# Assignment<sup>1</sup> of the human $\alpha$ 1,3-fucosyltransferase IX gene (FUT9) to chromosome band 6q16 by in situ hybridization

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## Rationale and significance

Lewis x (Lex) is defined as the terminal structure of carbohydrate chains consisting of the trisaccharide structure, Galβ1-4(Fucα1-3)GlcNAc, and is considered to play important roles in cell-cell interactions during embryonic development, differentiation, and oncogenesis. We recently isolated a novel human α1,3-fucosyltransferase gene (FUT9, alias hFucTIX), which is involved in the last step of Lex synthesis (Kaneko et al., 1999). The amino acid sequences of FUT9 are very highly conserved between human and the mouse homologue (Kudo et al., 1998; Kaneko et al., 1999). This high conservation is not the case for other fucosyltransferases which were previously cloned. FUT9 has been under a strong selective pressure during its evolution, suggesting that it plays essential roles in ontogeny. We report here the chromosomal mapping of FUT9.

## Materials and methods

Isolation of a PAC clone containing FUT9

A PAC genomic library (RPCl 4 and 5 libraries) was screened to obtain a genomic DNA clone using a pair of PCR primers (forward primer, 5'-AGGC-CACCCTTTCAGAAATG-3' and reverse primer, 5'-AGTTTTCCCTA-GATGGACCC-3') which amplify a 456-bp fragment from FUT9 cDNA. The reaction conditions were as follows: 30 cycles of 30 s at 96°C, 30 s at 55°C and 30 s at 72°C, by an automated PCR thermal cycler (Astec Co. Ltd.). We obtained a PAC clone (PAC-FUT9) encoding the FUT9 gene. The insert size of PAC-FUT9 was determined by pulse-field gel electrophoresis to be approximately 130 kb.

Chromosome mapping of FUT9 by FISH

FISH analysis and counterstaining with DAPI were carried out (Takahashi et al., 1990). Normal human lymphocyte metaphases were used. The probe, PAC-FUT9 was labeled by nick translation with DIG-11-dUTP using the Nick Translation Kit (Bochringer Mannheim).

Probe name: PAC-FUT9 Probe type: genomic DNA Insert size: 130 kb Vector: pCYPAC2

Proof of authenticity: Southern blot analysis and DNA sequencing Gene reference: Kaneko et al. (1999); GenBank accession no. AB023021

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<sup>&</sup>lt;sup>1</sup> To our knowledge this gene has not previously been mapped.

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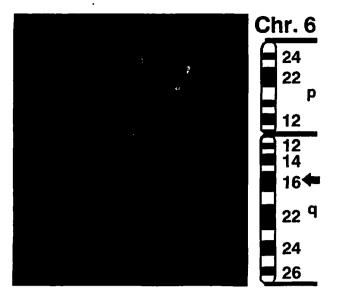


Fig. 1. FISH analysis demonstrated that FUT9 was located at 6q16, the long arm of human chromosome 6. An arrow indicates the position of fluorescent signals. Thus, the FUT9 gene was mapped to a chromosome distinct from those where the other members of the  $\alpha$ 1,3 Fuc-T family are found.

# Results

Mapping data Location: 6q16

Number of cells examined: 100

Number of cells with specific signal: 99

Most precise assignment: 6q16

Location of background signals (site with >2 signals): none

observed

### References

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