ORIGINAL ARTICLE

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Three hundred twenty-six genetic variations in genes encoding nine members of ATP-binding cassette, subfamily B (ABCB/MDR/TAP), in the Japanese population

Received: September 18, 2001 / Accepted: November 2, 2001

Abstract We screened DNAs from 48 Japanese individuals for single-nucleotide polymorphisms (SNPs) in nine genes encoding components of ATP-binding cassette subfamily B (*ABCB/MDR/TAP*) by directly sequencing the entire applicable genomic regions except for repetitive elements. This approach identified 297 SNPs and 29 insertion/deletion polymorphisms among the nine genes. Of the 297 SNPs, 50 were identified in the *ABCB1* gene, 14 in *TAP1*, 35 in *TAP2*, 48 in *ABCB4*, 13 in *ABCB7*, 21 in *ABCB8*, 21 in *ABCB9*, 13 in *ABCB10*, and 82 in *ABCB11*. Thirteen were located in 5' flanking regions, 237 in introns, 37 in exons, and 10 in 3' flanking regions. These variants may contribute to investigations of possible correlations between genotypes and disease-susceptibility phenotypes or responsiveness to drug therapy.

Key words Single-nucleotide polymorphism (SNP) · ATP-binding cassette, subfamily B (ABCB) · Multidrug resistance (MDR) · P-glycoprotein · Transporter associated with antigen processing (TAP) · Major histocompatibility complex (MHC)

Introduction

The ATP-binding cassette (ABC) is a protein complex that plays an important role in membrane transport. Its full-molecule (FM) form includes two ATP-binding segments and two transmembrane (TM) regions, and the half-molecule (HM) contains one ATP-binding and one TM

domain (Hyde et al. 1990). ABC proteins constitute a superfamily consisting of eight subfamilies: ABC1, MDR/TAP, CFTR/MRP, ALD, OABP, GCN20, WHITE, and ANSA (Kerb et al. 2001; Human ABC gene nomenclature committee, <http://www.gene.ucl.ac.uk/nomenclature/genefamily/abc.html>).

Members of the MDR/TAP subfamily include the ATP-binding cassette, subfamily B (ABCB) and the transporter associated with antigen processing (TAP). The *ABCB1* [ATP-binding cassette, subfamily B, member 1, also called multidrug resistance (*MDR*)-1] gene encodes P-glycoprotein (PGP; Gros et al. 1986), which is expressed in normal human tissues such as small and large intestine, adrenal gland, kidney, liver, and capillary endothelial cells of brain and testis (Fojo et al. 1987; Thiebaut et al. 1987; Sugawara et al. 1988; Cordon-Cardo et al. 1989). Experiments in mice carrying a homozygous disruption of the *mdr1a* gene have shown that PGP plays an important role in the blood–brain barrier, since its absence results in elevated drug concentrations in the brain and many other tissues (Schinkel et al. 1994). Those observations suggest that PGP plays a role in protection against toxic xenobiotics. Among human subjects, various polymorphisms have been identified in the *ABCB1* gene (Mickley et al. 1998; Hoffmeyer et al. 2000; Cascorbi et al. 2001; Tanabe et al. 2001), and phenotypic consequences have been observed for three of those SNPs: (1) individuals homozygous for a C-to-T synonymous substitution at codon 1145 showed significantly lower duodenal PGP expression and had increased plasma levels of digoxin, a substrate of PGP (Hoffmeyer et al. 2000); (2) A T-to-C polymorphism (exon 2, +202) and a G-to-T or -A polymorphism in exon 22 were correlated with levels of PGP expression in the placenta; and (3) individuals heterozygous (T/C) at an SNP site in the promoter region had significantly lower levels of PGP than T/T wild types (Tanabe et al. 2001).

The transporter associated with antigen processing (TAP), a molecule that provides peptides to major histocompatibility complex (MHC) class I molecules in the endoplasmic reticulum, is comprised of two subunits (TAP1 and TAP2) that form a heterodimeric complex in the mem-

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A *ATP-binding cassette, subfamily B, member1 (ABCB1)*

