strains.

4.2 Skin transplantation

Due to the isogenicity of inbred animals, rejection of tissue transplantation within the strain does not occur. Therefore, skin transplantation technique can be used to verify the isogenic histocompatibility of inbred strains. Tissue rejection is determined from an array of gene loci which are designated as histocompatibility (H) loci. The major histocompatibility complex (MHC) has significant importance, however, the minor histocompatibility loci also have influence on the transplantation result (Simpson 1991). Skin transplantation offers

the possibility to check the identity of the H-loci between the donor and the recipient animal. With divergent MHC the transplant would be acutely rejected (< 2 weeks); with disparity of one or more minor histocompatibility loci the rejection reaction can take a chronic course and is prolonged over several months; but an acute course can also take place. Normally orthotopic tail skin transplantation (Bailey and Usama, 1960) is conducted, however, occasionally the earlobe is heterotopically transplanted to the side of the chest wall. Typically transplantations are performed on groups of animals using standardized methods (reciprocal circular system by Bailey and Kohn, 1965, or the double ring system of Zeiss, 1966).

4.3. General marker sets

4.3.1 Definition of general market sets

A general marker set is a collection of specific loci or gene products that are suitable for generating a genetic profile of an inbred strain and thus monitor genetic authenticity of the strain. The marker set should provide the following requirements:

- The marker set should have a minimum of 1 to 3 markers per chromosome in adequate genetic or physical distance. The number of markers per chromosome is determined by the chromosomal size and the markers of a specific chromosome should be evenly distributed. Furthermore, additional informative markers must be available in sufficient numbers and density for special purposes.
- The individual markers of the sets should exhibit a high degree of genetic polymorphism to ensure optimal informative output for the genetic monitoring program.
- The marker set must be able to facilitate differentiation between closely related inbred strains.
- The genotypes or phenotypes of the markers can be determined with reasonable and justifiable costs.

4.3.2 Biochemical markers

Biochemical markers represent proteins (in most cases enzymes), which are subject to a genetic polymorphism. Biochemical markers are not typed at the genetic level but are analyzed at the protein level. To this end organic homogenates or body fluids are separated by electrophoresis and subsequently subjected to a protein specific reaction.

Formerly the program for genetic monitoring of isogenic mouse strains of The Jackson Laboratory contained a set of 25 biochemical markers. These markers are listed in Appendix 1 including their chromosomal position and reference literature. In addition the “ICLAS Manual for Genetic Monitoring of Inbred Mice” has published a set of biochemical markers (Esaki et al., 1984). Furthermore a set of biochemical markers for authenticity control of isogenic rat strains has been published by Hedrich (1990, see Appendix 2).

4.3.3 Immunological markers

Immunological markers either represent gene products with immunological functions or proteins that can be detected by means of immunological methods. The original genetic mouse monitoring program of The Jackson Laboratory contained three immunological markers for analyses, subjacent also the MHC. These markers are listed in Appendix 3 inclusive of chromosomal position and reference literature. In the "ICLAS Manual for Genetic Monitoring of Inbred Mice" (Esaki et al., 1984) there is also a list of immunological markers which can be used for authenticity control of isogenic mouse strains. In the monograph by Hedrich (1990) an immunological marker set for authenticity control of isogenic rat strains can also be found (see Appendix 4). The MHC represents a significantly important immunological marker group. Since all T cell and most B cell immune reactions are restricted by the MHC, monitoring of the MHC locus is particularly important for immunological animal experiments. Under the internet address of The Jackson Laboratory a list can be seen of inbred murine haplotypes and available intra-MHC recombinant inbred strains (ftp://ftp.informatics.jax.org/pub/datasets/misc/H2Haplotypes/H2_haplotypes.html). Various MHC haplotypes of rats are described by Hedrich (1990) and are listed in the archives of the Institute for Laboratory Animal Science of the Hannover Medical School. On this internet site information with regard to the genetic profile of many rat strains is also available (http://www.mh-hannover.de/2652.html). For many MHC antigens of mice, monoclonal antibodies have been developed, most of them are available for purchase. Therefore, verification of the appropriate haplotype can be easily confirmed with the help of a flow-cytometer. Monoclonal antibodies for individual MHCI and MHCII alleles for rats are seldom commercially available. Therefore the MHC haplotypes of rats are mostly determined and confirmed by the use of polyclonal antisera. The methods most commonly used are the dextrane haemagglutination test (MHC class I) and the complement-dependent cytotoxicity test (MHC class I and II). Today, as an alternative to serological verification, proven molecular-genetic methods can be deployed to define the MHC haplotypes of mice and rats. In the meantime polymorphic microsatellites and single nucleotide polymorphisms (SNP) have been introduced, which are positioned in the coding and non-coding regions of the major histocompatibility complex; however, they do not always clearly define the respective haplotype. PCR, hybridization reactions and sequencing techniques are suggested as methods for genotyping.

