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InFocus

Center of Comparative Medicine & Pathology
Research Animal Resource Center
Laboratory of Comparative Pathology

Genetic Mutation in a Commercial Vendor's C57BL/6 Colony has the Potential to Invalidate Research Studies

The utilization of rodents, especially mice, as animal models has allowed scientists to explore and discover key aspects of many human and animal diseases. Mice are the most commonly used mammalian model due to their small size, high reproductive rate, low maintenance cost, and close genetic similarity to humans[13]. The mouse has become a powerful research tool due to our ability to manipulate its genome indirectly, by actively selecting for specific phenotypes, or directly, using genome editing tools (i.e., CRISPR, etc.)[16].

Some of the earliest studies conducted with mice involved analyzing the genetics of coat color within a population. It was through the work of Clarence Cook Little and his colleague in the early 1900's that inbred strains were developed in hopes of maintaining genetic homozygosity within a colony[12]. An inbred strain is created by conducting 20 or more brother-sister matings with all offspring descended from a single breeding pair[1,9]. Inbred strains are genetically uniform enabling the effects of their genetic background to be examined closely and reproducibly[5]. However, the often-forgotten cost of maintaining genetic uniformity within inbred strains has been its vulnerability to substrain divergence and genetic drift.

Substrains develop when distinct colonies of the same inbred strain become increasingly genetically divergent through generations of independent breeding[16]. More specifically, a colony is considered a substrain when more than 20 generations have passed since its separation from the parent colony and/or phenotypic differences from the parent colony are discovered[8]. They result from the incorporation and fixation of spontaneous mutations driven by genetic drift, and may give rise to additional substrains if the breeding colony doesn't share the same foundation colony[1].* Substrains are designated by specific nomenclature to help researchers identify their origin and

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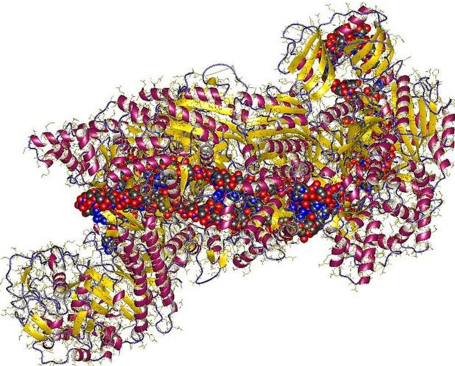

Clustered regularly interspaced short palindromic repeats, aka-CRISPR, one of several genetic engineering tools available for mouse manipulation.
Image credit: https://en.wikipedia.org/wiki/CRISPR

"A rose by any other name…", not so where mice are concerned. No Nicknames Please! "B6" just won't do anymore. The Jackson Laboratory provides the "Mouse Nomenclature Quick Guide" PDF, available for download at: https://www.jax.org/jax-mice-and-services/customer-support/technical-support/genetics-and-nomenclature

The Importance of Knowing What's Colonizing Your Mice

Confusion often exists regarding the health status of mice used in biomedical research. Investigators sometimes assume that mice are free of opportunistic pathogens or that health statuses are uniform within or across institutions, but this more often than not, is not the case. Mice, like humans, are each colonized with trillions of microorganisms, including bacteria, viruses, and fungi. Numerous studies have shown that mice from different vendors, institutions, or even locations within the same institution often host distinct microbial populations and that each mouse's microorganism profile can have significant effects on research. This article will discuss some of the recent findings and address how to be better informed regarding rodent colony health status.