Chromosomal localization : 7q21.1

Fig. 1A–I. Locations of single-nucleotide polymorphisms (SNPs) in the *ABCB1* (A), *TAP1* (B), *TAP2* (C), *ABCB4* (D), *ABCB7* (E), *ABCB8* (F), *ABCB9* (G), *ABCB10* (H), and *ABCB11* (I) genes, indi-

cated by vertical lines. Open boxes represent exons; hatching on the chromosomes indicates regions of repetitive elements. ATG and TGA or TAA, initiation and stop codons, respectively

brane of the endoplasmic reticulum and proximal Golgi (Neefjes et al. 1993). TAP is essential for MHC class I-restricted presentation of antigen, as demonstrated by absent or low expression of MHC class I molecules in TAP-deficient mice and cell lines (Spies and DeMars 1991). Tu-

mor cell lines frequently show impaired TAP expression and/or function, underlining the key role of TAP in immune surveillance and prevention of tumor growth mediated by MHC class I proteins (Algarra et al. 2000). Chen et al. (1996) have described a case of human lung tumor with

B *Transporter associated with antigen processing 1 (TAP1)*

Chromosomal localization : 6p21.3

C *Transporter associated with antigen processing 2 (TAP2)*

Chromosomal localization : 6p21.3

D *ATP-binding cassette, subfamily B, member 4 (ABCB4)*

Chromosomal localization : 7q21.1

Fig. 1A-I. Continued

impaired TAP function due to a mutation in the Walker B region of *TAP1*, and a number of investigators have examined potential associations of *TAP1* and *TAP2* genotypes with altered susceptibilities to MHC-associated diseases. Results from these investigations suggest that mutations in the *TAP1* and *TAP2* genes may be associated with ankylosing spondylitis (Colonna et al. 1992), insulin-dependent diabetes mellitus (Colonna et al. 1992; Jackson and Capra 1993), and celiac disease (Colonna et al. 1992).

The *ABCB4* gene, also known as *MDR3* or *MDR2*, the homologue of murine *mdr2* (Linke et al. 1991), encodes the MDR3 protein, which is a member of the superfamily of ABC transporters (van der Bliek et al. 1987). High concentrations of ABCB4 in the bile canalicular membrane indicate its possible involvement in transport of phospholipids from hepatocytes to the bile duct, although its function has not yet been clarified (Smit et al. 1993). Mutations in the human *ABCB4* gene are reportedly associated with progressive familial intrahepatic cholestasis (de Vree et al.

E *ATP-binding cassette, subfamily B, member 7 (ABCB7)*

Chromosomal localization : Xq12-q13

F *ATP-binding cassette, subfamily B, member 8 (ABCB8)*

Chromosomal localization : 7q36

G *ATP binding cassette, subfamily B, member 9 (ABCB9)*

Chromosomal localization : 12q24

Fig. 1A-I. Continued

1998; Jacquemin et al. 1999) and intrahepatic cholestasis of pregnancy (Dixon et al. 2000).

cDNAs encoding *ABCB7* (Shimada et al. 1998), *ABCB8* (Hogue et al. 1999), *ABCB9* (Zhang et al. 2000a), and *ABCB10* (Zhang et al. 2000b) were isolated by screening an expressed sequence tags (EST) database. *ABCB7*, an ABC half-transporter located in the mitochondrial inner membrane, may play a central role in the maturation of cytosolic proteins containing the iron-sulfur (Fe/S) cluster (Shimada et al. 1998). Mutations in the *ABCB7* gene are associated with X-linked sideroblastic anemia and ataxia (XLSA/A) (Allikmets et al. 1999; Bekri et al. 2000). *ABCB8* is an ABC half-transporter located in the mitochondria, but its func-

tion is unclear (Hogue et al. 1999). *ABCB9* mRNA is expressed at a high level in testis and at moderate levels in brain and spinal cord, although the function of the gene product, located in lysosomes, similarly is unknown (Zhang et al. 2000a). *ABCB10* is expressed ubiquitously, but is especially abundant in bone marrow; its product is located in mitochondria, but the function of *ABCB10* is also obscure (Zhang et al. 2000b).

Gerloff et al. (1998) identified a rat gene, *Spgp*, which encoded a novel ABC transporter (*ABCB11*), Sister of P-Glycoprotein (SPGF). In the rat, SPGF is expressed only in the liver canalicular membrane, and it appears to function as a bile salt transporter or a bile salt export pump

H *ATP binding cassette, subfamily B, member 10 (ABCB10)*

Chromosomal localization : 1q42

I *ATP-binding cassette, subfamily B, member 11 (ABCB11)*

Chromosomal localization : 2q24

Fig. 1A-I. Continued

(Gerloff et al. 1998). In humans, mutations in the *ABCB11* coding sequence cause progressive familial intrahepatic cholestasis, evidence that *ABCB11* is the major canalicular bile salt export pump in human (Strautnieks et al. 1998).

To investigate the nature of apparent genotype/phenotype correlations for some ABC-transporters in

more detail, we began by searching for additional SNPs in the nine *ABCB* genes described above, including their promoter regions and introns, but excluding repetitive elements, and we report here a total of 326 genetic variations, of which 226 have not been previously reported.