4.3.4 DNA Markers

In the past a great number of methods were developed to detect DNA polymorphisms, for example "restriction fragment length polymorphisms" (RFLP, Dai et al., 2005); "variable number of tandem repeats" (VNTR, Thebault-Baumont et al., 2003), "random amplification of polymorphic DNA" (RAPD, Alexandrova and Shvemberger, 2005; Keshava et al., 1999) and "amplified fragment length polymorphisms" (ALFP, Vos et al., 1995). Because these methods are costly and time consuming and frequently do not have satisfactory reproducible results, they are only applied in specific cases for genotyping of inbred

strains. Today, in most cases microsatellite analyses and “single nucleotide polymorphisms” (SNP) are used for authenticity control of isogenic mouse and rat strains.

4.3.4.1 Microsatellites

Microsatellites are composed of tandem replications of DNA motifs, which consist of 2 to 4 nucleotides. Microsatellites are therefore also referred to as “short tandem repeats” (STR, Miesfeld et al., 1981). They can be found with high frequency in all mammalian genomes and show a strong degree of genetic polymorphism which is characterized by the number of repeats of the underlying DNA motif and is thus also designated as “simple sequence length polymorphism” (SSLP). Microsatellite polymorphisms can easily be amplified with the help of PCR primers which are homologous to the specific flanking sequences of the microsatellite. The size of the amplified sequence can be determined by gel electrophoresis (agarose or polyacrylamide gel) or by automated systems if fluorescent primers have been used for amplification. A considerable number of databanks are readily available informing about mouse microsatellites and their genotypes in specific isogenic strains. The most prevalent information source is certainly the “Mouse Genome Informatics” (MGI) databank (http://www.informatics.jax.org/). In this extensive databank information can be obtained with regard to specific murine microsatellites, chromosomal position, alleles, strain polymorphism, typing techniques and literature references. For establishment of microsatellite-based authenticity control systems specific data collections are very useful, offering specific microsatellite sets and genotypes for multiple isogenic strains. For example, Dietrich et al. (1992) have described a set of 317 microsatellites and provide information about the genotypes of the inbred strains C57BL/6J-Lep<ob>, Cast/Ei, C57BL/6J, SPRET/Ei, DBA/2J, A/J, C3H/HeJ, BALB/cJ, AKR/J, NON/Lt, NOD/MrkTacBr and LP/J. Similarly, Schalkwyk et al. (1999) describe a panel of 128 microsatellites and polymorphisms for the strains DBA/2, BALB/c, AKR, C57BL/6, C57BL/10, A/J, C3H, 129/J, SJL/J, JF/1 and PWB. Information can also be obtained from the “Center for Inherited Disease Research” with regard to polymorphism of 314 microsatellites for a total of 54 inbred strains. This data collection refers to a publication from Witmer and Colleagues (2003). Lastly, under the Internet address ftp://ftp.informatics.jax.org/pub/datasets/misc/Moore/Moore.xls an excel table can be downloaded which displays a palette of 1562 microsatellites which can identify polymorphisms existing between the 129/SvJ strain and seven other inbred strains (C57BL/6J, DBA/2J, C3H/HeJ, BALB/cJ, Cast/Ei, FVB/NJ, C57BLKS/J). The original work for microsatellites for rats was published by James and Lindpaintner (1997). In the meantime there is a multitude of information available on the Internet with regard to microsatellites of rats. The Rat Genome Database has proved to be user-friendly and practical (http://rgd.mcw.edu/). Information can be obtained from this source for specific rat microsatellites, for chromosomal position, alleles, strain polymorphisms, verification techniques and literature references. Data collections are available from which a selected set of microsatellites for genotyping defined isogenic rat strains can be obtained.

4.3.4.2 Single Nucleotide Polymorphisms (SNPs)

SNPs represent genetic elements which are characterized by the exchange of a single nucleotide. Although principally all four possible nucleotides should be found in a defined DNA position; most SNPs are only found in 2 alleles because the likelihood for a further base exchange at this position is extremely low (Vignal et al. 2002). In spite of this low-level genetic polymorphism (as compared to the multi-allelic microsatellites), SNPs are being increasingly used for molecular genetic analyses. The basis for this is the large number of these elements that can be detected in the genome. For murine SNPs there is a broad range of databanks which are accessible for information; an excellent source is the "Mouse Genome Informatics" (MGI) databank of The Jackson Laboratory (http://www.informatics.jax.org/), they also have a specific SNP databank (http://www.informatics.jax.org/javawi2/servlet/WIFetch?page=snpQF) which supplies information on chromosomal position, strain genotypes, flanking DNA sequences (primer design) and literature references for specific murine SNPs. In addition the "National Center for Biotechnology Information" (http://www.ncbi.nlm.nih.gov/) has an extensive data collection, including an SNP databank.

