Fine Mapping of the Human Bone Morphogenetic Protein-4 Gene (BMP4) to Chromosome 14q22–q23 by *in Situ* Hybridization

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Bone morphogenetic protein-4 (BMP-4) is a member of the transforming growth factor-β (TGF-β) superfamily and is involved in morphogenesis and bone cell differentiation (2). Recombinant BMP-4 can induce ectopic cartilage and bone formation when implanted subcutaneously or intramuscularly in rodents. This ectopic bone formation process resembles the process of bone formation during embryogenesis and fracture healing (10). A cosmid clone containing the complete human bone morphogenetic protein-4 gene (BMP4) was isolated (details to be published elsewhere) and used as a probe to determine the precise chromosomal localization of the human BMP4 gene. This cosmid clone was labeled with biotin-14–dATP and hybridized *in situ* to chromosomal preparations of metaphase cells as described previously (6). In 20 metaphase preparations, an intense and specific fluorescence signal (FITC) was detected on the q arm of chromosome 14. The DAPI-counterstained chromosomes were computer-con-

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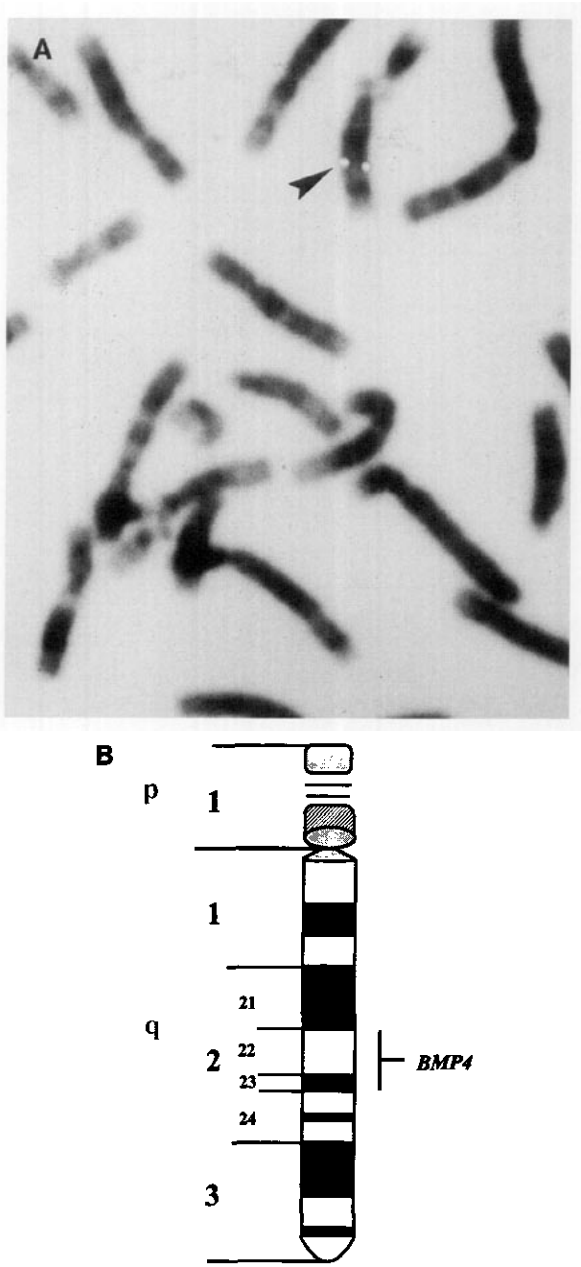

FIG. 1. Fluorescence *in situ* hybridization of the BMP4-containing cosmid to region q22–q23 of human chromosome 14. (**A**) Positively imaged chromosomes showing hybridization signal (arrowhead) and (**B**) chromosome 14: morbid anatomy with localization of BMP4.

verted into GTG-like banding patterns (Fig. 1), allowing the regional localization of BMP4 within 14q22–q23. This result is in agreement with a previous mapping of the human BMP4 gene to chromosome 14 (7). For this latter study a human–rodent somatic cell hybrid panel and a BMP-4 cDNA probe were used, which did not allow a more detailed chromosomal sublocalization. The present fine localization of BMP4 to 14q22–q23 makes BMP-4 a possible candidate gene for Holt–Oram syndrome (HOS). HOS is a heritable disorder of skeletal and cardiac development (5). The HOS phenotype is probably determined early in embryogenesis. This heterogeneous disorder (8) may be caused by several types of gene mutations or deletions on either chromosome 12q or 14q. Although one HOS locus has been localized to chromosome 12, other forms of HOS have also been described (1), which are not linked to chromosome 12 (6). In one case, a direct association has been established between HOS and a deletion of the 14q23–q24.2 region (9). The BMP4 localization to 14q22–q23 shown here in combination with the putative role of BMP-4 in limb development (3) and its distribution during development (4) suggests that the disturbed development of skeleton and heart in some HOS patients may be due to a disturbed BMP4 expression or an altered gene product. However, such an association of BMP4 with HOS still must be confirmed by genetic linkage and mutation analyses.

REFERENCES

1. Braulke, I., Herzhog, S., Thies, U., and Zoll, B. (1991). Holt–Oram syndrome in four half-siblings with unaffected parents: Brief clinical report. *Clin. Genet.* **39:** 241–244.
2. Centrella, M., Horowitz, M. C., Wozney, J. M., and McCarthy, T. (1994). Transforming growth factor-β gene family members and bone. *Endocr. Rev.* **15:** 27–39.
3. Francis, P. H., Richardson, M. K., Brickell, P. M., and Tickle, C. (1994). Bone morphogenetic proteins and signalling pathway that controls patterning in the developing chick limb. *Development* **120:** 209–218.
4. Jones, C. M., Lyons, K. M., and Hogan, L. M. (1991). Involvement of *Bone Morphogenetic Protein-4* (BMP4) and *Vgr-1* in morphogenesis and neurogenesis in the mouse. *Development* **111:** 531–542.
5. McKusick, V. A. (1994). "Mendelian Inheritance in Man," 11th ed., Johns Hopkins Univ. Press, Baltimore, MD.
6. Suijkerbuijk, R. F., Gebbink, M. F. B. G., Geurts van Kessel, A., and Moolenaar, W. (1993). Fine mapping of the human protein tyrosine phosphatase gene DTPRL1 to 18p11.2 by fluorescence in situ hybridization. *Cytogenet. Cell Genet.* **64:** 245–246.
7. Tabas, J. A., Hahn, G. V., Cohen, R. B., Seaunez, H. N., Modi, W. S., Wozney, J. M., Zasloff, M., and Kaplan, F. S. (1993). Chromosomal assignment of the human gene for bone morphogenetic protein 4. *Clin. Orthop. Relat. Res.* **293:** 310–316.
8. Terret, J. A., Newbury-Ecob, R., Cross, G. C., Fenton, I., Raeburn, J. A., Young, I. D., and Brook, J. D. (1994). Holt–Oram syndrome is a genetically heterogeneous disease with one locus mapping to human chromosome 12q. *Nature Genet.* **6:** 401–408.
9. Turleau, C., de Grouchy, J., and Chavin-Colin, F. (1984). Two patients with interstitial deletion (14q), one with features of the Holt–Oram syndrome: Exclusion mapping of PI (alpha-1-antitrypsin). *Ann. Genet.* **27:** 237–240.
10. Wang, E. A. (1993). Bone morphogenetic proteins (BMPs): Therapeutic potential in healing bony defects. *Trends Biotechnol.* **11:** 379–383.