

## FCER1A gene proximal promoter polymorphisms in Caucasians and East Asians

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### Summary

Two *FCER1A* gene proximal promoter polymorphisms, –344C > T and –95T > C, both associated with total serum IgE and/or allergic disorders in Caucasians and East Asians, were shown to influence the gene expression in additive manner. In face of the rarity of other proximal promoter variants in Caucasians or their lack in East Asians, future investigations of the *FCER1A* locus in these ethnic groups should probably focus on three common haplotypes of the common –344C > T and –95T > C polymorphisms.

The high affinity IgE receptor (FcεRI) plays a central role in allergic response. Thus, not surprisingly, its α-subunit gene (*FCER1A*) variability was recently studied not only in human (Shikanai *et al.*, 2002; Hasegawa *et al.*, 2003; Potaczek *et al.*, 2006, 2007; Bae *et al.*, 2007) but also in other mammals (Vychodilova-Krenkova *et al.*, 2005).

Two frequent polymorphisms located in the proximal promoter of *FCER1A* gene, –344C > T<sup>a</sup> and –95T > C, were shown to be associated with allergic disorders and/or total serum IgE levels in Caucasian (Shikanai *et al.*, 2002; Potaczek *et al.*, 2006, 2007) and East Asian (Hasegawa *et al.*, 2003; Bae *et al.*, 2007) allergic subjects. Both mutations were shown to be functional (Hasegawa *et al.*, 2003; Bae *et al.*, 2007) affecting *FCER1A* gene expression in an additive manner (Kanada *et al.*, 2008). They are completely linked in Caucasians (Potaczek *et al.*, 2006, 2007) as evidenced by the presence of only three of four theoretically possible haplotypes (–344C/–95C, –344C/–95T and –344T/–95T), significant likelihood

ratio test for linkage disequilibrium (Excoffier *et al.*, 2005) and Lewontin's index of diversity (*D'*) value of 1.0<sup>b</sup>. *D'* values obtained in two East Asian populations of Koreans (0.74) and Japanese (1.0) are conflicting, however, only three of four theoretical haplotypes (–344C/–95C, –344C/–95T and –344T/–95T) were observed in the latter (Bae *et al.*, 2007; Kanada *et al.*, 2008).

Two more polymorphisms were reported to be localized upstream in that region in American Caucasian and African Americans (Shikanai *et al.*, 2002) but no haplotypic linkage analysis of them was done. To describe full haplotypes of *FCER1A* proximal promoter polymorphisms in Caucasians, we tested for their presence in 48 Poles. In brief, genomic DNA extracted from peripheral blood leucocytes and amplified with primers pairs AATGGTTTAACTTCTGCCTTCC (1F)/ACCA GAGTACCAACGCTTTT (1R) and TGCCTAACT GATCAGCAGGA (2F)/AGTGAGGTAAAGTAACT TGTTTTGG (2R) were subjected to single-strand conformation polymorphism (SSCP) technique as described previously (Potaczek *et al.*, 2006, 2007). Amplification products having altered electrophoretic mobility were cloned (TA Cloning; Invitrogen, Carlsbad, CA, USA) and sequenced (Applied Biosystems, Foster City, CA, USA). Two polymorphisms, –778A > C and –673G > A, most probably corresponding to those reported by Shikanai *et al.* (2002) were found. Further genotyping was completed using restriction fragment length polymorphism (RFLP) method (–778A > C: primers pair CACAGTAGAGAAAGAGGAGTCTCTTA<sup>c</sup> [3F]/[2R], restriction endonucleases: *Tru*II (Fermentas UAB, Vilnius, Lithuania) or *Mse*I (New England Biolabs, Ipswich, MA, USA); –673G > A: primers pair [1F]/GGGATTACCAGAGCTACCC<sup>c</sup>A [3R], restriction endonucleases: *Hsp*92II (Promega, Madison, WI, USA) or *Nla*III (New England Biolabs). Both polymorphisms genotyped in total of 88 Caucasians of the Polish origin were shown to be rare, with minor allele frequencies of

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<sup>a</sup> All polymorphisms were numbered according to translation start site.

<sup>b</sup> *D'* value was calculated currently using Arlequin 3.1 (see footnote 'd') on data obtained in 368 Polish Caucasians, 308 genotyped previously (Potaczek *et al.*, 2007) and 60 new cases genotyped as described elsewhere (Potaczek *et al.*, 2006).

<sup>c</sup> The mutagenic nucleotides introducing digestion sites in the primers sequences were underlined.

0.023 (–778C) and 0.017 (–673A). Haplotype analysis<sup>d</sup> evidenced five haplotypes of –778A > C, –673G > A, –344C > T and –95T > C polymorphisms, i.e. A|G|C|C, A|G|C|T, C|A|C|T and C|G|C|T. Lewontin's *D'* was of 1.0 for all possible variants pairs. Total serum IgE levels did not apparently differ between carriers of the rare alleles and wild-type homozygotes (–778AC vs. –778AA,  $35.0 \pm 18.1$  vs.  $23.5 \pm 20.9$  IU mL<sup>–1</sup>, respectively; –673GA vs. –673GG,  $27.0 \pm 10.3$  vs.  $23.9 \pm 21.1$  IU mL<sup>–1</sup>, respectively), however, any reliable statistical comparisons were rather not possible due to low numbers of subjects carrying rare alleles of –778A > C and –673G > A polymorphisms. Neither –778A > C nor –673G > A polymorphisms were found in 91 East Asian subjects of the Japanese origin, which was in line with the results by Bae *et al.* (2007) obtained in Koreans. Due to absence of –778C and –673A variants in East Asians, and their very low frequencies and linkage to frequent, functional –344C > T and –95T > C polymorphisms in Caucasians, their importance in these racial groups is probably limited. A pairwise linkage disequilibrium of the rare variants (–778C|–673A) and their genetic linkage to a particular haplotype of the common variants (–344C|–95T) suggest its recent origin in Caucasians.

In conclusion, further investigations, especially functional and pharmacogenetic, of *FCER1A* proximal promoter in Caucasians or East Asians should focus on three common haplotypes of its two most frequent polymorphisms.

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<sup>d</sup> Haplotypes distribution was analysed using Arlequin 3.1 (<http://cmpg.unibe.ch/software/arlequin3/>) (Excoffier *et al.*, 2005). *D'* value was determined using Haploview\_4.0 (<http://www.broad.mit.edu/mpg/haploview/>) (Barrett *et al.*, 2005).