In order to understand the implication differing health statuses, it is first important to define the terms used to categorize rodent colonies[1,2]. The strictest health status is "germ-free" or axenic, meaning that no exogenous* biological agents are present. These mice are in fact "sterile". In order to be axenic, animals generally must be housed within sterile isolators[#] and procedures are employed that prevent the introduction of microorganisms. Isolators are tested regularly to ensure they, and the animals contained within, remain sterile. Axenic rodents are a subset of rodents referred to as "gnotobiotic." This term is derived from the Greek gnotos meaning "known" and describes animals for which all exogenous organisms, if present, have been defined. Gnotobiotes are maintained in isolators to prevent colonization by other microorganisms. They are often used to study the effects of a single organism or set of organisms without the confounding effects of additional unknown microbes. However, for many studies, these mice prove to be poor research subjects, as their lack of commensal[+] microorganisms significantly alters gastrointestinal physiology and immunologic function and thus they are not representative of the majority of animals or humans. One particularly notable abnormality is that gnotobiotic rodents have severely enlarged ceca which are prone to torsion, resulting in

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Inside:

✓ New A Quality Assurance Specialist joins the IACUC

✓ What are the ARRIVE Guidelines? See page 3 for details.

Don't judge a book by its cover. Appearances are deceiving- gnotobiotic, defined flora or specific pathogen free mice are visually indistinguishable but highly individual!
Image credit: https://www.jax.org/news-and-insights/jax-blog/2013/may/the-difference-between-germ-free-and-specific-pathogen-free-mice

Genetic Mutation in a Commercial Vendor's Colony *Cont. from pg. 1*

history. Once a colony is determined to be a substrain, it is given a laboratory code that consists of one to five letters identifying the institute, laboratory, or investigator that produced and/or maintained a particular animal strain[14]. Laboratory codes are assigned by the Institute of Laboratory Animal Research (ILAR). Substrains can have distinct phenotypes. For example, the C3H/HeJ mouse is a substrain of the C3H inbred strain that originated and was maintained by an investigator, Walter Heston, prior to being maintained at the Jackson Laboratory (JAX)[5]. This particular substrain is resistant to the effects of lipopolysaccharide (LPS) due to a mutation in its Toll-like 4 receptor[5]. The C3H/HeOuJ substrain, on the other hand, is sensitive to LPS and contains a wild-type Toll-like 4 receptor[5]. This particular substrain was given to Henry Oustin by Walter Heston in 1952 prior to arriving and being maintained at JAX[5]. Although both the C3H/HeJ and C3H/HeOuJ substrains originated from the same parent colony in 1952, independent breeding by the two investigators led to a mutation within one of the colonies and a marked phenotypic difference that has been utilized in immunological research. To ensure reproducibility[#] it is important for investigators to specify the substrain used within the Materials and Method sections of publications due to the potential of phenotypic and genetic differences to develop in substrains. More information on reproducibility and the ARRIVE (Animal Research: Reporting of In Vivo Experiments) Guidelines can be found in this edition of In Focus[4,6].

Mice, like other living organisms, have an intrinsic genetic drive to change[5]. With each passing generation, spontaneous mutations can potentially develop from DNA repair processes or during meiosis[5]. Mutations can result from single base-pair changes, deletions, duplication, or inversions, and are the source of genetic variation found in all biological organisms[1]. It is important to understand and recognize that inbred strains are not exempt from this biological phenomenon. Genetic drift is the constant tendency of genes to evolve even in the absence of selective forces[5]. It is the development of spontaneous mutations that drives genetic drift and causes genetic variation to become fixed within a population at random[5]. Genetic drift can be beneficial within a population since it can contribute to species and phenotypic diversity. On the other hand, it can create unwanted mutations that have the potential to confound research, as recently described in a Cell Reports paper published by Mahajan and colleagues[7]. This paper describes multiple hematopoietic phenotypes that were thought to have resulted from altered sialic acid physiology within a transgenic line they created. Their transgenic mice had a germline loss of either Siae (sialic acid acetyl esterase) or Cmah (cytidine monophosphate-N-acetylneuraminic acid hydroylase) that initially was thought to cause them to lack marginal zone (MZ) B cells and exhibit hyperactive B cell receptor signaling[7]. The authors subsequently discovered that the phenotype was not a result of the genetic changes they induced but resulted from a spontaneous mutation that disrupted the function of Dock2 which was present in the C57BL/6NHsd (B6/NHsd) substrain they used for backcrossing. The authors demonstrated that the mutation was present in mice of the B6/NHsd substrain obtained from a single commercial vendor, but not in colonies of the same inbred strain (B6/N) maintained by other vendors[7]. The vendor Envigo (formerly known as Harlan) subsequently discovered that the mutation was inconsistently present in their B6/NHsd colonies within the US and across the globe raising concerns, within CCMP, of their genetic stability program.

