

Table S1. Characteristics of investigated polymorphisms, variant allele frequency and agreement with Hardy-Weinberg equilibrium in controls.

SNP	Nucleotide/ amino acid change	Location	SNP function	VAF (controls)	p_{HWE} (controls)
<i>SOD2</i> rs4880	p.Ala16Val	Coding region nsSNP	Decreased enzyme activity [1]	0.493	0.304
<i>CAT</i> rs1001179	c.-262C > T	5'UTR	Altered expression [2]	0.249	0.536
<i>GPX1</i> rs1050450	p.Pro198Leu	Coding region nsSNP	Decreased enzyme activity [3]	0.307	0.070
<i>GSTP1</i> rs1695	p.Ile105Val	Coding region nsSNP	Decreased enzyme activity [4]	0.333	0.871
<i>GSTP1</i> rs1138272	p.Ala114Val	Coding region nsSNP	Decreased enzyme activity [4]	0.100	0.805
<i>IL1B</i> rs1143623	c.-1560G > C	5'UTR	Altered expression [5]	0.286	0.741
<i>IL1B</i> rs16944	c.-598T > C	5'UTR	Altered expression [6]	0.656	0.340
<i>IL6</i> rs1800795	c.-174G > C	5'UTR	Altered expression [7]	0.423	0.106
<i>TNF</i> rs1800629	c.-308 G > A	5'UTR	Altered expression [8]	0.177	0.546

HWE–Hardy-Weinberg equilibrium, ns–non-synonymous, SNP–single nucleotide polymorphism, UTR–untranslated region, VAF–variant allele frequency.

Table S2. Primers used for multiplex PCR (a) and thermal cycling conditions used for genotyping for multiplex PCR (b) and KASP chemistry (c).

a).		Primer sequences	
GSTM1	Forward	5' CTGGATTGTAGCAGATCATGC 3'	
	Reverse	5' CTCCTGATTATGACAGAAGCC 3'	
GSTT1	Forward	5' TTCCTTACTGGTCCTCACATCTC 3'	
	Reverse	5' TCACCGGATCATGGCCAGCA 3'	
β -globin	Forward	5' GAAGAGCCAAGGACAGGTAC 3'	
	Reverse	5' CAACTTCATCCACGTTTCACC 3'	
b)		Multiplex PCR protocol	
Stage	Temperature	Duration	Cycles no.
Start denaturation	94 °C	10 min	1
Denaturation	94 °C	30 s	35
Annealing	60 °C	30 s	
Extension	72 °C	30 s	
Final extension	72 °C	10 min	1
c)		61-55 °C Touchdown protocol	
Stage	Temperature	Duration	Cycles no.
Hot-start Taq activation	94°C	15 min	1
Touchdown	94°C	20 s	10
	61 °C (61 °C decreasing 0.6 °C per cycle to achieve a final annealing / extension temperature of 55 °C)	60 s	
Amplification	94 °C	20 s	30
	55 °C	60 s	
Read stage	30 °C	60 s	1