Table 1A. Summary of genetic variations detected in the *ABCB1* gene

No.	Location	Position ^a	Genetic variation	NCBI SNP ID
1	5' Flanking	−196	T/C	
2	5' Flanking	−16	T/C	
3	Intron 1	71660	A/C	
4	Intron 1	80091	A/C	
5	Intron 1	103126	T/C	
6	Intron 1	103148	C/T	
7	Intron 1	108428	A/G	
8	Intron 1	112042	A/G ^d	
9	Exon 2	202	T/C(5'UTR) ^{b,d}	
10	Intron 2	491	G/del	
11	Intron 4	36	C/T	
12	Intron 5	1596	T/C	
13	Intron 7	139	T/C ^{b,c}	rs1202168
14	Intron 7	251	G/A	rs1202169
15	Intron 8	1789	C/T	
16	Intron 9	7225	A/G	rs1922240
17	Exon 13	12	T/C(Gly412Gly) ^{b,c,d}	rs2032588
18	Intron 14	24	T/C	
19	Intron 14	81	C/T	
20	Intron 15	38	A/G	
21	Intron 17	73	A/G	
22	Intron 17	472	T/A ^{b,c}	
23	Intron 18	564	G/A	
24	Intron 18	2062	C/T	
25	Intron 18	2293	A/G	
26	Intron 20	557	G/A	
27	Intron 21	24	G/A	
28	Intron 21	2725	A/G	
29	Intron 21	4725	A/G	
30	Exon 22	196	G/T/A(893Ala/Ser/Thr) ^{c,d}	rs2032582
31	Intron 22	49	T/C	rs2032583
32	Intron 22	8507	T/C	
33	Intron 22	8537	T/A	
34	Intron 22	8565	T/C	
35	Intron 22	8952	G/A	
36	Intron 22	9520	A/G	
37	Intron 22	9836	C/T	
38	Intron 24	377	C/A	
39	Intron 24	1493	A/del	
40	Intron 24	1495	A/T	
41	Intron 25	342	C/T	
42	Intron 26	134	C/G	
43	Intron 26	1043	G/A	rs1922243
44	Intron 26	1272	A/G	
45	Intron 26	1394	A/G	
46	Intron 26	1987–1988	AAAG/ins	
47	Exon 27	153	C/T(Ile1145Ile) ^{b,c,d}	rs1045642
48	Intron 27	59	G/T	
49	Intron 27	80	T/C	
50	Intron 28	1220	A/G	rs1186746
51	Intron 28	1266	G/T	rs1186745
52	Exon 29	400	A/G(3'UTR) ^d	rs3842
53	3' Flanking	264	G/A	rs1055302

ABCB1, ATP-binding cassette, subfamily B, member 1; NCBI, National Center for Biotechnology Information; SNP, single-nucleotide polymorphism; UTR, untranslated region; del, deletion; ins, insertion

^a For SNPs in the 5' flanking region, intron region, or 3' flanking region, nucleotide positions are counted from the first intronic nucleotide at the exon/intron junction (for SNPs in the exon region, nucleotide positions are counted from the first exonic nucleotide at the exon/intron junction)

^b SNPs previously reported by Hoffmeyer et al. (2000)

^c SNPs previously reported by Cascorbi et al. (2001)

^d SNPs previously reported by Tanabe et al. (2001)

Table 1B. Summary of SNPs detected in the *TAP1* gene

No.	Location	Position ^a	SNP	NCBI SNP ID
1	5' Flanking	-673	G/C	rs1351382
2	5' Flanking	-646	T/G	rs1351383
3	5' Flanking	-563	A/C	
4	5' Flanking	-236	G/T	
5	Intron 3	408	C/T	
6	Exon 4	153	A/G(Ile393Val) ^{b,c}	rs1057141
7	Intron 4	289	G/T	
8	Intron 4	291	C/G	rs2071539
9	Intron 5	1139	C/T	
10	Intron 7	375	C/T	rs735883
11	Exon 11	284	G/T(3'UTR) ^b	rs1057373
12	3' Flanking	71	G/A	rs2071540
13	3' Flanking	129	T/C	rs2071541
14	3' Flanking	459	G/A	rs2071463

TAP1, transporter associated with antigen processing 1

^b SNPs previously reported by Colonna et al. (1992)^c SNP previously reported by Jackson and Capra (1993)