The B6/N strain is widely used for research studies as well as backcrossing[7]. Mahajan et al speculated that the B6/NHsd substrain acquired this mutation either through genetic drift or genetic contamination from the 129 inbred strain at a remote time during their history[7]. However, Envigo subsequently found that only some of their B6/NHsd colonies were affected, suggesting this event occurred within the last 2 decades based on colony records. The principal difference between genetic drift and genetic contamination is that contamination is due to human error (i.e., accidental crossing with a different strain) and consequently can be prevented; however, drift is a natural, evolutionary process that can only be slowed (not stopped) within a population[5,14,20]. Mahajan et al's findings highlight the impact that substrain divergence can have on research when spontaneous mutations accumulate and go unnoticed.

Most commercial vendors have developed strategies to reduce the incidence of genetic drift within their production colonies. For example, JAX developed their Genetic Stability Program (GSP) to limit genetic drift by rebuilding foundation stocks from cryopreserved, pedigreed embryos every five generations (approximately every 18 months)[18]. This program was initiated in '03 and is used to manage multiple strains, including C57BL/6NJ, DBA/2J, FVB/NJ, and C57BL/6J.[18] JAX regularly

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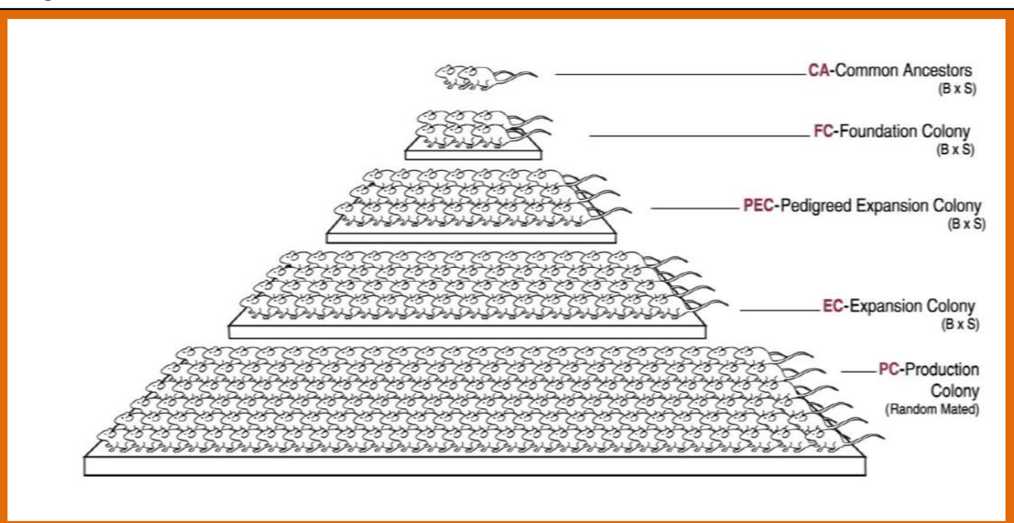
Figure 1: Pyramidal Breeding Scheme

At the top of the pyramid is the relatively small Foundation Colony (FC), shown here as being derived from a pair of genetically tested Common Ancestors. There is a single FC for each inbred strain. At the base of the pyramid is the large Production Colony (PC), and in between are the two types of expansion colonies, the Pedigreed Expansion Colony (PEC) and the Expansion Colony (EC). The FC is the only self-perpetuating colony (i.e., it produces its own new breeders), and all matings are brother x sister (BxS). The pedigreed expansion colony is an extension of the FC and receives all of its breeders from it; all matings in the PEC are also BxS. The PEC produces breeders for the EC. The EC produces breeders for the PC, where random mating is utilized to scale up breeding for production of animals used in research[15].