For setting up an SNP-based authenticity program specific SNP sets with genotype information of isogenic mouse strains are more helpful than databanks. Petkov and colleagues (2004) have described a set of 1638 SNPs with genotype informations about 102 isogenic murine strains. This article can be found under http://www.genome.org/cgi/content/full/14/9/1806, it is available for free and the SNP and genotype collection can be found as an excel table under the category "Supplemental Research Data". In addition the "Center for Inherited Disease Research (CIDR, http://www.cidr.jhmi.edu/) provides information for genotyping SNPs for inbred strains. Presently a lot of effort is being put into developing a high resolution SNP map for the rat genome (Zimdahl, et al., 2004; Smits et al., 2005). But as yet an acceptable economic system for typing of rat SNPs has not been established. Hence the genotyping of rat strains via SNPs has not yet attained the importance which this technology has reached for genetic monitoring of isogenic mouse strains.

5. Methods for strain discrimination in a breeding unit

5.1 Critical marker sets

If defined isogenic mouse or rat strains are bred and maintained concurrently for long periods of time in the same breeding unit (room or barrier), genetic authenticity in the first line is endangered by genetic contamination between these strains; thus by outcrossing within the breeding unit. Under these exceptional circumstances and only over a limited period of time genetic monitoring can be confined purely to the discrimination of those strains co-habiting in the same breeding unit. For these specific inquiries, analysis with a critical marker set is sufficient. The required critical marker set only needs to comprise

those markers that give a clear discrimination between the animal strains within the specific breeding unit.

5.2 Genotyping methods of differential loci

Principally, suitable genotyping protocols need to be established for all differential alleles (e.g. transgenes, knock-out defects, knock-in mutations, spontaneous mutation); which are carried by isogenic strains of a specific facility. Usually, these protocols are based on PCR technology. However, in exceptional cases more complex procedures must be set up. As an example, Lieschke et al. (1994) and Kaneko et al. (1995) describe techniques to type the osteopetrotic (op) allele of the murine Csf1 gene position. If co-isogenic or congenic strains with the same genetic background are maintained in the same breeding unit; a suitable critical marker set must be able to discriminate all differential alleles of all strains existing in this unit.

6. Literature

Adams M, Baverstock PR, Watts CH, Gutman GA (1984) Enzyme markers in inbred rat strains: genetics of new markers and strain profiles. Biochem Genet 22: 611-629.

Alexandrova SA, Shvemberger IN (2005) Genetic variability of the mouse hepatoma cells MH-22a revealed by RAPD-PCR-fingerprinting under different conditions of cultivation. Exp Oncol. 27: 114-9.

Bailey DW, Usama B (1960) A rapid method of grafting skin on tails of mice. Transplant Bull. 7: 424-5.

Bailey DW, Kohn HI (1965) Inherited histocompatibility changes in progeny of irradiated and unirradiated inbred mice. Genet Res. 6: 330-40.

Barclay AN, Jackson DI, Willis AC, Williams AF (1987) Lymphocyte Specific Heterogeneity in the Rat Leukocyte Common Antigen (T200) Is Due to Differences in Polypeptide Sequences near the Nh2-Terminus. EMBO J 6:1259-1264,

Benavides F, Cazalla D, Pereira C, Fontanals A, Salaverri M, Goldman A, Buggiano V, Dran G, Corley E (1998) Evidence of genetic heterogeneity in a BALB/c mouse colony as determined by DNA fingerprinting. Lab Anim 32: 80-5

Benavides FJ (1999) Genetic Contamination of an SJL/J Mouse Colony: Rapid Detection by PCR-based Microsatellite Analysis. Contemp Top Lab Anim Sci. 38: 54-55.

Bender K, Cleve H, Gunther E (1981) A Previously Described Serum-Protein Polymorphism in the Rat Identified as Gc (Vitamin-D-Binding Protein). Anim Blood Groups Biochem Genet.12: 31-6.

Bender K, Nagel M, Gunther E (1982) Es-6, a Further Polymorphic Esterase in the Rat. Biochemical Genetics 20: 221-228.

Bender K, Adams M, Baverstock PR, den Bieman M, Bissbort S, Brdicka R, Butcher GW, Cramer DV, von Deimling O, Festing MF, et al. (1984) Biochemical markers in inbred strains of the rat (Rattus norvegicus). Immunogenetics 19: 257-266.

