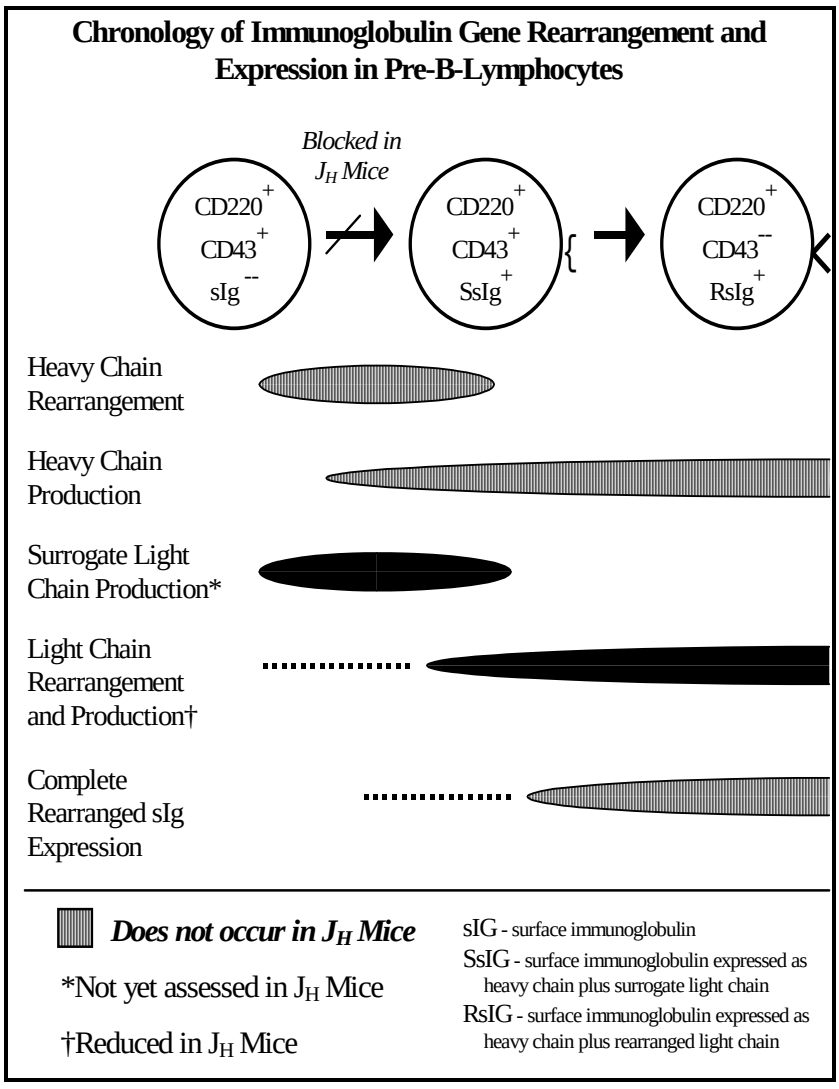
Jh Targeted Mutation Mice

Deletion of the J_H gene for antibody heavy chain production provides a mouse model devoid of mature B lymphocytes, that is suitable for exploration and manipulation of humoral and cell-mediated immune responses, both in healthy and disease states.

Applications for the Jh Targeted Mutation Mouse Model (model 001147)

- Assessing cellular control mechanisms for antibody gene assembly and expression
- Defining the temporal inter-relationships among heavy and light chain gene rearrangement, transcription, and translation
- Correlation of temporal changes in B-lymphocyte surface proteins with antibody gene rearrangement and activity
- Discovery of biochemical approaches to enhance immunity in B cell-deficient animals
- Characterization of non-B cell immune responses in pathogen-induced disease
- Providing a null background for gene replacement experimentation in normal immune responses and autoimmune disease
- Development of an antibody-dependent immune deficiency model



Features of the Jh Targeted Mutation Mouse[1]

