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Identification of 197 genetic variations in six human methyltransferase genes in the Japanese population

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Abstract Methylation is an important event in the biotransformation pathway for many drugs and xenobiotic compounds. We screened DNA from 48 Japanese individuals for single-nucleotide polymorphisms (SNPs) in six methyltransferase (MT) genes (catechol-*O*-MT, *COMT*; guanidinoacetate *N*-MT, *GAMT*; histamine *N*-MT, *HNMT*; nicotinamide *N*-MT, *NNMT*; phosphatidylethanolamine *N*-MT, *PEMT*; and phenylethanolamine *N*-MT, *PNMT*) by direct sequencing of their entire genomic regions except for repetitive elements. This approach identified 190 SNPs and seven insertion/deletion polymorphisms among the six genes. Of the 190 SNPs, 33 were identified in the *COMT* gene, 6 in *GAMT*, 41 in *HNMT*, 8 in *NNMT*, 98 in *PEMT*, and 4 in *PNMT*. Nine were located in 5′ flanking regions, 156 in introns, 10 in exons, and 15 in 3′ flanking regions. These variants may contribute to a more precise understanding of possible correlations between genotypes and disease-susceptibility phenotypes or risk for side effects from drugs.

Key words Single-nucleotide polymorphism (SNP) · Catechol-*O*-methyltransferase gene (*COMT*) · Guanidinoacetate *N*-methyltransferase gene (*GAMT*) · Histamine *N*-methyltransferase gene (*HNMT*) · Nicotinamide *N*-methyltransferase gene (*NNMT*) · Phosphatidylethanolamine *N*-methyltransferase gene (*PEMT*) · Phenylethanolamine *N*-methyltransferase gene (*PNMT*)

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Introduction

Methylation is an important feature of the biotransformation pathway for many drugs and xenobiotic compounds (Weinshilboum 1989). The reaction involves transfer of the activated methyl group of S-adenosyl-L-methionine (AdoMet) to the substrates.

Among the enzymes involved in such reactions, called methyltransferases, catechol-*O*-methyltransferase (COMT; EC 2.1.1.6) catalyzes the transfer of a methyl group from S-adenosylmethionine to catecholamines, a class of molecules that includes the neurotransmitters dopamine, epinephrine, and norepinephrine (Kopin 1985). *O*-methylation drives one of the major pathways for degrading catecholamine transmitters (Zhu and Conney 1998). In addition to its role in the metabolism of endogenous substances, COMT is important for metabolizing catechol drugs such as L-DOPA (Lautala et al. 2001). COMT is found in two forms in tissues, a cytoplasmic soluble form (S-COMT) and a membrane-bound form (MB-COMT) located in the rough endoplasmic reticulum (Salminen et al. 1990; Bertocci et al. 1991; Lundström et al. 1991; Ulmanen et al. 1997). The soluble enzyme appears to be the predominant form of COMT in most tissues. However, in humans the membrane-bound enzyme predominates in the brain and adrenal medulla, and also in pheochromocytomas (Tenhunen et al. 1994; Eisenhofer et al. 1998). The MB-COMT transcript differs from the S-COMT transcript in that the former contains a longer 5′ untranslated region (UTR) and its translation-start site lies 50 codons upstream from that of S-COMT (Tenhunen et al. 1994).

Lotta et al. (1995) identified a G to A polymorphism in the COMT gene at codon 158, which leads to substitution of methionine for valine. This polymorphism is associated with a three- to fourfold difference in enzymatic activity (Lachman et al. 1996); i.e., homozygosity for the A allele, $COMT^{Met}$, is associated with low enzyme activity and thermolability. Individuals who are homozygous for the normal (valine) allele display relatively high activity and heat stability of COMT; heterozygotes usually

display intermediate levels of COMT activity and heat stability.

Guanidinoacetate methyltransferase (GAMT; EC2.1.1.2) catalyzes the last step of creatine biosynthesis in mammals. Creatine is an important metabolite for cellular energy metabolism in a variety of tissues, e.g., skeletal, cardiac and smooth muscle, and brain (Walker 1979; Wallimann and Hemmer 1994). GAMT deficiency causes an inborn error in creatine biosynthesis (Stöckler et al. 1994, 1996a, 1996b, 1997; Carducci et al. 2000; Ilas et al. 2000). Deficiency of GAMT readily explains the accumulation of guanidinoacetate and the depletion of the total creatine pool in affected individuals.

Histamine *N*-methyltransferase (HNMT; EC 2.1.1.8) catalyzes a major metabolic pathway of histamine, a known neurotransmitter (Schwartz et al. 1991). Histamine plays an important role in allergic responses and is involved in regulating secretion of gastric acid (Wasserman 1983; Loiselle and Wollin 1993). Biochemical genetic studies of HNMT in human red blood cells demonstrated fivefold individual differences in the level of this enzyme's activity, mainly reflecting common genetic polymorphisms (Scott et al. 1988; Price et al. 1993). Preuss et al. (1998) demonstrated a C314T polymorphism in exon 4 in the *HNMT* gene that resulted in a Thr105Ile amino acid substitution; they found that the T314 allele was associated with decreased levels of both HNMT enzymatic activity and immunoreactive protein. Therefore, the presence of a T314 allele would be expected to result in reduced histamine metabolism and increased bronchoconstriction. A number of investigators have examined potential associations of *HNMT* genotypes with altered susceptibilities to various diseases such as asthma (Yan et al. 2000a) and schizophrenia (Yan et al. 2000b).