Bender K, Bissbort S, Brdicka R (1985) Studies of Some Selected Protein Polymorphisms in Inbred and Wild Rats. Transplantation Proceedings 17:1872-1874.

Bitter-Suermann H, Lewis MG (1980) Rejection of skin and spleen "isografts" in strain of Lewis rats. Transplantation. 30:158.

Bogden AE, Aptekman PM (1960) The R-1 Factor, a Histocompatibility Antigen in the Rat. Canc. Res. 20: 1372-1382.

Brdicka R, Sulc K (1965) Different Haemoglobin Patterns in Inbred Lines of Rats. Folia Biologica 11: 328

Chen SH, Donahue RP, Scott CR (1973) The Genetics of Glutamic-Pyruvic Transaminase in Mice: Inheritance, Electrophoretic Phenotypes, and Postnatal Changes. Biochem. Genet. 10: 23-28.

Cohen BL (1960) Genetics of Plasma Transferrin in the Mouse. Genet. Res. 2: 431-438.

Cramer DV (1981) Genetic-Variation of Urinary Pepsinogen and Its Probable Linkage to Albinism in the Rat. Immunogenetics 13: 555-558.

Cramer DV, Mowery PA, Adams M (1986) Biochemical markers in rats: linkage relationships of aconitase (Acon-1), aldehyde dehydrogenases (Ahd-2 and Ahd-c), alkaline phosphatase (Akp-1), and hydroxyacid oxidase (Hao-1). Biochem Genet 24: 217-227.

Dai JG, Min JX, Xiao YB, Lei X, Shen WH, Wei H (2005) The absence of mitochondrial DNA diversity among common laboratory inbred mouse strains. J Exp Biol. 208: 4445-50.

DeLorenzo RJ and Ruddle FH (1969) Genetic Control of Two Electrophoretic Variants of Glucosephosphate Isomerase in the Mouse (Mus musculus). Biochem. Genet. 3: 151-162.

DeLorenzo RJ and Ruddle FH (1970) Glutamate Oxalate Transaminase (GOT) Genetics in Mus musculus: Linkage, Polymorphism, and Phenotypes of the Got-2 and Got-1 Loci. Bioche. Genet. 4: 259-273.

Dietrich W, Katz H, Lincoln SE, Shin HS, Friedman J, Dracopoli NC, Lander ES (1992) A genetic map of the mouse suitable for typing intraspecific crosses. Genetics. 131: 423-47

Drake JW, Charlesworth B, Charlesworth D, Crow JF (1998) Rates of spontaneous mutation. Genetics. 148: 1667-86.

Eicher EM, Stern RH, Womack JE, Davisson MT, Roderick TH, and Reynolds SC (1976) Evolution of Mammalian Carbonic Anhydrase Loci by Tandem Duplication: Close Linkage of Car-1 and Car-2 to the Centromere Region of Chromosome 3 of the Mouse. Biochem. Gen., 14: 651-660.

Esaki K, Festing MFW, Hedrich HJ, Hoffman HA, Mobraaten LE, Moutier R, Nomura T, Radzikowski C, Tomita T, Yosida T (1984) ICLAS Manual for Genetic Monitoring of Inbred Mice, Nomura T, Esaki K, Tomita T (Eds) University of Tokyo Press

Fox RR and Black PM (1987) Erythrocyte antigen-9 (Ea-9)., Mouse News Lett. 78:57

Gasser DL (1972) Seminal Vesicle Protein in Rats - Gene in Fourth Linkage Group Determining Electrophoretic Variants. Biochem. Genet. 6: 61

Greiner DL, Barton RW, Goldschneider I, Lubaroff DM (1982) Genetic-Linkage and Cell Distribution Analysis of T-Cell Alloantigens in the Rat. J. Immunogenetics 9: 43-50.

Hamada H, Petrino MG, Kakunaga T (1982) A novel repeated element with Z-DNA-forming potential is widely found in evolutionarily diverse eukaryotic genomes. Proc. Natl. Acad. Sci. USA 79: 6465-6469

Hamada S, Yamada J, Bender K, Adams M (1987) A New Polymorphic Pepsinogen Locus (Pg-2) in the Rat (Rattus-Norvegicus). Exp. Anim. 36: 267-272.

Hedrich HJ, von Deimling O (1987) Re-evaluation of Lg-V of the Rat and Assignment of 12 Carboxylesterases to 2 Gene Clusters. 78: 92-96

Hedrich HJ, Reetz IC (1990) Typing of Inbred Strains for Rt3 and Mapping of This Antigenic System to Linkage Group-X of the Rat. Transplantation Proceedings 22: 2559-2560

Hedrich, H. J. (1990). Genetic monitoring of inbred strains of rats, Gustav Fischer, Stuttgart

Henderson NS (1965) Isozymes of Isocitric Dehydrogenase: Subunit Structure and Intacellular Location. J. Exp. Zool. 158: 263-273.