- Deletion of all four J_h gene segments and the JhQ52 segment of the antibody heavy chain locus
- No rearrangement of $V_H(D)_HJ_H$ segments in the genome of B-lymphocyte precursor cells
- An accessible (transcribed) V_H locus in B-lymphocyte precursor cells
- No detectable IgM or IgG in serum
- Absence of mature (surface immunoglobulin) B-lymphocytes, bone marrow, and periphery
- Precursor B cells ($B220^+$ cells) reduced in bone marrow by two to four fold and represent the immature large cell fraction
- B cell differentiation blocked at the large, $CD43^+$ precursor stage
- Apparently normal development and surface phenotype of T cells
- Normal quantity of T cells in spleen (relative quantity enriched two to five fold due to reduced cells in the B-lymphocyte lineage)
- No Peyer's patches or M cells
- No follicular dendritic cells

Scientific Profile of the Jh Targeted Mutation Mouse Model

Immunoglobulin (antibody) synthesis by B-lymphocytes is an integral part of the immune defense system. The production of antibodies that together provide an astonishing diversity of antigen recognition sites is accomplished in part by somatic DNA recombination in developing B-lymphocytes. Specifically, loci encoding the variable regions of antibody light chains and heavy chains undergo recombination in early stages of B-lymphocyte differentiation. Each B cell becomes committed to expressing one of the many possible variable chain combinations for each of the light and heavy chains. In addition to ensuring that each B cell contributes a unique antigen-recognition site to the vast diversity generated by the B-lymphocyte population, these recombination events also are regulators of B-lymphocyte development that ensure maturation of fully functional B cells.

During recombination, regions of DNA that will compose the final encoding regions for the antibody light chain or heavy chain are spliced together in a prescribed order. The spliced (recombined) DNA assemblage for the variable region of the heavy chain is composed of one gene segment from each of three loci, J_H, $(D)_H$, and V_H. Prior to recombination these segments were widely separated on the chromosome. Similarly, the recombined light chain assemblage consists of distant gene segments (located on a different chromosome from the heavy chain loci), designated J_L and V_L. In both mouse and humans, each of these five loci in the embryonic genome includes anywhere from a few to a few hundred tandemly arranged genes. For example, four J_H gene segments are present on the mouse chromosome prior to recombination, but only one is incorporated into the final heavy chain encoding sequence after recombination.

Current evidence supports the concept that somatic recombination proceeds in each pre-B cell in a specific temporal pattern. Through investigation using murine models, it appears that the recombination of the heavy chain components occurs first, beginning with linkage of a $(D)_H$ gene segment to a J_H segment. Subsequent attachment of the V_H segment to the $(D)_H$ portion of the $(D)_HJ_H$ segment generates the linear DNA region from which the $V_H(D)_HJ_H$ (heavy chain variable region) transcript is derived. Light chain recombination, in which V_L and J_L segments become linked to produce a single, V_LJ_L light-chain DNA assemblage, appears to follow assembly and expression of the recombined heavy chain. However, evidence, including from the Jh Targeted Mutation Mouse, exists to suggest that some degree of light chain rearrangement can precede heavy chain rearrangement and expression.

The Jh Targeted Mutation Mouse Model carries a targeted deletion of the J_H locus, such that mice are homozygous for absence of all four J_H gene segments.[1] As a result, cells cannot produce a complete, recombined version of the variable region of the heavy chain. Analyses of cells from the B-lymphocyte lineage ($B220^+$ cells) revealed that there are no rearranged $V_H(D)_H$ or $V_H(D)_HJ_H$ segments. Thus, it appears that loss of J_H genes prevents not only the assembly of the complete heavy chain variable region, but prevents linkage of the V_H and $(D)_H$ regions, in spite of the ability of the cells to transcribe the V_H gene alone.[1] These data confirm the need for linkage of $(D)_H$ and J_H segments prior to the addition of the V_H segment.