Materials and methods

Exon–intron boundaries of the *ABCB1*, *TAP1*, *TAP2*, *ABCB4*, *ABCB7*, *ABCB8*, *ABCB9*, *ABCB10*, and *ABCB11* genes were defined by comparing genomic sequences with mRNA sequences. Accession numbers of the genomic sequences obtained for this study are as follows:

ABCB1 (AC002457.1, AC005068.1)*TAP1* (X66401.1)*TAP2* (X66401.1)*ABCB4* (AC079591.1, AC079303.3, AC005045.2)*ABCB7* (AL360179.3, AC002417.1)*ABCB8* (AC010973.4)*ABCB9* (AC026362.9, AC073857.10)*ABCB10* (AL121990.9)*ABCB11* (AC008177.3, AC069137.3)**Table 1C.** Summary of genetic variations detected in the *TAP2* gene

No.	Location	Position ^a	Genetic variation	NCBI SNP ID
1	5' Flanking	-63	C/T ^b	
2	5' Flanking	-55	G/A ^b	
3	Exon 1	61	T/C(5'UTR) ^b	
4	Intron 1	39	G/A ^b	
5	Intron 1	311	A/G ^b	
6	Intron 2	48	G/C ^b	
7	Intron 3	7	G/A ^b	
8	Intron 3	8	G/A	
9	Intron 3	265	A/G ^b	
10	Intron 3	1474	T/C ^b	rs241429
11	Intron 4	104	C/T	
12	Intron 5	111	G/A ^b	rs241430
13	Intron 5	124	G/A ^b	
14	Exon 6	190	G/A(Val379Ile) ^{b,c}	rs1800454
15	Exon 7	15	G/T(Gly386Gly) ^b	
16	Intron 7	1379	G/A	rs1015166
17	Intron 7	1399	G/A	rs117821
18	Intron 9	168	C/T	rs241436
19	Intron 10	23	C/T	rs241437
20	Intron 10	87	G/A	rs241438
21	Intron 10	170	A/C	rs241439
22	Intron 10	219	A/G	
23	Intron 10	346	G/A	rs241440
24	Exon 11	17	G/A(Gly604Gly)	rs241441
25	Intron 11	9	C/T	rs241442
26	Intron 11	62	C/A	rs241443
27	Intron 11	68	C/T	rs241444
28	Intron 11	105	G/A	rs241445
29	Intron 11	210	C/T	rs241446
30	Intron 11	317–319	GTG/del	
31	Exon 12	19	T/C(Cys651Arg)	
32	Exon 12	61	A/G(Thr665Ala) ^{c,d}	rs241447
33	Exon 12	127	T/C(stop687Gln) ^{c,d}	rs241448
34	Exon 12	159	G/T(3'UTR) ^{c,d}	rs241449
35	Exon 12	291	G/A(3'UTR)	rs1871666
36	Exon 12	332	A/G(3'UTR)	rs241451
37	Exon 12	356–357	GG/TGGTGGGGTGGGA(3'UTR)	

TAP2, transporter associated with antigen processing 2

^b SNPs previously reported by Jeffreys et al. (2000)^c SNPs previously reported by Colonna et al. (1992)^d SNPs previously reported by Powis et al. (1992)

Table 1D. Summary of genetic variations detected in the *ABCB4* gene

No.	Location	Position ^a	Genetic variation	NCBI SNP ID
1	Exon 3	3	C/T(5'UTR)	
2	Intron 3	45	T/C	
3	Intron 3	498	C/T	
4	Intron 3	515	A/G	
5	Intron 6	1030	G/C	
6	Intron 6	1437	G/A	
7	Intron 6	2449	G/A	
8	Intron 7	451	A/C	
9	Intron 7	530	C/G	
10	Intron 7	−1023	A/G	rs988448
11	Intron 7	−152	T/C	
12	Exon 8	40	C/T(Leu59Leu)	
13	Intron 8	130	T/C	
14	Intron 8	248	A/G	
15	Intron 8	531	A/G	
16	Intron 8	4240	T/A	
17	Intron 8	4343	C/T	
18	Intron 8	4677	G/C	
19	Intron 9	113	A/G	
20	Intron 9	982	T/A	
21	Exon 10	160	C/T(Asn168Asn)	rs1202283
22	Intron 10	218	T/C	rs1473154
23	Intron 11	241	A/G	
24	Intron 11	457	A/G	
25	Intron 11	589	T/G	rs1473152
26	Intron 11	1337	C/G	
27	Exon 12	3	A/T(Ile237Ile)	
28	Intron 12	1288	T/C	
29	Intron 12	2697	G/T	rs1014283
30	Intron 13	206	G/A	
31	Intron 13	988	T/C	
32	Intron 13	1386	A/G	rs1468615
33	Intron 13	1413–1414	T/ins	
34	Intron 13	1931	A/G	
35	Intron 14	402	A/C	rs1149222
36	Intron 18	519	G/A	rs31674
37	Intron 20	1819	C/T	rs31669
38	Intron 21	16	T/C	rs31668
39	Intron 21	72	C/T	rs31667
40	Intron 22	767	A/del	
41	Intron 23	784	T/C	
42	Intron 25	158	T/C	
43	Intron 25	1044	C/T	rs31662
44	Intron 25	2920	T/A	
45	Intron 27	580	A/G	rs31659
46	Intron 27	2068	G/A	rs31658
47	Intron 29	411	A/C	
48	Intron 30	2991	C/T	rs31653
49	Intron 31	642	C/T	rs1526090
50	3' Flanking	458	T/C	