Herzenberg, L.A., Dora K. Tachibana, Leonore A. Herzenberg, and Leon T. Rosenberg (1963) A Gene Locus Concerned with Hemolytic Complement in Mus musculus. Genetics 48: 711-715.

Hjorth JP (1979) Genetic Variation in Mouse Salivary Amylase Rate of Synthesis. Biochem. Genet. 17: 665-682

James MR and Lindpaintner K (1997) Why map the rat? Trends Genet. 13: 171-173

Kaneko H, Iizuka K, Morohashi T, Kokai Y, Fujimoto J (1995) Non-phenotypic detection of osteopetrotic (op/op) mutation by using PCR-SSCP analysis. Laboratory Animals 29: 442-6.

Kendall C, Geiger D, Wagner JE (1986) Survey of genetic authenticity of commercially produced inbred rats. Lab Anim Sci.36: 655-8.

Kendall PB (1983) The Gene Esterase-12 and Esterase-13 Polymorphisms in the Norway Rat. 17: 221-224.

Kendall PB (1985) Glucose phosphate dehydrogenase polymorphism and the genetics of linkage group II in the Norway rat (Rattus norvegicus). Lab Anim 19: 169-172.

Keshava C, Keshava N, Zhou G, Whong WZ, Ong TM (1999) Genomic instability in silica- and cadmium chloride-transformed BALB/c-3T3 and tumor cell lines by random amplified polymorphic DNA analysis. Mutat Res. 425: 117-23.

Klein J, Figueroa F, Klein D (1982) H-2 haplotypes, genes, and antigens: second listing. I. Non-H-2 loci on chromosome 17. Immunogenetics 16: 285-317.

Kluge R, Hedrich HJ, von Deimling O (1990) Genetic Variation and Biochemical Properties of Esterase-18 (Es-18) in the Laboratory Rat (Rattus-Norvegicus) - a New Locus of Esterase Cluster 2 in Linkage Group-V. Biochem. Genet. 28: 57-68

Kondo Y, Hamada S, Serikawa T, Yamada J (1987) 2 Polymorphic Genetic-Loci of Rat Tear Proteins. Transplantation Proceedings 19: 3146-3147.

Kunz HW and Gill TJ (1978) Red blood alloantigenic systems in the rat. J. Immunogenet. 5: 365-382

Lewis WHP and Truslove GM (1969) Electrophoretic Heterogeneity of Mouse Erythrocytes Peptidase, Biochem. Genet. 3: 493-498.

Lieschke GJ, Stanley E, Grail D, Hodgson G, Sinickas V, Gall JA, Sinclair RA, Dunn AR (1994) Mice lacking both macrophage- and granulocyte-macrophage colony-stimulating factor have macrophages and coexistent osteopetrosis and severe lung disease. Blood 84: 27-35.

Matsumoto K, Matsuhashi A, Aizawa M (1982) A New Genetic-Variation of the Malate Dehydrogenase-Like Enzyme (Mdl-1) in Inbred Rats and Its Possible Linkage. 20: 443-448.

Matsumoto K, Gasser DL (1983) Polymorphism of Glutathione S-Transferase in Laboratory Rats. Biochem. Genet. 21: 1209-1215.

Matsumoto K, McCafferty E, Neilson EG, Gasser DL (1984) Mapping of the Genes for Tubular Basement-Membrane Antigen and a Submaxillary-Gland Protease in the Rat. Immunogenetics 20: 117-123.

Meo T, Douglas T, and Rijnbeek AM (1977) Glyoxalase I Polymorphism in the Mouse: a New Genetic Marker Linked to H-2. Science 198: 311-313.

Miesfeld R, Krystal M, Arnheim N (1981). A member of a new repeated sequence family which is conserved throughout eucaryotic evolution is found between the human delta and beta globin genes. Nucleic. Acids Res. 9: 5931-5947

Mizuno M, Susuki K (1978) Genetic-Variation of Pancreatic Alpha-Amylases in Rat, Rattus-Norvegicus. Japan. J. Genet. 53:137-142.

Naggert JK, Mu JL, Frankel W, Bailey DW, Paigen B. (1995) Genomic analysis of the C57BL/Ks mouse strain. Mamm Genome. 6: 131-3.

Nichols EA, Chapman VM, and Ruddle FH (1973) Polymorphism and Linkage for Mannosephosphate Isomerase in Mus musculus. Biochem. Genet. 8: 47-53.

Nichols EA and Ruddle FH (1975) Polymorphism and Linkage of Glutathione Reductase in Mus musculus. Biochem. Genet. 13: 323-329.