Jh Targeted Mutation Mice have no detectable IgM or IgG in serum.[1] This implies a complete interruption of functional antibody formation in the absence of J_H genes. However, a low level (about 1% of normal) of rearrangement of the light chain kappa (κ) gene family is detected in total bone marrow.[1] Cell sorting of bone marrow samples from Jh Targeted Mutation Mice revealed that this light chain recombination is attributable to a subset of $CD220^+$ cells (B cell lineage) that are large, immature, and carry the surface protein $CD43^+$. About 2.5% of these $CD220^+/CD43^+$ cells demonstrated reassembled light chains, and at a level equivalent to about half that achieved by the same cell population in wild-type mice.[1]

Cells of the B lineage in Jh Targeted Mutation Mice are drastically altered by the absence of the J_H genes, both in their developmental progression and in cell quantity.[1] Cells beyond the $CD220^+/CD43^+$ stage (such as smaller, $CD43^-$ cells) are not seen in bone marrow, and there are no mature (immunoglobulin-bearing) B-lymphocytes in spleen, bone marrow, lymph nodes, peripheral blood, or peritoneum. Furthermore, total quantities of B-lineage cells are reduced in bone marrow compared to wild-type mice by two to four fold. However, T-lymphocyte development appears to proceed normally in Jh Targeted Mutation Mice, as evidenced by a normal surface phenotype and absolute quantity of cells in the spleen. Splenic lymphocytes are enriched (from two to five times normal) for T cells due to the B cell deficit.

The Jh Targeted Mutation Mouse Model provides a null background for the antibody heavy chain J_H gene segment, which makes it suitable for experimentation in mechanisms of B cell differentiation and antibody production. Transgenic rescue of antibody synthesis by adding specific murine genes to this background can aid in defining immune mechanisms in normal and disease states. For example, a mouse line has been developed through intercrossing of Jh Targeted Mutation Mice and another line with severe autoimmune manifestations.[2] Animals from the resultant cross lacked mature B cells and antibodies, had no signs of autoimmune tissue destruction and a reduced T cell activity. This same intercrossed strain was further bred with mice carrying a mutant transgene for a reassembled heavy chain (IgM) that would appear on cell surfaces but not be secreted. This model has provided insight into the essential role of surface immunoglobulins on B-lymphocytes in autoimmune disease expression.[3] Additionally, transgenes encoding portions of the heavy chain locus derived from humans were able to restore some aspects of humoral immune function when bred into the Jh Targeted Mutation Mouse Model.[4]

Origin of the Model

The Jh Targeted Mutation Mouse Model was developed by Dennis Huszar and colleagues at GenPharm International. Elimination of the J_H gene region, which normally carries four J_H loci, was accomplished by electroporation of cultured AB-1 embryonic stem cells derived from strain 129 Sv/Ev Mice, with a targeting vector in which J_H genes were replaced with a *neo* expression cassette.[1] Cells that successfully incorporated the construct were identified and injected into C57BL/6J blastocysts to generate chimeric mice. Male chimeras then were bred with C57BL/6J females. Heterozygous offspring expressing the targeted heavy chain locus were interbred to generate mice that were homozygous for the J_H deletion, as identified by Southern blot hybridization of tail DNA.

The Jh Targeted Mutation Mice were transferred from GenPharm to Yale University in 1992 on a mixed 129 x B6 background. At Yale the mice were backcrossed several generations to C.B-Igh1*b* congenic mice and then backcrossed to BALB/c mice for two generations before being bred to homozygosity. Thus, the mice are approximately 99% BALB/c in origin. The foundation colony is maintained through homozygous brother x sister matings in a plastic isolator. The production is maintained in a Murine Pathogen Free (MPF™) Isolated Barrier Unit.

Taconic provides Jh Targeted Mutation Mice in collaboration with Medarex, Inc., which has offered to make this promising model available to the scientific research community. Under agreement with Medarex, Taconic will provide Jh Mice to researchers with the stipulation that *cross-breeding of Jh Targeted Mutation Mice prohibits breeding the Mice with a mouse of another strain containing human Ig genes.* Any crossbreeding with the Jh Mice is subject to these terms, and an agreement to this effect is to be made by researchers who wish to crossbreed the Jh Targeted Mutation Mouse Model.