The ARRIVE Guidelines: What they are and why they are important

The ARRIVE (Animal Research: Reporting of In Vivo Experiments) Guidelines were established by the National Centre for the Replacement, Refinement, and Reduction of Animals in Research (NC3Rs) in '10 to improve the design, analysis, and reporting of research using animals to promote repeatability and minimize unnecessary experimental repetition[3]. The ARRIVE Guidelines state that scientific publications should include sufficient information to allow the reader to completely understand how the study was conducted and its biological relevance providing the reader the ability to assess the reliability and validity of the findings allowing the experiments to be repeated[1]. Ensuring that all relevant information is included in research publications is fundamental. Failure to carefully design experiments, clearly and sufficiently describe research methods, and to correctly interpret results has negative scientific and socio-economic implications[2]. For example, reporting animal numbers is necessary for the assessment of the biological and statistical significance of results and for the experiment to be repeatable. Furthermore, experiments using laboratory animals need to adhere to effective reporting guidelines to be ethically acceptable[3].

The ARRIVE Guidelines were developed in consultation with scientists, statisticians, journal editors, and organizations that fund research[3]. The guidelines consist of a checklist of 20 items (listed in the panel below) outlining the minimum information that should be included in publications describing research using animals (see below) or access a downloadable file at http://www.nc3rs.org.uk/arrive-guidelines. This includes the number and specific characteristics of the animals (e.g., species, strain, sex, and genetic background); details of housing and husbandry; and, the experimental, statistical, and analytical methods used (e.g., randomization and blinding). These guidelines are intended to: (1) improve reporting of scientific experiments using animals; (2) describe essential information to include in a manuscript while being flexible to be relevant to different research fields; (3) promote reproducible, transparent, accurate, comprehensive, concise manuscripts; and, (4) improve the communication of results to the broader research community. The guideline authors also specify that they are not intended to promote homogeneity, prevent originality, or be a guide for study design. They are appropriate for any biomedical research discipline in which laboratory animals are used. The guidelines have already been published in several bioscience research journals and publishers have endorsed these guidelines by incorporating them into the Instructions for Authors[3].