ABCB4, ATP-binding cassette, subfamily B, member 4

Accession numbers of the mRNA sequences are as follows:

ABCB1 (M14758.1)
TAP1 (NM_000593.2)
TAP2 (XM_039609.1)
ABCB4 (NM_000443.2)
ABCB7 (AF038950.1)
ABCB8 (NM_007188.2)
ABCB9 (AF216494.1)
ABCB10 (NM_012089.1)
ABCB11 (NM_003742.1)

Table 1E. Summary of genetic variations detected in the *ABCB7* gene

No.	Location	Position	Genetic variation	NCBI SNP ID
1	Intron 1	220	C/A	
2	Intron 1	480	G/A	
3	Intron 1	512–513	AT/del	
4	Intron 1	1690	G/A	
5	Intron 1	5309	G/A	
6	Intron 1	−11274	A/G	
7	Intron 1	−11085	A/G	
8	Intron 1	−10037	T/C	
9	Intron 1	−21	G/A	
10	Intron 3	135–136	A/ins	
11	Intron 3	333	G/A	
12	Intron 12	524	C/T	
13	Intron 13	1543	G/A	
14	Intron 13	2400	C/G	
15	Intron 15	2201	G/C	

ABCB7, ATP-binding cassette, subfamily B, member 7

Table 1F. Summary of genetic variations detected in the *ABCB8* gene

No.	Location	Position	Genetic variation	NCBI SNP ID
1	5' Flanking	−2272	T/C	
2	5' Flanking	−2070	G/A	
3	Intron 1	25	A/C	
4	Exon 2	308	G/A(Val135Ile)	
5	Intron 2	334	C/G	
6	Intron 4	12	C/T	
7	Intron 5	547	G/A	
8	Exon 7	57	A/T(Arg328Arg)	
9	Intron 9	1231	C/T	
10	Intron 9	1245	G/A	rs1469549
11	Intron 9	2164	C/T	
12	Intron 9	2645	C/del	
13	Intron 9	2646	G/A	
14	Intron 9	3229	G/A	
15	Intron 12	113–114	GG/ins	
16	Intron 13	128	C/T	
17	Intron 13	305	A/G	
18	Intron 14	135	C/G	
19	Intron 14	159	A/T	
20	Intron 15	747	A/G	
21	3' Flanking	48	C/A	rs891506
22	3' Flanking	333	G/A	
23	3' Flanking	1168	G/A	
24	3' Flanking	1719–1721	GTC/del	

ABCB8, ATP-binding cassette, subfamily B, member 8

Amplification of samples and detection of polymorphisms

Total genomic DNAs were isolated from peripheral leukocytes of 48 unrelated Japanese individuals by the standard phenol/chloroform extraction method. On the basis of sequence information from GenBank, we designed polymerase chain reaction (PCR) primers to amplify DNA fragments from all nine genes, excluding repetitive elements, by invoking the REPEAT MASKER computer program (<http://ftp.genome.washington.edu/cgi-bin/RepeatMasker>). PCR experiments and DNA sequencing were performed according to methods described previously (Iida et al. 2001; Saito et al. 2001; Sekine et al. 2001). All SNPs detected by the

Table 1G. Summary of SNPs detected in the *ABCB9* gene

No.	Location	Position	SNP	NCBI SNP ID
1	Intron 1	69	G/C	
2	Intron 1	8873	T/C	
3	Intron 1	8940	G/A	
4	Intron 1	11410	T/C	
5	Intron 1	12863	G/A	
6	Intron 1	19731	G/A	
7	Intron 1	29649	T/C	
8	Intron 1	31793	C/T	
9	Intron 1	37537	C/A	
10	Intron 1	38293	G/A	
11	Intron 1	44661	A/G	
12	Intron 1	49576	C/T	
13	Intron 1	55608	C/T	rs1790094
14	Intron 1	59085	C/T	rs1617156
15	Intron 1	60516	T/G	rs1790106
16	Intron 1	64083	G/A	rs949143
17	Intron 1	64669	C/A	
18	Exon 2	448	G/A(Val121Met)	
19	Intron 7	3364	A/G	
20	Intron 11	113	G/T	
21	Exon 12	370	A/G(3'UTR)	

