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Assignment[a] of WNT7B to human chromosome band 22q13 by in situ hybridization

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Rationale and significance

Wnt genes constitute a growing family of structurally related glycoproteins with oncogenic potential, which are normally involved in the early embryonic development of a variety of species including mammals, insects and amphibians. The mammalian *Wnt* gene family consists of at least 16 members, which have distinct temporal and spatial expression patterns during embryogenesis. For example, murine *Wnt7a* is expressed in the flanking ectoderm of the trunk prior to limb bud outgrowth and throughout the dorsal ectoderm during growth and patterning of the early limb (Parr et al., 1993; Parr and McMahon, 1995). *Wnt7a* has a high transforming potential and has been implicated in mammary tumorigenesis (Wong et al., 1994). *Wnt7a*$^{-/-}$ mice exhibit limb defects in accordance with the expression pattern of the gene, and the recent localization of the human orthologue at 3p25 should guide the systematic search for mutations in this gene in human disorders (Parr and McMahon, 1995; Ikegawa et al., 1996). The closely related *Wnt7b* gene is expressed in specific regions of the embryonic forebrain, the collecting duct epithelium of the kidney and, rather uniformly, throughout the limb ectoderm (Parr et al., 1993; Kispert et al., 1996). We have isolated part of the human WNT7B gene and mapped this gene to chromosome 22q13.3. This chromosome band has been implicated to carry a third locus on chromosome 22 that is involved in meningiomas (OMIM 156100; Arinami et al., 1986).

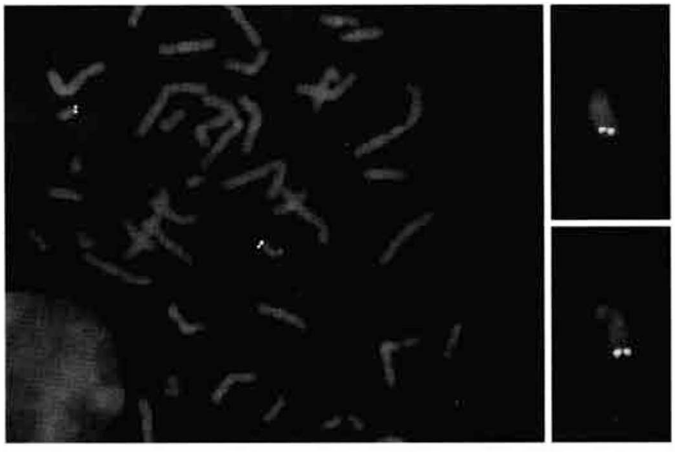

Fig. 1. Chromosome mapping of human WNT7B by fluorescence in situ hybridization. A double signal is present on both chromosome 22 homologs at a location corresponding to bands q13.2→q13.3. The position of the signals can be appreciated better at the enlarged images and the DAPI stained chromosomes 22.

Materials and methods

Isolation of human WNT7B clones

Cloning of human WNT7B cDNA sequences has been reported in Huguet et al. (1994). This clone was used to hybridize a Southern blot containing *Hin*dIII-digested DNA from human-rodent monochromosomal hybrids. A specific hybridizing band of approximately 20 kb was detected both in human control DNA and in the chromosome 22 hybrid (data not shown). Next, the same cDNA probe was used to screen the Lawrence Livermore chromosome 22 cosmid library to obtain a probe suitable for in situ hybridization. Four positive cosmids were obtained that were reactive with the WNT7B probe at a *Hin*dIII fragment of approximately 20 kb. The presence of WNT7B sequences in these cosmids was verified by sequencing.

Fluorescence in situ hybridization (FISH)

Metaphase spreads from lymphocytes were prepared using standard procedures and FISH was performed as described previously (Suijkerbuijk et al., 1991). Cosmid DNA was labeled with digoxigenin (Boehringer) and immu-

[a] To our knowledge this is the first time this gene has been mapped.

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nocytochemical detection was achieved with sheep-antiDIG-FITC, followed by successive steps with rabbit-antiFITC and goat-anti-rabbit-FITC. Chromosomes were stained with 4',6-diamidino-2-phenylindole-dihydrochloride (DAPI).

Probe names: clones 53G2, 62A9, 85D12 and 123F9 from library LL22NC03
Probe type: cosmid
Insert size: 30–40 kb
Vector: Lawrist16
Proof of authenticity: DNA sequencing
Gene reference: Huguet et al. (1994)

Results

Mapping data
Location: 22q13.2→q13.3
Number of cells examined: 35
Number of cells with specific signal: 29
1 (0), 2 (6), 3 (2), 4 (21) chromatids per cell
Most precise assignment: 22q13.3
Location of background signals (sites with >2 signals): none observed

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