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SHORT COMMUNICATION

DINUCLEOTIDE REPEAT IN THE THIRD INTRON OF THE FABP3/MDGI PUTATIVE TUMOR SUPPRESSOR GENE

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The product of the cardiac fatty acid binding protein (also known as mammary-derived growth inhibitor: MDGI) has been shown to have modest antiproliferative activity for human breast cancer cells *in vitro* (Grosse *et al.*, 1991). It has recently been demonstrated that human breast cancer cells transfected with a MDGI expression construct exhibit a reduced proliferation rate and reduced tumorgenicity in nude mice as compared to controls (Huynh *et al.*, 1995). These results suggest that MDGI has tumor suppressor activity. The FABP3/MDGI gene has been mapped to chromosome 1 at p32–p33 (Peeters *et al.*, 1991; Troxler *et al.*, 1993). Here we describe a polymorphic dinucleotide repeat within the third intron of the FABP3/MDGI gene. This highly informative marker will be extremely useful in genotyping studies of chromosome 1 in the region of FABP3/MDGI. In particular, this polymorphism will be useful in studies of tumor types in which loss of MDGI function might contribute to the tumor phenotype.

A λEMBL3 FABP3 genomic clone was isolated by hybridization with the FABP3 cDNA. Sequence analysis revealed a $(GT)_{14}$ repeat located in the third intron of the gene. Unique sequences flanking this repeat were identified and used to design polymerase chain reaction (PCR) primers for amplification of the repeat-containing region. This repeat was found to be highly polymorphic (GenBank Accession no. U40222).

Primers were developed flanking this polymorphism [Forward (98F-1): 5'-TGCCTGTCTTAAGGATTTGCTG-3' (Coding strand); Reverse (98F-2): 5'-CACCATTGCGAGCATTCTACCC-3' (Antisense strand)]. PCR amplifications were performed in a total volume of 10 μl containing 20 ng genomic DNA, 0.25 μM of each primer, 0.025 μM ^{32}P end-labeled forward primer, 10 mM Tris-HCl pH 8.3, 50 mM KCl, 1.25 mM $MgCl_2$, 25 μM each dNTP, 0.2 U *Taq* polymerase. Cycling parameters were as follows: 94°C for 30 seconds, 60°C for 30 seconds, and 72°C for 60 seconds for 28 cycles. Samples were electrophoresed in a 6% polyacrylamide gel for 2 hours at 2000V. The gels were dried and exposed to film (OMAT-AR, Kodak). Allele sizes were determined by comparison to an M13mp18 DNA sequencing ladder.

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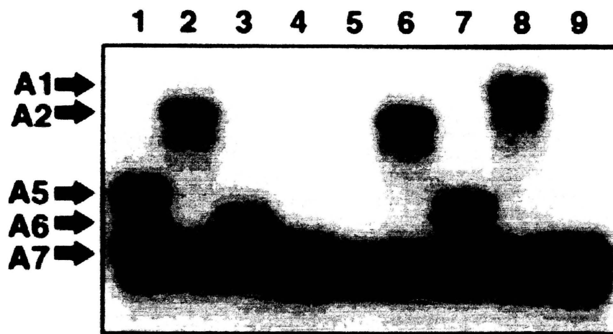

Figure 1. Autoradiograph demonstrating the detectionof five alleles in nine unrelated individuals (1–9) based on variation in (GT) repeat number.

Table 1. Frequency of FABP3/MDGI polymorphism alleles in 55 unrelated subjects.

Allele	Size (bp)	Frequency
A1	174	0.02
A2	172	0.20
A3	170	0.02
A4	168	0.05
A5	166	0.05
A6	164	0.13
A7	162	0.53

Seven different alleles were detected following electrophoresis of PCR products. Allele frequencies were determined based on 55 unrelated subjects (110 chromosomes). The observed heterozygosity was 0.60 and the polymorphism information content was 0.62. The size in basepairs and the frequency of each allele are listed in Table 1. A representative autoradiograph of alleles from nine individuals is shown in Figure 1.

The development of a polymorphic DNA marker at a putative tumor suppressor gene locus is very useful in the study of cancer. Specifically, the described polymorphism within the FABP3/MDGI gene is of interest in the study of breast cancer as this gene has been implicated in the breast tumorigenesis process.

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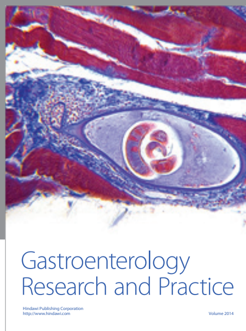
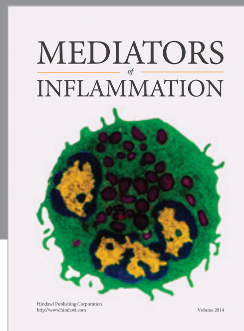
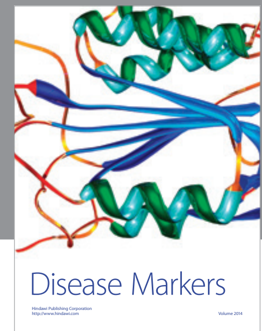
REFERENCES

Grosse, R., Boehmer, F.D., Langen, P., Kurtz, A., Lehmann, W., Mieth, M., Wallukat, G. (1991). Purification, biological assay, and immunoassay of mammary-derived growth inhibitor.

Methods Enzymol., **198**, 425–440.
Huynh, H.T., Larsson, C., Narod, S., Pollak, M. (1995). Tumor suppressor activity of the gene encoding mammary-derived growth inhibitor. *Cancer Res.*, **55**, 2225–2231.
Peeters, R.A., Veerkamp, J.H., van Kessel, A.G., Kanda, T., Ono, T. (1991). Cloning of the cDNA encoding human skeletal-muscle fatty-acid-binding protein, its peptide sequence, and chromosomal localization. *Biochem. J.*, **276**, 203–207.
Troxler, R.F., Offner, G.D., Jiang, J.W., Wu, B.L., Skare, J.C., Milunsky, A., Wyandt, H.E. (1993). Localization of the gene for human fatty acid binding protein to chromosome 1p32-1p33. *Hum. Genet.*, **92**, 563–566.







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