ABCB9, ATP-binding cassette, subfamily B, member 9

Table 1H. Summary of genetic variations detected in the *ABCB10* gene

No.	Location	Position	Genetic variation	NCBI SNP ID
1	5' Flanking	-424	C/T	
2	Exon 1	491	G/T(Ala150Ser)	
3	Intron 1	37	T/G	
4	Intron 1	217	C/T	
5	Intron 1	405	T/del	
6	Exon 3	185	C/T(Ser301Ser)	
7	Intron 6	1269	C/G	
8	Intron 7	393	G/A	rs446924
9	Intron 9	632	G/A	
10	Intron 10	2373	G/C	
11	Intron 11	108	T/G	
12	Intron 11	2379	T/A	
13	Exon 13	1460	C/A(3'UTR)	rs1053512
14	Exon 13	1571	A/G(3'UTR)	rs1053513

ABCB10, ATP-binding cassette, subfamily B, member 10

PolyPhred Computer Program (Nickerson et al. 1997) were confirmed by sequencing both strands of each PCR product.

Results

We screened 96 Japanese chromosomes for SNPs in nine genes belonging to ATP-binding cassette, subfamily B (*ABCB/MDR/TAP*) by direct DNA sequencing. Resequencing of a total of about 278 kilobases of genomic DNA (70 kb for the *ABCB1* gene, 9 kb for *TAP1*, 6 kb for *TAP2*, 43 kb for *ABCB4*, 33 kb for *ABCB7*, 13 kb for *ABCB8*, 42 kb for *ABCB9*, 17 kb for *ABCB10*, and 45 kb for *ABCB11*) identified a total of 297 SNPs and 29 insertion/deletion polymorphisms. On average, we identified one SNP in every 936 nucleotides.

Figure 1 (A–I) illustrates the location of each variation in the respective genes; detailed information about nucleotide positions and substitutions is summarized in Table 1 (A–I). The numbers and types of genetic variations are summarized in Table 2. Among the 297 SNPs listed here, 50 were identified in *ABCB1* (average of one per 1400 bp), 14 in *TAP1* (1/643 bp), 35 in *TAP2* (1/171 bp), 48 in *ABCB4* (1/896 bp), 13 in *ABCB7* (1/2538 bp), 21 in *ABCB8* (1/619 bp), 21 in *ABCB9* (1/2000 bp), 13 in *ABCB10* (1/1308 bp), and 82 in *ABCB11* (1/549 bp). Of the 29 insertion/deletion polymorphisms, we found 3 in the *ABCB1* gene, 2 in *TAP2*, 2 in *ABCB4*, 2 in *ABCB7*, 3 in *ABCB8*, 1 in *ABCB10*, and 16 in *ABCB11*. Of the 326 genetic variations identified in our screening, 226 (69%), including 29 insertion/deletion polymorphisms, had not been reported previously. Classification of SNPs on the basis of their locations is summarized in Table 3. Among the 297 SNPs, 13 were located in 5' flanking regions, 237 in introns, 37 in exons, and 10 in 3' flanking regions. Among the 37 SNPs detected in exons, 3 were located in 5' untranslated regions (UTRs), 23 in coding regions, and 11 in 3'UTRs.

Among the 37 SNPs detected in exons, 23 were present in coding regions; 10 of those would cause amino acid substitutions: Ala893Ser/Thr in the *ABCB1* gene, (rs2032582); Ile393Val in *TAP1* (rs1057141); Val379Ile (rs1800454), Cys651Arg, Thr665Ala (rs241447), and stop687Gln (rs241448) in *TAP2*; Val135Ile in *ABCB8*; Val121Met in *ABCB9*; Ala150Ser in *ABCB10*; and Val444Ala in *ABCB11*. Of these, 5 were novel (Cys651Arg in *TAP2*, Val135Ile in *ABCB8*, Val121Met in *ABCB9*, Ala150Ser in *ABCB10*, and Val444Ala in *ABCB11*). Among the 13 synonymous SNPs (2 in the *ABCB1* gene, 2 in *TAP2*, 3 in *ABCB4*, 1 in *ABCB8*, 1 in *ABCB10*, and 4 in *ABCB11*), 7 were novel (59Leu and 237Ile in *ABCB4*, 328Arg in *ABCB8*, 301Ser in *ABCB10*, and 36Asp, 90Phe, and 269Tyr in *ABCB11*).