~ Christopher Cheleuitte, DVM, PhD

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	ITEM	RECOMMENDATION
Title	1	Provide as accurate and concise a description of the content of the article as possible.
Abstract	2	Provide an accurate summary of the background, research objectives, including details of the species or strain of animal used, key methods, principal findings and conclusions of the study.
INTRODUCTION		
Background	3	a. Include sufficient scientific background (including relevant references to previous work) to understand the motivation and context for the study, and explain the experimental approach and rationale. b. Explain how and why the animal species and model being used can address the scientific objectives and, where appropriate, the study's relevance to human biology.
Objectives	4	Clearly describe the primary and any secondary objectives of the study, or specific hypotheses being tested.
METHODS		
Ethical statement	5	Indicate the nature of the ethical review permissions, relevant licences (e.g. Animal [Scientific Procedures] Act 1986), and national or institutional guidelines for the care and use of animals, that cover the research.
Study design	6	For each experiment, give brief details of the study design including: a. The number of experimental and control groups. b. Any steps taken to minimise the effects of subjective bias when allocating animals to treatment (e.g. randomisation procedure) and when assessing results (e.g. if done, describe who was blinded and when). c. The experimental unit (e.g. a single animal, group or cage of animals). A time-line diagram or flow chart can be useful to illustrate how complex study designs were carried out.
Experimental procedures	7	For each experiment and each experimental group, including controls, provide precise details of all procedures carried out. For example: a. How (e.g. drug formulation and dose, site and route of administration, anaesthesia and analgesia used [including monitoring], surgical procedure, method of euthanasia). Provide details of any specialist equipment used, including supplier(s). b. When (e.g. time of day). c. Where (e.g. home cage, laboratory, water maze). d. Why (e.g. rationale for choice of specific anaesthetic, route of administration, drug dose used).
Experimental animals	8	a. Provide details of the animals used, including species, strain, sex, developmental stage (e.g. mean or median age plus age range) and weight (e.g. mean or median weight plus weight range). b. Provide further relevant information such as the source of animals, international strain nomenclature, genetic modification status (e.g. knock-out or transgenic), genotype, health/immune status, drug or test naïve, previous procedures, etc.
Housing and husbandry	9	Provide details of: a. Housing (type of facility e.g. specific pathogen free [SPF]; type of cage or housing; bedding material; number of cage companions; tank shape and material etc. for fish). b. Husbandry conditions (e.g. breeding programme, light/dark cycle, temperature, quality of water etc for fish, type of food, access to food and water, environmental enrichment). c. Welfare-related assessments and interventions that were carried out prior to, during, or after the experiment.
Sample size	10	a. Specify the total number of animals used in each experiment, and the number of animals in each experimental group. b. Explain how the number of animals was arrived at. Provide details of any sample size calculation used. c. Indicate the number of independent replications of each experiment, if relevant.
Allocating animals to experimental groups	11	a. Give full details of how animals were allocated to experimental groups, including randomisation or matching if done. b. Describe the order in which the animals in the different experimental groups were treated and assessed.
Experimental outcomes	12	Clearly define the primary and secondary experimental outcomes assessed (e.g. cell death, molecular markers, behavioural changes).
Statistical methods	13	a. Provide details of the statistical methods used for each analysis. b. Specify the unit of analysis for each dataset (e.g. single animal, group of animals, single neuron). c. Describe any methods used to assess whether the data met the assumptions of the statistical approach.
RESULTS		
Baseline data	14	For each experimental group, report relevant characteristics and health status of animals (e.g. weight, microbiological status, and drug or test naïve) prior to treatment or testing (this information can often be tabulated).
Numbers analysed	15	a. Report the number of animals in each group included in each analysis. Report absolute numbers (e.g. 10/20, not 50%[2]). b. If any animals or data were not included in the analysis, explain why.
Outcomes and estimation	16	Report the results for each analysis carried out, with a measure of precision (e.g. standard error or confidence interval).
Adverse events	17	a. Give details of all important adverse events in each experimental group. b. Describe any modifications to the experimental protocols made to reduce adverse events.
DISCUSSION		
Interpretation/ scientific implications	18	a. Interpret the results, taking into account the study objectives and hypotheses, current theory and other relevant studies in the literature. b. Comment on the study limitations including any potential sources of bias, any limitations of the animal model, and the imprecision associated with the results[2]. c. Describe any implications of your experimental methods or findings for the replacement, refinement or reduction (the 3Rs) of the use of animals in research.
Generalisability/ translation	19	Comment on whether, and how, the findings of this study are likely to translate to other species or systems, including any relevance to human biology.
Funding	20	List all funding sources (including grant number) and the role of the funder(s) in the study.

ARRIVE

The ARRIVE Guidelines: Animal Research: Reporting of *In Vivo* Experiments. Originally published in *PLOS Biology*, June 2010[1]

Genetic Mutation in a Commercial Vendor's Colony

Cont. from pg. 2

refreshes their foundation stocks from genetically defined offspring produced from frozen embryos[18]. By regularly refreshing mutant colonies by backcrossing to their GSP inbred strains, JAX ensures that the genetic background of these mutant colonies remain genetically similar to the parent inbred strain[18]. Another commonly used commercial vendor, Taconic, uses multiple strategies to ensure genetic quality across their sites including: (1) cryopreserving all inbred strains and stocks, (2) cryorecovering their foundation colonies every 5 years, (3) maintaining a single foundation colony for each inbred strain sourcing all production colonies from this single foundation colony, and (4) maintaining a maximum of 10 generations between their foundation and production colonies[15]. Envigo's genetic monitoring program, on the other hand, did not utilize cryopreservation or cryorecovery of their foundation colonies during the time when Mahajan et al discovered the spontaneous mutation in their B6/NHsd mice and they did not utilize a single foundation colony to support their colonies at distinct sites.

