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Human pancreatic amylase is encoded by two different genes

Peter C.Groot, Maria J.Bleeker, Jan C.Pronk, Fré Arwert, Willem H.Mager[1], Rudi J.Planta[1], Aldur W.Eriksson and Rune R.Frants

Institute of Human Genetics, Vrije Universiteit, van der Boechorststraat 7, 1081 BT Amsterdam and [1]Department of Biochemistry, Vrije Universiteit, de Boelelaan 1083, 1081 HV Amsterdam, The Netherlands
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Human alpha-amylase consists of two types of isozymes produced by salivary and pancreatic cells. The genes encoding the salivary isozyme (designated AMY1) form part of an extended multigene family [1,2; Groot *et al.* manuscript in preparation]. The gene encoding the pancreatic isozyme (AMY2) belongs to this family but, on the basis of an observed protein polymorphism [3], is assumed to represent a single gene. The AMY genes are about 9 kb DNA in length and consist of 11 and 10 exons for AMY1 and AMY2, respectively [4]. Using an AMY2 cDNA probe, two recombinants, each carrying a complete amylase gene were isolated from a human lambda library. About 900 nucleotides of the 5'-portions of both genes were sequenced, using synthetic oligonucleotides for both DNA strands. Comparison with AMY gene-sequences published previously [4] revealed them to represent pancreatic amylase genes (see Fig 2). Although showing considerable homology, the two AMY2 sequences differ at a number of positions. This indicates them to be distinct copies of the AMY2 gene, tentatively designated AMY2A and AMY2B, according to the nomenclature proposed by Gumucio *et al.* [2]. The restriction maps of the two lambda recombinants showed the presence of several differences both mutually and in comparison with AMY1 gene-containing DNA fragments (results not shown). On the basis of these differences certain restriction fragments could be specifically assigned to AMY1, AMY2A or AMY2B. For instance, a 3 kb BglII fragment hybridizing with a cDNA probe encompassing exons 4-8 indicates the presence of an AMY2B gene whereas the hybridization of an 8 kb BglII fragment points to an AMY2A gene. Since genomic Southern analysis of random families (see Fig. 1) always revealed the presence of both these fragments, it is unlikely that AMY2A and AMY2B are allelic. These results strongly suggest that the pancreatic amylase gene is duplicated in the human genome.

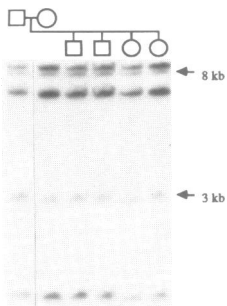

Fig. 1. Southern blot analysis of DNA from a random family. DNA was digested with BglII and probed with cDNA encompassing exons 4-8. Fragments of 3 kb and 8 kb, indicating the presence of AMY2B and AMY2A respectively, are marked with arrows.

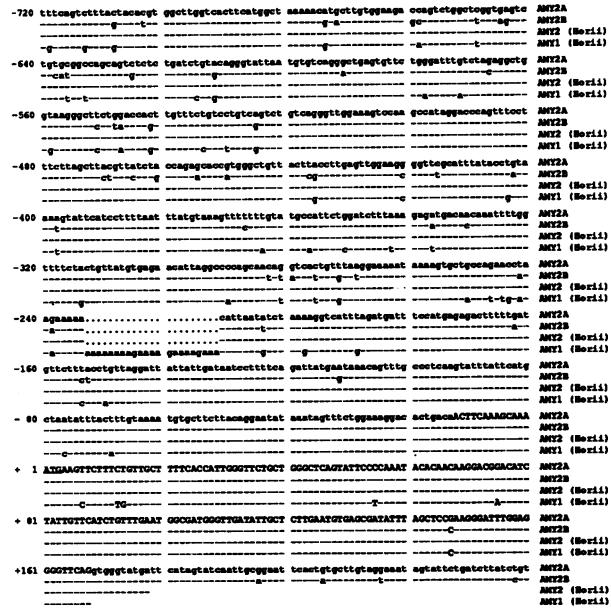

Fig. 2. Comparison of sequences of AMY2A and AMY2B with the sequences of AMY1 and AMY2 according to Horii *et al.*[4]. The sequence is numbered relative to the ATG start codon (A = +1). Exonic sequences are given in capitals, a dash indicates homology and a dot indicates a deletion.

References
1.Pronk, J.C., Frants, R.R., Jansen, W., Eriksson, A.W. and Tonino, G.J.M. (1982) Hum. Genet. 60, 32-35
2.Gumucio, D.L., Wiebauer, K., Caldwell, R.M., Samuelson, L.C. and Meisler, M.H. (1988) Mol. Cell Biol., in press
3.Merritt, A.D., Rivas, M.L., Bixler, D. and Newell, R (1973) Am. J. Hum. Genet. 25, 510-522
4.Horii, A., Emi, M., Tomita, N., Nishide, T., Ogawa, M. and Matsubara, K. (1987) Gene 60, 57-64