Discussion

We identified 326 genetic variations (297 SNPs and 29 insertion/deletion polymorphisms) by screening the entire genomic regions, except for repetitive sequences, representing nine *ABCB* genes (*ABCB1*, *TAP1*, *TAP2*, *ABCB4*, *ABCB7*, *ABCB8*, *ABCB9*, *ABCB10*, and *ABCB11*) in 96 chromosomes from Japanese volunteers. The polymorphisms published here should be useful for examining relationships between genotypes and susceptibilities to certain diseases.

On average, we identified one SNP in every 936 nucleotides. SNPs were especially frequent in the *TAP2* gene (average of one per 171 bp), but fewer were present in *ABCB1* (1/1400 bp), *ABCB7* (1/2538 bp), *ABCB9* (1/2000 bp), or *ABCB10* (1/1308 bp).

In the *ABCB1* gene, we identified two synonymous polymorphisms and one that was nonsynonymous (Ala893Ser/Thr). The synonymous 1145Ile polymorphism has been

Table 11. Summary of genetic variations detected in the *ABCB11* gene

No.	Location	Position	Genetic variation	NCBI SNP ID
1	5' Flanking	–(2596–2595)	TT/ins	
2	5' Flanking	–1746	G/A	
3	5' Flanking	–(325–314)	(T) _{9–12}	
4	5' Flanking	–135	T/C	
5	Intron 1	511	A/del	
6	Intron 1	581	C/T	
7	Intron 1	1938–1951	(A) _{10–13}	
8	Intron 1	4517	G/A	
9	Intron 1	5651	T/C	
10	Intron 1	12200–12201	CT/del	
11	Intron 1	13023	G/A	
12	Intron 2	739	C/T	
13	Intron 2	921–922	CAGATCTTCTTCAGCTAATTTAGAAATGT/ins	
14	Intron 3	644	G/A	
15	Intron 3	2231	A/G	
16	Intron 3	2406	T/C	
17	Exon 4	10	T/C(Asp36Asp)	
18	Intron 4	434	A/G	
19	Intron 4	518	G/T	
20	Exon 5	120	T/C(Phe90Phe)	
21	Intron 5	320	T/C	
22	Intron 5	16076	T/G	
23	Intron 6	303	G/C	
24	Intron 7	1141	A/G	
25	Intron 8	2463	A/C	
26	Intron 8	2677	A/C	
27	Intron 8	2699	T/A	
28	Exon 9	24	T/C(Tyr269Tyr)	
29	Intron 9	108	A/G	
30	Intron 10	2475	C/A	
31	Intron 10	2478	T/A	
32	Intron 10	2711	C/T	
33	Intron 10	3539	C/G	
34	Intron 10	3623	T/C	
35	Intron 10	3661	A/T	
36	Intron 10	5100	A/G	
37	Intron 10	5292	G/A	
38	Intron 10	5912	A/del	
39	Intron 12	116	G/A	
40	Intron 12	326	G/C	
41	Intron 12	335	A/G	
42	Intron 12	2572	C/T	
43	Exon 13	23	T/C(Val444Ala)	
44	Intron 13	70	C/T	
45	Intron 13	1578–1579	C/ins	
46	Intron 14	32	C/T	
47	Intron 14	80	C/T	
48	Intron 14	439	A/G	
49	Intron 14	1262–1263	T/ins	
50	Intron 14	1283	A/C	
51	Intron 14	1339	G/A	
52	Intron 14	1359	T/C	
53	Intron 14	1480	G/A	
54	Intron 15	370	G/A	
55	Intron 16	550–559	(T) _{9–12}	
56	Intron 17	188	T/G	
57	Intron 17	194	T/G	
58	Intron 17	197–198	T/ins	
59	Intron 17	289–296	(A) ₇ G(A) ₄ /(A) ₁₂ /(A) ₈	
60	Intron 17	1070	C/T	
61	Intron 17	1651	T/C	
62	Intron 17	2226	T/A	
63	Intron 17	2979	T/del	
64	Intron 17	3288	T/G	
65	Intron 17	3289	C/T	
66	Intron 18	97	A/G	
67	Intron 18	98	T/C	
68	Intron 18	892	C/T	
69	Intron 18	2681	A/G	
70	Intron 18	3780	C/G	
71	Intron 18	5741	C/T	

