

Table SI. Polymorphisms in the metabolic pathway and response to methotrexate.

Chromosome	Gene	Polymorphism	dbSNP ^b	Sequence of the primer pairs	Allele change (ancestral allele)	Residue change	Chromosome position, bp ^a	Location in gene	Functional outcome
1	MTHFR	C677T	rs1801133	F, CgAAgCAGggAgCTTTgAggCTG R, AggACggTgCggTgAgAgTg	G(C)C>GTC	Alanine>Valine	11,796,321	Coding region	Missense
		A1298T	rs1801131	F, CTTTggggAgCTgAAggACTACTAC R, CACTTTTgTgACCAATCCggTTTg	G(A)A>GCA	Glutamate>Alanine	11,794,419	Coding region	Missense
2	ATIC	C347G	rs2372536	F, TTTTATAgAgTTgttgCCTgCA R, TCCTCTCTAgTCTTCTAAATggTT	A(C)T>AGT	Threonine>Serine	215,325,297	Coding region	Missense
			rs16853834	F, AACATTTCTCACTgTgTgACgT R, gACCTCAgAgATgAACCCAgCT	C>T	-	215,345,513	Intron variant	-
5	DHFR		rs10072026	F, ACCTTgTgATATTAAGCagCTT R, TTCACCCACAAGAAAATCATgTCT	T>C	-	80,649,321	Intron variant	-
9	FPGS		rs1544105	F, CCTCCATgCAATCCAAATCCAACTC R, CCCAgggACACTgCTAATT	C>T	-	127,800,446	promoter	-
21	RFC1	A80G	rs1051266	F, gCTgAgTCTCAAAATAgCTTCATg R, CAgcagATTggAAACACTgT	CAC>CGC	Histidine>Arginine	45,537,880	Coding region	Missense

^aOrder of the polymorphisms is according to the chromosomal position. ^bSNP ID in dbSNP. F, forward; R, reverse; MTHFR, methylenetetrahydrofolate reductase; ATIC, 5-aminoimidazole-4-carboxamide ribonucleotide formyltransferase; DHFR, dihydrofolate reductase; FPGS, folypolyglutamate synthetase; RFC, reduced folate carrier; SNP, single nucleotide polymorphism.

Table SII. PCR thermocycling conditions.

A, rs 1072026, rs 2372536, rs 1051266, rs 1801133, rs 1801131, rs 1544105

Parameter	Denaturation	Cycling Quantification			Melting Melting curve			Cooling
Analysis mode	none	45			1			None
Cycles	1							1
Segment	1	1	2	3	1	2	3	1
Target, °C	95	95	55	72	95	40	85	40
Hold (mm:ss)	10:00	00:05	00:10	00:06	00:20	00:20	-	00:30
Ramp rate, °C/s	4/4	4/4	2/2	4/4	4/4	2/2	4/4	20
Acquisition mode	None	None	Single	None	None	None	Continuous	None

B, rs 16853834

Parameter	Denaturation	Cycling Quantification			Melting Melting curve			Cooling
Analysis mode	none	45			1			None
Cycles	1							1
Segment	1	1	2	3	1	2	3	1
Target, °C	95	95	60	72	95	40	75	40
Hold, mm:ss	10:00	00:05	00:10	00:06	00:20	02:00	-	00:30
Ramp rate, °C/s	4/4	4/4	2/2	4/4	4/4	1/5	0/19	1/5
Acquisition mode	None	None	Single	None	None	None	Continuous	None