Nitzki F, Kruger A, Reifenberg K, Wojnowski L, Hahn H (2006) Identification of a genetic contamination in a commercial mouse strain using two panels of polymorphic markers. Lab Anim. UK: 41: 218-228

Peters J and Nash HR (1976) Polymorphisms of Es-10 in Mus musculus. Biochem. Genet. 14: 119-125.

Peters J and Nash HR (1977) Polymorphisms of Esterase-11 in Mus musculus, a Further Esterase Locus on Chromosome 8. Biochem. Genet. 15: 217-226.

Petkov PM, Ding Y, Cassell MA, Zhang W, Wagner G, Sargent EE, Asquith S, Crew V, Johnson KA, Robinson P, Scott VE, Wiles MV. (2004) An efficient SNP system for mouse genome scanning and elucidating strain relationships. Genome Res. 14: 1806-11.

Popp RA and Popp DM (1962) Inheritance of Serum Esterases Having Different Electrophoretic Patterns Among Inbred Strains of Mice. J. Hered. 53: 111-114.

Popp RA (1965) Hemoglobin Variants in Mice. Fed. Proc. 24: 1252-1257

Popp RA (1966), Inheritance of an Erythrocyte and Kidney Esterase in the Mouse. J. Hered. 57: 197-201.

Rosenberg LT and Tachibana DK (1962) Activity of Mouse Complement, J. Immunol. 89: 861-867.

Ruddle FH and Roderick TH (1968) Allelically Determined Isozyme Polymorhisms in Laboratory Populations of Mice. Ann. NY Acad. Sci. 151: 531-539.

Ruddle FH, Shows TB, and Roderick TH (1968) Autosomal Control of an Electrophoretic Variant of Glucose-6-Phosphate Dehydrogenase in the Mouse (Mus musculus). Genetics 58: 599-606.

Schalkwyk LC, Jung M, Daser A, Weiher M, Walter J, Himmelbauer H, Lehrach H (1999) Panel of microsatellite markers for whole-genome scans and radiation hybrid mapping and a mouse family tree. Genome Res. 9: 878-87.

Searle AG (1968) Comparative Genetics of Coat Colour in Mammals. Logos Press Limited

Seeley TL and Holmes RS (1981) Genetics and Ontogeny of Butyryl CoA Dehydrogenase in the Mouse and Linkage of Bcd-1 with Dao-1. Biochem. Genet. 19: 333-345.

Sharp DW (1981) Rejection of islet isografts in a strain of Lewis rats. Transplantation 31: 229-230

Shiroishi T, Sagai T, Moriwaki K (1981) A simplified micro-method for cytotoxicity testing using a flat-type titration plate for the detection of H2 antigens. Microbiol Immunol 25: 1327-1334.

Shows TB and Ruddle FH (1968) Malate Dehydrogenase: Evidence for Tetrameric Structure in Mus musculus. Science 160: 1356-1357.

Shows TB, Ruddle FH, Roderick TH (1969) Phosphoglucomutase Electrophoretic Variants in the Mouse, Biochem. Genet. 3: 25-35.

Shreffler DC (1960) Genetic Control of Serum Transferrin Type in Mice. Proc. Natl. Acad. Sci., USA 46: 1378-1384.

Sick K and Nielsen JT (1964) Genetics of Amylase Isoenzymes in Mice. Hereditas 51: 291-296.

Silvers WK (1979) The Coat Colors of Mice: A Model for Mammalian Gene Action and Interaction, Springer Verlag, New York (elektronisch verfügbar unter http://www.informatics.jax.org/wksilvers/)

Simpson E (1991) Minor histocompatibility antigens. Immunol Lett. 29: 9-14.

Simpson EM, Linder CC, Sargent EE, Davisson MT, Mobraaten LE, Sharp JJ (1997) Genetic variation among 129 substrains and its importance for targeted mutagenesis in mice. Nat Genet. 16: 19-27

Smits BM, Guryev V, Zeegers D, Wedekind D, Hedrich HJ, Cuppen E (2005) Efficient single nucleotide polymorphism discovery in laboratory rat strains using wild rat-derived SNP candidates.. BMC Genomics. 6: 170.

Soars ER (1979) Identification of a New Allele of Es-1 Segregating in an Inbred Strain of Mice. Bioche. Genet. 17: 577-583.

Thebault-Baumont K, Dubois-Laforgue D, Krief P, Briand JP, Halbout P, Vallon-Geoffroy K, Morin J, Laloux V, Lehuen A, Carel JC, Jami J, Muller S, Boitard C (2003) Acceleration of type 1 diabetes mellitus in proinsulin 2-deficient NOD mice. J Clin Invest. 111: 851-7.