Genetic alterations can occur anywhere within the DNA, including non-coding and protein-coding regions[10]. It has been calculated that the spontaneous mutation rate in mice is approximately 1 per 26Mb per generation which equates to the introduction of approximately 100 SNPs per genome each generation.[10] Assuming we have a small inbred breeding population (which is common in vendors' foundation and investigators' colonies), we can estimate that 1 new mutation in a coding sequence will become fixed every 6-9 generations[5]. If fortunate, the mutation will contribute to a phenotypic change that can be easily detected within the population, such as a coat color change. However, there is always a possibility that the mutation will be "invisible" and result in no obvious phenotype, such as the loss-of-function variant of Snca that are seen in C57BL/6JOlaHsd[7]. Nevertheless, the moment a spontaneous mutation develops and becomes fixed within a mouse colony, it is imperative to know that this population is no longer genetically identical to its original parent colony, and is now a substrain[16,17]. By cryopreserving lines, investigators can "rescue" and rederive their transgenic lines that might have accumulated mutations over the years and are therefore, genetically different from their original strain.

Researchers can also play a part in limiting the impact of genetic drift within their breeding colonies. JAX encourages investigators to closely monitor their colonies for phenotypic change, such as coat color[5]. Phenotypic changes can indicate the presence of mutations and genetic drift. In addition, investigators should avoid applying selection pressure and randomize breeding[5]. By actively selecting animals that are larger or have a shinier coat, as examples, they can inadvertently fix mutations within their colonies. Breeders should be refreshed every 10 generations to ensure genetic background remains similar to the parental inbred strain[5].

The accumulation of impactful mutations within the genome is one of the contributing factors to the development of substrains. Although these mutations can be beneficial when they lead to a biologically important and identifiable phenotype potentially resulting in a new animal model, the unintentional changes induced by genetic drift can often have small, but significant impacts on a mouse strain and consequently, the research studies that utilize them[5]. Mahajan et al highlighted this concept by demonstrating their transgenic line's phenotype was erroneously attributed to an induced gene knockout, when in fact it was due to a spontaneous mutation found in the substrain used to backcross. The B6/NHsd substrain is used commonly in many research studies, including by some our own investigators. Scientists who use this substrain should be aware of the potential impact this mouse strain can have or has had on their experiments either directly when used as experimental subjects or indirectly when used for backcrossing. It is important for researchers to recognize that genetic drift exists within all inbred populations, and they should seek ways to mitigate its effects on their colonies and research.

~ Samantha Peneyra, DVM

References: (see panel in left margin)

Endnotes:

*Foundation colonies serve as the genetic and health standard for an inbred mouse strain. These animals are derived from a pair of genetically tested common ancestors and provide breeders through sibling mating for the expansion and production colonies of a pyramid mating system[15].

#Reproducibility is the ability to achieve similar or nearly identical results using comparable materials and methodologies[4].

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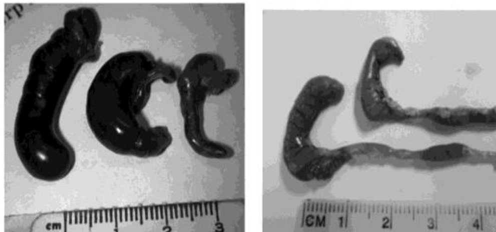
Enjoy the FALLing leaves!