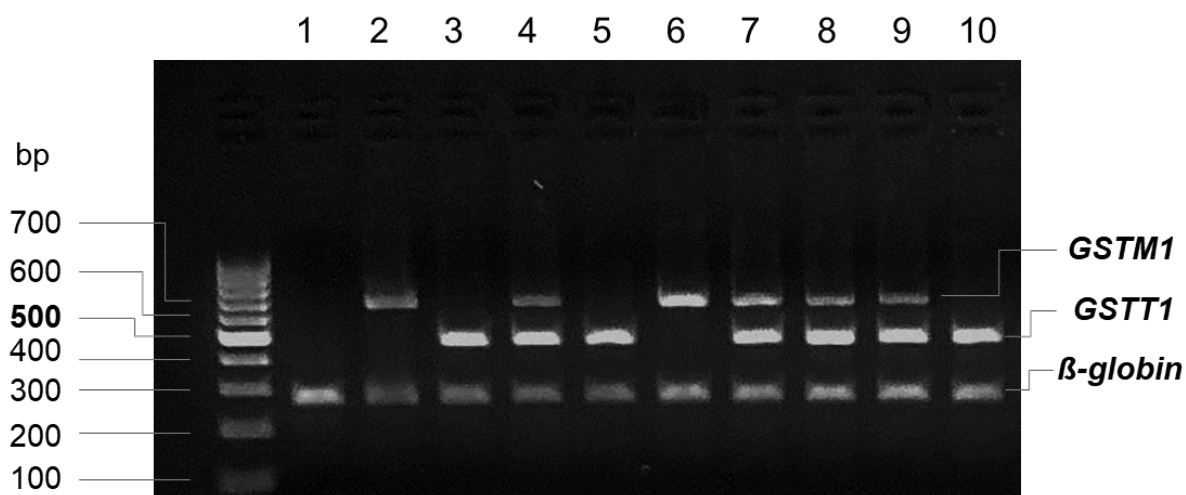


Figure S1. Representative gel image of *GSTT1* and *GSTM1* genotyping analysis. *GSTT1* and/or *GSTM1* deletions were determined simultaneously with multiplex PCR followed by electrophoresis (40 min at 100 V) on 2% agarose gel in TBE buffer stained with ethidium bromide. *GSTM1* and *GSTT1* genotypes were determined by the presence or absence of the respective amplification products. β -globin served as internal control of amplification. Amplicon lengths: *GSTM1* - 600 bp, *GSTT1* - 480 bp, β -globin gene - 268 bp. Lane 1: homozygous *GSTM1* and *GSTT1* deletion, lanes 2 and 6: homozygous *GSTT1* deletion, lanes 3, 5 and 10: homozygous *GSTM1* deletion, lanes 4, 7, 8, and 9: *GSTM1* and *GSTT1* gene present. The 100 bp DNA Ladder (Fermentas, Thermo Fisher Scientific, Waltham, MA, USA) was used as a reference.

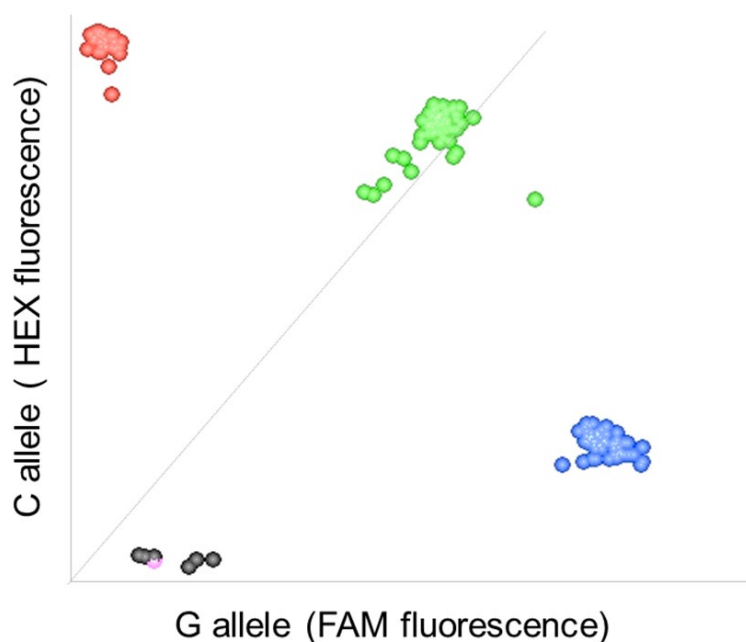


Figure S2. Representative cluster image for *IL6* rs1800795 analysis obtained after KASP competitive allele specific PCR. Fluorescence of amplified products was measured with microplate reader (FLUOstar Omega, BTG LABTECH, Ortenberg, Germany) and processed with KlusterCaller program (LGC Genomics, Hoddesdon, UK). Genotypes were determined considering fluorescence signal detected; for homozygous samples either FAM (labelled G allele) or HEX (labelled C allele) fluorescence signal was detected and for heterozygous both of fluorescence signals were detected. Black cluster: no template controls (NTC); red cluster: homozygous for allele C (CC genotype); green cluster: heterozygous (CG genotype); blue cluster: homozygous for allele G (GG genotype).

References

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