Threadgill DW, Yee D, Matin A, Nadeau JH, Magnuson T (1997) Genealogy of the 129 inbred strains: 129/SvJ is a contaminated inbred strain. Mamm Genome. 8: 390-3.

van Zutphen LF, Lagerwerf A, Bouw J, den Bieman MG (1981) Biochemical polymorphisms in the rat. Biochem Genet. 19:173 - 186,

Vignal A, Milan D, SanCristobal M, Eggen A (2002) A review on SNP and other types of molecular markers and their use in animal genetics. Genet Sel Evol. 34: 275-305.

Vos P, Hogers R, Bleeker M, Reijans M, van de Lee T, Hornes M, Frijters A, Pot J, Peleman J, Kuiper M et al. (1995) AFLP: a new technique for DNA fingerprinting. Nucleic Acids Res. 23: 4407–4414.

Walker MC and JM Phillips-Quagliata (1985) A new erythrocytic antigen of C57BL/10 (B10) mice. Proc Soc Exp Biol Med 178: 402-6

Wilcox FH, Hirschhorn L, Taylor BA, Womack JE, Roderick TH (1979) Genetic Variation in Alkaline Phosphatase of the House Mouse (Mus musculus) with emphasis on a Manganese-Requiring Isoenzyme. Biochem. Genet. 17: 1093-1107.

Witmer PD, Doheny KF, Adams MK, Boehm CD, Dizon JS, Goldstein JL, Templeton TM, Wheaton AM, Dong PN, Pugh EW, Nussbaum RL, Hunter K, Kelmenson JA, Rowe LB, Brownstein MJ (2003) The development of a highly informative mouse Simple Sequence Length Polymorphism (SSLP) marker set and construction of a mouse family tree using parsimony analysis. Genome Res. 13: 485-91.

Womack JE and Eicher EM (1977) Liver-specific Lysosomal Acid Phosphate Deficiency (Apl) on Mouse Chromosome 17. Mol. Gen. Genet. 155: 315-317.

Zeiss IM (1966) Demonstration of isohistogenicity in groups of rats from partially inbred strains. Transplantation. 4: 506-9.

Zimdahl H, Nyakatura G, Brandt P, Schulz H, Hummel O, Fartmann B, Brett D, Droege M, Monti J, Lee YA, Sun Y, Zhao S, Winter EE, Ponting CP, Chen Y, Kasprzyk A, Birney E, Ganten D, Hubner N. (2004) A SNP map of the rat genome generated from cDNA sequences. Science. 303: 807.

Appendix 1

Biochemical markers used by The Jackson Laboratory for authenticity control of isogenic mouse strains:

Biochemical Marker	Gene	Chromo-some	Literature
Salivary Amylase	*Amy1*	3	Hjorth, 1979; Sick und Nielsen, 1964
Acyl-CoA Dehydrogenase	*Acads*	5	Seeley und Holmes, 1981
Alkaline Phosphatase 1	*Akp1*	1	Wilcox et al., 1979
Apolipoprotein A1 & Transferin	*Apoa1 und Trf*	9	Cohen, 1960; Shreffler, 1960
Carbonic Anhydrase 2	*Car2*	3	Eicher et al., 1976
Esterase 3, 10, and 11	*Es3, Es10 und Es11*	11, 14 und 8	Popp, 1966; Peters and Nash, 1976; Peters und Nash, 1977
Esterase 1	*Es1*	8	Popp und Popp, 1962; Ruddle und Roderick, 1968; Soars, 1979
Glucose-6-Phosphate Dehydrogenase	*G6pd1*	4	Ruddle et al., 1968
Glyoxalase-1-Structural	*Glo1s*	17	Meo et al., 1977
Glutamat Oxaloacetate Transaminase 1 und 2	*Got1 and Got2*	19 und 8	DeLorenzo und Ruddle, 1970
Glucose Phosphate Isomerase 1 Structural	*Gpi1s*	7	DeLorenzo und Ruddle, 1969
Glutamic Pyruvic Transaminase 1	*Gpt1*	15	Chen et al, 1973
Glutathione Reductase 1	*Gr1*	8	Nichols und Ruddle, 1975
Beta Chain Hemoglobin	*Hbb*	7	Popp, 1965
Isocitrate Dehydrogenase 1	*Idh1*	1	Henderson, 1965
Soluble Malic Enzyme	*Mod 1*	9	Shows und Ruddle, 1968
Mannose Phosphate Isomerase 1	*Mpi1*	9	Nichols et al., 1973
Neuraminidase 1	*Neu1*	17	Womack und Eicher, 1977
Peptidase 3	*Pep3*	1	Lewis und Truslove, 1969
Phosphoglucomutase 1 und 2	*Pgm1 und Pgm2*	5 und 4	Shows et al., 1969