Image credit: http://www.carlsams.com/inthewoods/ 2010/02 mouse-hide-and-seek-and- karens-mouse-keepers-journal/

The Importance of Knowing What's Colonizing Your Mice *Cont. from pg. 1*

Comparison of Cecal Morphology of GF, Monoassociated, ASF, & Conventional Mice

GF, Mono, and Conv Ceca — ASF and Conventional ceca



Morphological features of the ceca from gnotobiotic and conventional mice. Left panel: Representative images of the cecum excised from a germfree (left), a monoassociated (center), or a conventional (right) C3H/HeN mouse. Right panel: Representative images of the cecum excised from an ASF (left) or conventional (right) C3H/HeN mouse.

Source: http://ilarjournal.oxfordjournals.org/content/56/2/169.full

How much is that mouse in the window? Selective seeding of gut flora is favorable for many studies but can a mouse be too "clean"? Some studies may require an immunophenotype akin to that of humans and it has been shown that in some cases controlled exposure to pet store mice can achieve that goal.

Image credit: Image credit: http://www.everythingneon.com/proddetail.php?prod=n100-3474-pet-shop-neon-sign antenna/ratbrains/131.asp

Gnotobiotic, "defined flora" rodents (mice and rats) require strict husbandry practices via maintenance in isolators to prevent colonization by other microorganisms.
Image credit: https://thewalklab.files.wordpress.com/2015/ 04/walklab15.jpg

death of the animal. For these reasons, rodents maintained in isolators are sometimes inoculated with a cocktail of specific commensal bacteria, such as Altered Schaedler's Flora (ASF) used in mice, in order to establish a natural microbiome. Mice colonized with ASF are still considered gnotobiotic since all exogenous microorganisms are known. These mice are sometimes referred to as having a "defined flora".

Even with a defined flora, gnotobiotic animals are not always representative of animals living in natural environments. Furthermore, their maintenance is expensive and labor-intensive. Therefore, gnotobiotic conditions are generally reserved for production facilities and specialized studies, while most mice used in research are maintained outside isolators. Once animals are removed from isolators, they are colonized by organisms present in the environment and are no longer gnotobiotic. A similar process occurs during the perinatal period. After leaving the sterile environment of the uterus, the newborn immediately begins to acquire microbes from the birth canal and then from their mother while nursing and nesting. Even after this period, animals continue to acquire microbes from the environment and their conspecifics throughout life.

When rodents are maintained outside of an isolator, the primary focus becomes preventing colonization by agents that cause disease or can confound research. These animals are referred to as "specific pathogen free" or SPF. SPF animals are free of specific agents that are catalogued. Conditions for maintaining SPF rodents may include the use of cage filter tops, ventilated caging, gamma-irradiated feed, sterile bedding, various types of personal protective equipment (PPE), regular sanitation of housing areas and ensuring that the materials with which the animals come in contact or are administered do not carry agents for which the animals are intended to be free. SPF rodents are often maintained within "barriers" which implies a set of physical and operational measures designed to prevent the introduction of pathogens.

It is critical to note that SPF rodents are only free of certain agents for which they have been tested. No definitive statements can be made regarding the presence of organisms beyond the list of excluded agents. Furthermore, the agents excluded can differ among institutions, between vendors, or even between different housing areas within an institution. A recent study that investigated the effects of *Escherichia coli* on a mouse model of inflammatory bowel disease serves as example[3]. The authors found that when using dextran sulfate sodium to induce colitis, mice of the same strain obtained from a vendor showed significantly more muscle wasting than those maintained within their own vivarium. This difference was attributed to the presence of an *E. coli* strain that was present in their colony but not in those obtained from the vendor. The bacterium prevented muscle wasting by sustaining signaling in the IGF-1/PI3K/AKT pathway, which is an important regulator of muscle size. Although both colonies were SPF, neither specifically excluded *E. coli* which ultimately led to phenotypic differences in animals from each group.

