Phenotyping Challenges in Genetically Engineered Mice

Abstract
Genetically engineered mice (GEM) are a staple of modern biomedical research. While genetic engineering technology has become more and more sophisticated, proper pathological analysis of these animals is often lacking. Proper pathological analysis is often replaced by Do-it-Yourself (DIY) pathology, which often resulted in surprising “scientific discoveries”.

Do-it- Yourself (DIY) Pathology
Conflict between pathology and genetic manipulation in the mouse
- Molecular biology is the driving force behind genetic manipulation in the mouse
- Scientists who create GEM often do not have training or knowledge on normative biology and disease processes within the context of a whole organism
- Adequate pathologic analysis is usually not performed
- One can publish a phenotypic analysis in a leading biomedical journal without input from a pathologist

The rise of Do-it-Yourself pathology
- Pathologists with rodent pathology training are few
- Pathological analysis is done by not adequately trained basic scientists

Consequences
- Several instances of erroneous interpretation
- The percentage of scientific reports on phenotypic analysis without input from a pathologists is actually decreasing (Figure 1.)
Figure 1. The decrease of the percentage of scientific reports on phenotypic analysis without proper pathological analysis

Scientific Consequences of DIY Pathology

Histopathological interpretation by “wallpaper matching”

- GEM lesions are often unique
- Lesion interpretation in GEM should be done by an experienced comparative pathologist
- Histopathology knowledge cannot be acquired without the proper background in medicine and participation in a rigorous post-graduate training program

Normal organs identified as lesions

- Preputial glands identified as teratomas
Figure 2. The image above taken from a publication in Cell where the authors identified the bilaterally symmetrical preputial glands (white arrows) as “Mature cystic teratoma of hyperkeratotic skin with sebaceous glands”.

Misinterpretation of inflammatory lesions as neoplastic processes
- Inflammatory lesions are sometimes misinterpreted as lymphoma

Figure 3. The image above taken from a publication where this Mycoplasma-induced mixed inflammatory infiltrate around a bronchiole in rats treated with methyl-tertiary-butyl-ether was interpreted as lymphoma

Misinterpretation of proliferative lesions as neoplastic processes
Figure 4. The image above taken from a publication where crypt herniation (white arrows) through the muscularis mucosae was interpreted as an example of adenocarcinoma. Despite their appearance, these lesions will disappear with antibiotic treatment.

**Solutions to DIY Pathology**

The immediate future – large scale mouse mutagenesis programs that will require adequate pathology analysis

- Knock Out Mouse Project (KOMP) – NIH
- North American Conditional Mouse Mutagenesis Program (NorCOMM) – Canada
- European Conditional Mouse Mutagenesis Program (EUCOMM) – Europe
- International Mouse Knockout Consortium
- “Complex Traits” Consortium

**Responsibilities of scientific journals**

- Editors should recruit comparative pathologists to review manuscripts on GEM
- Editors should require expert pathology interpretation in each GEM manuscript
- Journals should use appropriately-sized histologic images in publications (i.e., much larger than a postage stamp)

**Responsibilities of funding agencies**

- Study sections should include comparative pathologists to review GEM grant proposals

**Responsibilities of biomedical investigators** – follow published reports (see examples in References section)

- Mouse models of intestinal cancer
- Prostate pathology of GEM
- Proliferative pulmonary lesions
- Classification of non-lymphoid hematopoietic neoplasms

**Responsibilities of training institutions**
Current veterinary and human pathology training programs should focus on comparative pathology as well.
Training programs should expose their trainees to pathology of genetically engineered mice.
Encourage their interested trainees to attend appropriate workshops on mouse pathology.

References
Fu et al. The circadian gene Period2 plays an important role in tumor suppression and DNA damage response in vivo. Cell 111, 41-50, 2002

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