Appendix 2

Biochemical markers used for authenticity control of isogenic rat strains:

Biochemical Marker	Gene	Chromo-some	Literature
Aconitase	*Acon1*	5	Adams et al., 1984
Acid phosphatase-2	*Acp2*	3	Bender et al., 1984
Aldehyde dehydrogenase-2	*Ahd2*	12	Adams et al., 1984
Aldehyde dehydrogenase-c-	*Aldh*	13	Cramer et al., 1986
Kidney alkaline phosphatase	*Akp1*	10	Adams et al., 1984
Plasma alkaline phosphatase	*Alp1*	6	Bender et al., 1984
Amylase-1	*Amy1*	2	Bender et al., 1985
Amylase-2	*Amy2*	2	Mizuno und Susuki et al., 1978
Catalase-1	*Cat*	3	Kendall, 1985
Esterase-1	*Es1*	19	Bender et al., 1984
Esterase-2	*Es2*	19	Bender et al., 1984
Esterase-3	*Es3*	19	Bender et al., 1984
Esterase-4	*Es4*	19	Bender et al., 1984
Esterase-6	*Es6*	8	Bender et al., 1982
Esterase-7	*Es7*	19	Bender et al., 1984
Esterase-10	*Es10*	19	Bender et al., 1984
Esterase-12	*Es12*	4	Bender et al., 1984
Esterase-13	*Es13*	10	Kendall, 1983
Esterase-14	*Es14*	19	Hedrich und von Deimling, 1987
Esterase-15	*Es15*	19	Hedrich et al., 1987
Esterase-16	*Es16*	19	Hedrich und von Deimling, 1987
Esterase-18	*Es18*	19	Kluge et al., 1990
Fumarate hydratase-1	*Fh1*	13	Adams et al., 1984
group-specific component/ Vitamine D binding protein	*Gc*	14	Bender et al., 1981
a-glycerophosphate dehydrogenase	*Gdc1*	8	Bender et al., 1984
Glyoxalase -1 regulatory	*Glo1r*	20	Bender et al., 1984
Glycolate oxidase	*Gox1*	3	Bender et al., 1984
Glutathion S-transferase	*Gst1*	8	Matsumoto und Gasser, 1983
Beta-hemoglobin	*Hbb*	1	Brdicka und Sulc, 1965
Leucine aryl-aminopeptidase	*Lap1*	1	Bender et al., 1984
Malate dehydrogease-like enzyme	*Mdl1*	3	Matsumoto et al., 1982
Major urinary protein	*Mup1*		van Zutphen et al., 1981
Peptidase-3	*Pep3*	13	Adams et al., 1984
Pepsinogen-1	*Pg1*	9	Cramer et al., 1981
Pepsinogen-2	*Pg2*	9	Hamada et al., 1987
Phosphogluconate dehydrogenase	*Pgd*	5	Bender et al., 1984
Pyruvate kinase-1	*Pk1*	2	Bender et al., 1984
Rat tear protein-1	*Rtp1*	5	Kondo et al., 1987

Rat tear protein-2	*Rtp2*	1	Kondo et al., 1987
Seminal vesicle protein 1	*Svp1*	3	Gasser, 1972
Seminal vesicle protein 2	*Svp2*	1	van Zutphen et al., 1981
Tamase-1	*Tam1*	1	Matsumoto et al., 1984

Appendix 3

Immunologic markers used by The Jackson Laboratory for authenticity control of isogenic mouse strains:

Immunologic Marker	Gene	Chromo-some	Literature
Major histocompatibility complex (MHC)	Klein et al, 1982	17	Shiroishi et al, 1981
Ea9 Hemagglutinine	*Ea9*	4	Fox und Black, 1987; Walker und Phillips-Quagliata, 1985
Complement Factor 5	*Hc*	2	Rosenberg und Tachibana, 1962; Herzenberg et al., 1963

Appendix 4

Immunologic markers used for authenticity control of isogenic rat strains:

Immunologischer Marker	Gen	Chromosom	Literatur
Major histocompatibility complex (MHC)	*RT1*	20	Bogden und Aptekman, 1960
Cell surface alloantigene, Ag-C	*RT2*	19	Kunz und Gill, 1978
Cell surface alloantigen Ag-D	*RT3*	13	Hedrich und Reetz, 1990
Cell surface alloantigen, ADP-Ribosyltransferase 2b,*RT6*	*Art2p*	1	Greiner et al., 1982
Protein tyrosine phosphatase, Receptor type c, *RT7*, *CD45*	*Ptprc*	13	Barclay et al., 1987