Given this finding, it is tempting to adopt the philosophy of excluding as many organisms as possible to reduce variability between populations. However, Beura et al suggested that certain microorganisms may help produce a phenotype that is more representative of the human population[4]. They found that C57BL/6 mice housed under SPF conditions had a CD8+ T cell population closely resembling naïve neonatal humans while pet store mice had a T-cell population more akin to human adults. Furthermore, exposing laboratory mice to pet store mice and their infectious microbes restored the more representative phenotype. In this case, the presence of additional exogenous microorganisms created an immunophenotype that more closely modeled that of humans.

It is also interesting to note that physiology is not just changed by pathogenic organisms, but commensals as well. This was observed by Ivanov et al who investigated the effects of a common commensal bacterium[5]. This study showed that inoculation of mice with a single species of segmented filamentous bacteria (SFB) resulted in the presence of Th17 cells in the lamina propria leading to a proinflammatory state resulting in resistance to the intestinal pathogen, *Citrobacter rodentium*. Several additional studies revealed the importance of the microbiome as well as the diversity of

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the microbiome between different colonies. Surprising results have included discovering that the microbiome changes significantly after arrival at a new institution and that the microbiome can affect studies of obesity, inflammation, diabetes, and immunology[6]. But while the intestinal microbiome is crucial to several areas of interest, this information is not readily available, regardless of health status. In cases where the microbiome has a role, more extensive and specialized testing, such as metagenomics or targeted amplicon sequencing must be performed.

In conclusion, is important to consider the implication of health status on each study conducted in animals. It is also crucial to consider differing health statuses when comparing data between studies or across institutions. One obstacle is that excluded pathogens are not always included in publications, so it is important that this becomes standard practice moving forward. Investigators may contact the Research Animal Resource Center's (RARC) biosecurity staff for information regarding the health status and excluded pathogens for the areas in which their animals are housed.

~ Nick Tataryn, DVM

Metagenomics? When you absolutely, positively have to know gut microbiome…

Metagenomics is based on the genomic analysis of microbial DNA that is extracted directly from communities in environmental samples. This technology- genomics on a huge scale- enables a survey of the different microorganisms present in a specific environment such as water or soil, to be carried out.

No snacking allowed!

Footnotes:

*This does not take into account endogenous retroviral elements, which include retroviral genes that have been integrated into the genome and have the potential to produce infective viral particles. Such endogenous retroviruses are common in mice as well as humans and cannot be eliminated. In mice, these include genes encoded by the murine leukemia virus and mouse mammary tumor virus among others.

#Isolators are housing units that are designed to maintain animals while remaining impregnable to outside contaminants. There are typically two ports for supply and exhaust air, both of which pass through high-efficiency particulate arrestance (HEPA) filters. Another port is designed to pass sterilized materials into the isolator for maintenance of the animals. Animals only enter isolators if they have been reared in a sterile environment (another isolator) or if they are being rederived by sterile cesarean section or embryo transfer procedures.

+Commensal microorganisms are those that inhabit the host without causing any harm or benefit. These include viruses, bacteria, and fungi that show no pathology in healthy animals. Common sites harboring these organisms are the skin, digestive tract, and terminal urogenital system. Recent interest in this population has led to several studies that suggest these organisms in fact have positive effects on the host, which would reclassify them as mutualistic.

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Welcome IACUC Quality Assurance Specialist Allison V. Maurice

We are pleased to announce that Allison V. Maurice has recently joined the WCM, HSS and MSKCC IACUCs as a Quality Assurance Specialist. Allison comes to the IACUCs from SUNY Downstate, where she was the Training Coordinator and Compliance Officer. Allison has considerable experience in post approval monitoring, training and conducting in-life audits of animal based research. Allison will be working with CCMP's Education & Quality Assurance section in formalizing the current IACUC Post Approval Monitoring program. She can be reached at MauriceA@mskcc.org, alm20732med.cornell.edu, or (646) 888-2417.