Localization of the Synapsin II (SYN2) Gene to Human Chromosome 3 and Mouse Chromosome 6

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The synapsins are a family of four synaptic vesicle-associated proteins, synapsins Ia, Ib, IIa, and IIb, that have been implicated in modulation of neurotransmitter release and in synaptogenesis (for review see Ref. 4). They are products from alternative splicing of two distinct genes, the synapsin I and synapsin II genes. The synapsin I (SYN1) gene has been mapped to the X chromosome in human and mouse (10). In this study, we have determined the chromosomal location of the synapsin II (SYN2) gene in both human and mouse.

The human SYN2 locus was mapped by Southern blot analysis of DNA from a panel of 19 human–hamster and 1 human–mouse somatic cell hybrids (BIOS Co., New Haven, CT). Hybridization of *Eco*RI-digested genomic DNA from these somatic cell hybrids and controls with a 256-bp rat synapsin II cDNA probe (1) detected a single 9.5-kb hamster and a 4.5-kb mouse fragment and two human fragments of 6.8 and 5.6 kb (data not shown). Further analysis using a human synapsin II cDNA probe (9) indicated that only the 5.6-kb fragment is the human synapsin II-specific band. This 5.6-kb human *Eco*RI fragment was present in only two hybrid cell lines, 423 and 1079, both of which contain human chromosome 3. In contrast, the 9.5-kb hamster *Eco*RI fragment was present in all 19 human–hamster hybrids, and the 4.5-kb mouse *Eco*RI fragment was present only in the human–mouse hybrid cell line 016 (data not shown).

This mapping result was confirmed by reprobing of the same blots with a human synapsin II genomic probe obtained by PCR from the human synapsin II cDNA sequence (9). This probe hybridized with only a single 5.6-kb *Eco*RI human fragment that was detected only in hybrid cell lines 423 and 1079 (data not shown). Comparison of these results with the human chromosome content chart for each hybrid cell line indicates that the human synapsin II-specific fragment segregates with chromosome 3 with perfect concordancy. All other chromosomes are excluded as possible locations for SYN2 by at least two discordant hybrids (data not shown).