Table 11. Continued

No.	Location	Position	Genetic variation	NCBI SNP ID
72	Intron 18	5882–5883	C/ins	
73	Intron 18	6061	C/A	rs853772
74	Intron 19	127	C/T	rs853773
75	Intron 19	10022	A/del	
76	Intron 19	11880	G/A	rs853785
77	Intron 19	12846	T/G	rs860510
78	Intron 21	322	C/del	
79	Intron 21	–1744	A/G	rs479682
80	Intron 21	–1488	A/G	rs567074
81	Intron 21	–1340	T/C	rs565412
82	Intron 22	257	T/C	
83	Intron 22	320	T/C	rs472614
84	Intron 22	552	G/C	
85	Intron 22	569	G/A	
86	Exon 24	28	A/G(Ala1028Ala)	rs497692
87	Intron 24	413	G/A	rs531772
88	Intron 24	848	T/C	rs527150
89	Intron 24	1201	G/A	rs551754
90	Intron 27	206	A/G	rs519035
91	Intron 27	282	A/G	rs519887
92	Intron 27	349	C/T	rs575671
93	Intron 27	801	A/G	rs579275
94	Exon 28	437	G/A(3'UTR)	rs473351
95	Exon 28	569	G/A(3'UTR)	rs495714
96	Exon 28	621	A/G(3'UTR)	rs496550
97	3' Flanking	243	G/A	
98	3' Flanking	292	C/T	rs478333

ABCB11, ATP-binding cassette, subfamily B, member 11

Table 2. Summary of genetic variations in nine *ABCB* genes

Gene	Total variations			
	Genetic variations	SNPs	Insertion/deletion polymorphisms	New genetic variations
<i>ABCB1</i>	53	50	3	38
<i>TAP1</i>	14	14	0	5
<i>TAP2</i>	37	35	2	6
<i>ABCB4</i>	50	48	2	34
<i>ABCB7</i>	15	13	2	15
<i>ABCB8</i>	24	21	3	22
<i>ABCB9</i>	21	21	0	17
<i>ABCB10</i>	14	13	1	11
<i>ABCB11</i>	98	82	16	78
Total	326	297	29	226

Table 3. Summary of SNPs in nine *ABCB* genes

Number of SNPs							
Gene	5' Flanking	Intron	3' Flanking	Exon			Total
				5'UTR	Coding	3'UTR	
<i>ABCB1</i>	2	42	1	1	3	1	50
<i>TAP1</i>	4	5	3	0	1	1	14
<i>TAP2</i>	2	23	0	1	6	3	35
<i>ABCB4</i>	0	43	1	1	3	0	48
<i>ABCB7</i>	0	13	0	0	0	0	13
<i>ABCB8</i>	2	14	3	0	2	0	21
<i>ABCB9</i>	0	19	0	0	1	1	21
<i>ABCB10</i>	1	8	0	0	2	2	13
<i>ABCB11</i>	2	70	2	0	5	3	82
Total	13	237	10	3	23	11	297

shown to correlate significantly with the expression level of this gene (Hoffmeyer et al. 2000), but it is unlikely that this SNP affects transcription directly. However, the non-synonymous (Ala893Ser/Thr) polymorphism we detected was located in the region encoding the second transmembrane domain and may have phenotypic consequences.

Many SNPs have been reported previously in the *TAP1* and *TAP2* genes, because of the high degree of polymorphism that pertains among *MHC* genes (Colonna et al. 1992; Powis et al. 1992; Jackson and Capra 1993; Jeffreys et al. 2000). However, 5 of the 14 SNPs we detected in the *TAP1* gene and 6 of the 37 genetic variations in *TAP2* were novel. The novel insertion/deletion polymorphism (GG to TGGTGGGGTGGGA) in the 3'UTR of the *TAP2* gene could influence mRNA processing.

For the remaining six genes (*ABCB4*, *ABCB7*, *ABCB8*, *ABCB9*, *ABCB10*, and *ABCB11*), potential associations of *ABCB* genotypes with altered susceptibilities to various diseases have not been examined. However, we detected four novel nonsynonymous polymorphisms (Val135Ile in *ABCB8*, Val121Met in *ABCB9*, Ala150Ser in *ABCB10*, and Val444Ala in *ABCB11*). The first three of these SNPs were close to the N-termini of the respective genes. The fourth was located in a codon close to, and corresponding to the cytoplasmic side of, the functionally important first ATP-binding domain (NCBI Protein ID: NP_003733). Any of these four polymorphisms could have phenotypic consequences.

All data on these SNPs is available on our website (<http://snp.ims.u-tokyo.ac.jp/>).

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