Ready for Your Experiments

Taconic Transgenic Models are produced and maintained in Isolator Barrier Unit (IBU™) facilities. Mice are shipped in Taconic Transport Cages (TTC™) and come with an up-to-date health report documenting their Murine Pathogen Free (MPF™) health status. Barrier housing conditions are recommended for maintenance of Jh Targeted Mutation homozygous mice.

Related Mouse Models from Taconic

Taconic provides a number of mouse models relevant to immune system function, including mutational, knockout, and microinjected models on a variety of backgrounds. Call or fax to inquire about the following additional models:

- **nude (models B6NU, B6NBO, BALBNU, BLBANU, NCRNU, NSWNU and NMRINU) –** heterozygous or homozygous for the nude gene, conferring in homozygous mice a T-lymphocyte deficiency due to absence of a functional thymus, but no altered T-cell immunity in carrier (heterozygous) mice
- **scid (models CB17SCRF, CB17SC, ICRSC and NODSC) –** homozygous for the *scid* (severe combined immunodeficiency) gene, lacking both T- and B-lymphocytes, serves as a model for immunity research and a host for xenotransplanation studies
- **scid-beige (model CBSCBG)** – homozygous for the *scid* (severe combined immunodeficiency) gene and *beige* mutation, lacking both T- and B-lymphocytes and exhibiting impaired macrophage and NK cell function; serves as a model for immunity research and a host in xenotransplantation studies
- **beige-nude-xid (model NIHBNX)** – homozygous for *nude* and *beige* mutations, and carrying an X-linked *xid* mutation; lacking normal thymic development and T-lymphocytes, exhibiting impaired function of macrophages, NK cells, and B-lymphocytes; serves as a model for immunity research and a host in xenotransplantation studies
- **Abb Targeted Mutation (models 004026 and ABBN12)** – exhibiting depletion of $CD4^+$ T-lymphocytes and deficiency in MHC Class II proteins due to disruption of the *Abb* gene, useful in transplantation, gene therapy, and immunological disease research
- **B2m Targeted Mutation (models 004020 and B2MN12)** – depleted of $CD8^+$ T-lymphocytes due to disruption of the *B2m* gene; useful in transplantation, gene therapy, and immunological disease research
- **Abb/B2m Double Targeted Mutation (model 004080)** – lacking MHC Class II and reduced in MHC Class I expression, deficient in both $CD4^+$ and $CD8^+$ T-lymphocytes
- **Fcer1g (FcRγ) Targeted Mutation (models 000584 and 000583)** – exhibiting impaired function of macrophages, neutrophils, mast cells, basophils, and NK cells due to lack of the gene encoding the γ subunit of the cell surface receptor proteins, FcγRIII and FcεRIγ
- **Fcgr2b (FcγRII) Targeted Mutation (models 000579 and 000580)** – exhibiting dysfunctional immune inhibitory pathways due to lack of the gene encoding FcγRIIβ, a low affinity IgG receptor
- **Pfp Targeted Mutation (model PFPN12)** – exhibiting a deficiency in perforin, a protein essential for cytotoxic activities of NK cells; useful for studies of immune suppression and transplantation
- **Rag2 Targeted Mutation (models 000461, 000601, RAG2 and RAGN12)** – lacking mature B- and T-lymphocytes, due to inactivation of the *Rag2* (recombination activating 2) gene required for V(D)J rearrangement; useful for vaccine development, transplantation studies, and hematopoiesis research
- **Pfp/Rag2 Double Targeted Mutation (model 001177)** – lacking both *Pfp* and *Rag2* genes, exhibiting a severe depletion of functional NK cells and of B- and T-lymphocytes; useful for vaccine development, transplantation studies, and studies of the immune system
- **Rag2 Targeted Mutation-HY Microinjected Mouse (model 004079)** – lacking endogenously derived mature B- and T-lymphocytes but with expression of receptors for the HY antigen, which rescues $CD8^+$ cell development in females

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