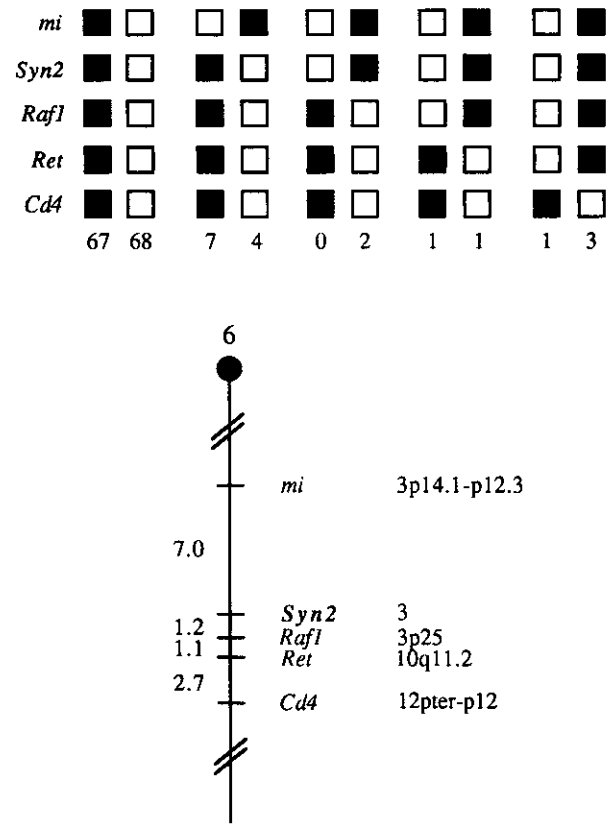

FIG. 1. *Syn2* maps to the central region of mouse chromosome 6. *Syn2* was placed on mouse chromosome 6 by interspecific backcross analysis. The segregation patterns of *Syn2* and flanking genes in 154 backcross animals that were typed for all loci are shown at the top of the figure. For individual pairs of loci, more than 154 animals were typed (see text). Each column represents the chromosome identified in the backcross progeny that was inherited from the (C57BL/6J × *M. spretus*)F_1 parent. The black boxes represent the presence of a C57BL/6J allele, and the white boxes represent the presence of a *M. spretus* allele. The number of offspring inheriting each type of chromosome is listed at the bottom of each column. A partial chromosome 6 linkage map showing the location of *Syn2* in relation to linked genes is shown at the bottom of the figure. A description of the probes and RFLPs for the loci linked to *Syn2* including microphthalmia (*mi*), ras-related fibrosarcoma oncogene (*Raf1*), ret protooncogene (*Ret*), and CD4 antigen (*Cd4*) has been reported previously (5, 6). Recombination distances were calculated as described (3) using the computer program SPRETUS MADNESS. Gene order was determined by minimizing the number of recombination events required to explain the allele distribution patterns. Recombination distances between loci in centimorgans are shown to the left of the chromosome, and the positions of loci in human chromosomes, where known, are shown to the right. References for the human map positions of loci cited in this study can be obtained from GDB (Genome Data Base), a computerized database of human linkage information maintained by The William H. Welch Medical Library of The Johns Hopkins University (Baltimore, MD).

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The mouse chromosomal location of *Syn2* was determined by interspecific backcross analysis using progeny derived from matings of [(C57BL/6J × *Mus spretus*)F_1 × C57BL/6J] mice. This interspecific backcross mapping panel has been typed for over 1800 loci that are well distributed among all of the autosomes as well as the X chromosome (2). C57BL/6J and *M. spretus* DNAs were digested with several restriction enzymes and analyzed by Southern blot hybridization for informative restriction fragment length polymorphisms (RFLPs) using a 1.5-kb mouse genomic *Syn2* DNA probe (1). This probe detected a 5.8-kb *Pvu*II fragment in C57BL/6J DNA and a major 1.7-kb *Pvu*II fragment in *M. spretus* DNA. The 1.7-kb *Pvu*II *M. spretus*-specific RFLP was used to follow the segregation of the *Syn2* locus in backcross mice. The mapping results indicated that *Syn2* is located in the central region of mouse chromosome 6 linked to *mi, Raf1, Ret,* and *Cd4*. Although 154 mice were analyzed for every marker and are shown in the segregation analysis (Fig. 1), up to 188 mice were typed for some pairs of markers. Each locus was analyzed in pairwise combinations for recombination frequencies using the additional data. The ratios of the total number of mice exhibiting recombinant chromosomes to the total number of mice analyzed for each pair of loci and the most likely gene order are centromere–*mi*–12/171–*Syn2*–2/162–*Raf1*–2/178–*Ret*–5/188–*Cd4*. The recombination frequencies [expressed as genetic distances in centimorgans (cM) ± the standard error] are *mi*–7.0 ± 2.0–*Syn2*–1.2 ± 0.9–*Raf1*–1.1 ± 0.8–*Ret*–2.7 ± 1.2–*Cd4*.

We have compared our interspecific map of chromosome 6 with a composite mouse linkage map that reports the map locations of many uncloned mouse mutations (Mouse Genome Data Base, a computerized database maintained at The Jackson Laboratory, Bar Harbor, ME). *Syn2* mapped to a region of the composite map that contains two mouse mutations, deaf waddler (*dfw*) and opisthotonos (*opt*), with a phenotype that might be expected for an alteration in this locus. The *dfw* mutation has been mapped 6.8 ± 1.1 cM distal of *mi,* and mice homozygous for *dfw* are deaf, bob their heads, and walk with a hesitant and wobbly gait (8). Homozygous *opt* mice can be recognized at about 10 days of age by their loss of balance when standing or moving. These mice also display severe opisthotonos (arching upward of the head and tail) and usually die before or by the weaning age (7). The *opt* mutation has been mapped 5.6 ± 1.1 cM from *mi* (7). It would be of interest to determine whether *Syn2* is altered in either of these mutations.

Finally, the central region of mouse chromosome 6 shares regions of homology with human chromosomes 3p, 10q, and 12p (summarized in Fig. 1). *Syn2* maps between *mi* and *Raf1,* both of which have been assigned to human 3p, suggesting that SYN2 will reside on the short arm of human chromosome 3.

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