Purkinje Cell Degeneration 7 Jackson, a remutation of the *Agtpbp1* gene

Richard M. Samples, Patricia F. Ward-Bailey, Leah Rae Donahue, Roderick T. Bronson, and Muriel T. Davisson

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Mutation (allele) symbol: *Agtpbp1*$^{pcd\text{-}7J}$/J

Mutation (allele) name: Purkinje Cell Degeneration 7 Jackson

Gene symbol: *Agtpbp1*

Strain of origin: C.129S2(B)- *Igh-6*^{tm1Cgn}/J

Current strain name: C(Cg)-*Agtpbp1*$^{pcd\text{-}7J}$/J

Stock #005501 (NOTE- As of 2-1-2007 is available only as DNA from the Jackson Laboratory DNA Resource)

Phenotype categories: Neurological

Origin and Description

The *Agtpbp1*$^{pcd\text{-}7J}$/J mutation was discovered by Andrea Snyder in a production colony of C.129S2 *Igh-6*^{tm1Cgn}/J mice at the Jackson Laboratory in July 2003. Mice homozygous for this spontaneous, recessive mutation are recognizable by a moderate ataxia that begins at 3-4 weeks of age and becomes more intensified by 5 weeks of age. In addition to the ataxia, mutants present the severe deficiency of Purkinje cells characteristic of other Agtpbp1 alleles and live through adulthood.

Genetic Analysis

In order to determine the mode of inheritance, ovaries from a female homozygous for this new mutation were transplanted into C3SnSmn.CB17-*Prkdc*^{scid}/J hosts which were then mated to an unrelated C57BL/6J male. No affected offspring were observed in the F1 generation produced from this mating (0 affected/25 born). Mice from this F1 generation were then mated together to produce F2s, and from this intercross both affected and unaffected animals were produced, showing that the mutation is recessive.

A direct test for allelism was set up by mating a heterozygous male from the BALB/cByJ-*Agtpbp1*$^{pcd\text{-}3J}$/Jstock with 2 heterozygous females carrying this new mutation. This mating produced 2 litters in which 1 offspring was affected (1/11 animals born), proving the new mutation to be an allele of the *Agtpbp1* gene. The chromosomal position of the *Agtpbp1* gene is at the 37 cM position on Chromosome 13 (MGD 2005).

A concurrent linkage cross to CAST/Ei substantiated the positive test for allelism. Using tail tip DNAs from 21 mutant F2 progeny and our standard PCR protocols, we found no recombinants with *D13Mit157,* which is located at 36 cM on Chromosome 13.

Pathology

A routine pathological assessment[1] done on one homozygous and one heterozygous mouse at 9 weeks of age showed the Purkinje cell loss typical of *Agtpbp1* mutants and the hippocampus appeared enlarged. The corpus callosum was absent in both mice, but this is a strain characteristic Of BALB/cJ mice and not caused by the mutation.

The results of auditory-evoked brainstem response[2] tests showed that two 5-week-old homozygous mutant mice and a heterozygous littermate all had normal hearing thresholds, but the response latency was longer for the homozygous mutants than for the heterozygote. The eyes of 2 homozygous and 1 heterozygous mutant were examined by an ophthalmoscope and found to be normal.

Discussion

We discovered and characterized a new remutation of the *Agtpbp1* gene (*pcd-7J*). This remutation exhibits a similar clinical and histopathological phenotype to the original *pcd* mutation. A direct test for allelism with *pcd-3J* and the new mutation proved to be allelic.

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[1] Standard Histology Protocol used in the Mouse Mutant Resource

For fixation of tissues, mice were deeply anesthetized with tribromoethanol (avertin) until they no longer displayed a withdrawal reflex in the hind limbs and then perfused intracardially with Bouin's fixative following a flush of the vasculature with saline solution. After soaking in Bouin's for one week to demineralize bones, tissues were dissected. Six segments of spine with axial muscles and spinal cord in situ, representing cervical, thoracic and lumbar spinal segments, were dissected. The brain was removed and sliced into 6 cross sectional pieces at the levels of olfactory lobes, frontal cortex, striatum, thalamus, midbrain, rostral and caudal medulla with cerebellum. Midsagittal slices of hind leg through the knees were prepared. Slices of basal skull through the pituitary and inner ears were taken. Both eyes, salivary glands and submandibular lymph node, trachea plus thyroid and sometimes parathyroid were removed and cassetted. A longitudinal slice of skin from the back was removed. The thymus, slices of lung, and a longitudinal slice of heart were cassetted. Similarly slices of liver through gall bladder, kidney with adrenal attached, pancreas and spleen were prepared. The stomach was sliced longitudinally to include both squamous and glandular portions. Loops of small intestine from 3 levels and slices of large intestine and cecum were removed, as were slices of urinary bladder. The whole uterus, with ovaries attached, was taken. In males testes were sliced longitudinally. The accessory male organs including seminal vesicles, coagulating gland and prostate were removed en block. Altogether in most cases all tissue fit into a total of 10 cassettes. The cassettes were processed in an automatic tissue processor to dehydrate tissues which were then embedded in paraffin. Six micron sections were cut and stained with hematoxylin and eosin (H&E). Sections of brain and spinal cord in vertebral bones also were stained with luxol fast blue (LFB) for myelin and cresylecht violet (CV) for cellular detail.

[2] Auditory-Evoked Brainstem Response (ABR) Thresholds

ABR thresholds in mice are determined using a semi-automated computer system (Intelligent Hearing Systems, Miami, Florida). Subdermal needle electrodes are inserted at the vertex and ventrolaterally to both ears of anesthetized mice. Specific auditory stimuli from 10-100 dB SPL are delivered binaurally through plastic tubes from high frequency transducers. ABR thresholds are obtained, in an acoustic chamber, for clicks and for 8, 16, and 32 kHz pure-tone pips. ABR thresholds of all mice and strains tested are entered in spreadsheet files for storage, easy access, and for the production of periodic progress reports. Click-evoked

ABR waveforms, obtained at threshold (T) and at T+10, T+20 and T+30 dB or each mouse, are also stored for future reference. Mice of the CBA/CaJ strain are tested periodically as references for normal hearing, and for monitoring the reliability of the equipment and testing procedures.