Nicotinamide *N*-methyltransferase (NNMT; EC 2.1.1.1) catalyzes the *N*-methylation of nicotinamide and other pyridines to form pyridinium ions (Rini et al. 1989). Several diseases have been associated with abnormal nicotinamide metabolism that results in elevated levels of *N*-methylnicotinamide; these include idiopathic Parkinson's disease (Williams et al. 1993) and hepatic cirrhosis (Cuomo et al. 1994). NNMT enzymatic activity in human liver varies in a fivefold range, with a bimodal frequency distribution (Rini et al. 1989). Phenotypic differences in NNMT activity could reflect genetic polymorphisms; however, no sequence differences were seen when investigators compared cDNAs of individuals with high and low NNMT activity (Smith et al. 1998; Yan et al. 1999).

Phosphatidylethanolamine *N*-methyltransferase (PEMT; EC2.1.1.17) converts phosphatidylethanolamine to phosphatidylcholine in mammalian liver (Vance et al. 1997). Immunolocalization experiments have detected two isoforms of PEMT, encoded by a single gene (Walkey et al. 1997). PEMT1, which constitutes the majority of PEMT activity, is located at the endoplasmic reticulum (Vance and Ridgway 1988), while PEMT2 is found on mitochondria-associated membranes (Cui et al. 1993). The activity of PEMT is very low when hepatocytes are undifferentiated and proliferating rapidly. At birth, as hepatocyte proliferation slows and final differentiation begins, PEMT2 expression increases dramatically. When hepatocyte proliferation was induced in adult rats by partial hepatectomy, PEMT2 expression was inhibited (Vance et al. 1997). Those results suggested that this enzyme might play a role in the regulation of hepatocyte growth and cell division (Walkey et al. 1999).

Phenylethanolamine *N*-methyltransferase (PNMT; EC 2.1.1.28) catalyzes the final step in the catecholamine biosynthetic pathway, converting norepinephrine to epinephrine (Axelrod and Weinshilboum 1972). It is primarily expressed in the adrenal medulla and the retina (Baetge et al. 1988). Its activity increases after stress in response to glucocorticoids and neuronal stimulation (Betito et al. 1994). Since defects in epinephrine metabolism have been implicated in the etiology of attention-deficit hyperactivity disorder (ADHD), aggression, and anxiety (Klinteberg and Magnusson 1989; Pliszka et al. 1994; Girardi et al. 1995; Hanna et al. 1996), PNMT is considered to be a candidate for a range of psychiatric disorders.

To investigate the nature of apparent genotype/phenotype correlations for methyltransferases more precisely, we began by searching for additional single-nucleotide polymorphisms (SNPs) in the six methyltransferase genes described above, including their promoter regions and introns but excluding repetitive elements. We report here a total of 197 genetic variations, of which 144 have not been reported before.

Materials and methods

Exon–intron boundaries of the *COMT*, *GAMT*, *HNMT*, *NNMT*, *PEMT*, and *PNMT* genes were defined by comparing genomic sequences with mRNA sequences. Accession numbers of the genomic sequences obtained for this study are as follows:

COMT (AC000080.2), *GAMT* (NT_000879.1),
HNMT (AC019304.3), *NNMT* (AC019290.3),
PEMT (AC020558.3), *PNMT* (AC040933.3)

Accession numbers of the mRNA sequences are as follows:

COMT (NM_000754.2, M65213.1),
GAMT (NM_000156.3), *HNMT* (NM_006895.1),
NNMT (U20970.1, U20971.1), *PEMT* (NM_007169.1, AF113126.1, AF176807.1), *PNMT* (NM_002686.1)

Amplification of samples. 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 six genes, excluding repetitive elements, by invoking the Repeat Masker computer program in the manner described by Seki et al. (2000). We used 60 ng of pooled DNA (20 ng from three individuals) for each PCR experiment. The reactions were performed as follows: initial denaturation at 94°C for 2 min,

followed by 35 cycles of amplification at 94°C for 30s, annealing at 60°C for 1min, and extension for 1min.

Direct sequencing and detection of polymorphisms. Products obtained from the PCR experiments were used as templates for direct sequencing and detection of SNPs, by the fluorescent dye-terminator cycle sequencing method (ABI PRISM Big Dye Terminator Cycle Sequencing Ready Reaction Kit; PerkinElmer, Foster City, CA, USA). These procedures were carried out according to methods we have described elsewhere (Ohnishi et al. 2000; Seki et al. 2000; Yamada et al. 2000).

Results

We screened 96 Japanese chromosomes for SNPs in six methyltransferase genes by direct DNA sequencing of a total of 103 kilobases of genomic DNA (11kb for the *COMT* gene, 5kb for *GAMT*, 27kb for *HNMT*, 5kb for *NNMT*, 52kb for *PEMT*, and 3kb for *PNMT*). In these regions we identified a total of 190 SNPs and seven insertion/deletion polymorphisms among the six genes examined. On average, we identified one SNP in every 542 nucleotides.

Fig. 1 (A–F) illustrates the location of each variation in the respective genes; detailed information about nucleotide positions and substitutions is summarized in Table 1 (A–F). Among the 190 SNPs listed there, 33 were identified in the *COMT* gene (average of one per 333bp), 6 in *GAMT* (1/833bp), 41 in *HNMT* (1/659bp), 8 in *NNMT* (1/625bp), 98 in *PEMT* (1/531bp), and 4 in *PNMT* (1/750bp). Of the seven insertion/deletion polymorphisms, one was present in the *COMT* gene, four in *HNMT*, and two in *PEMT*.

Among the 33 SNPs found in the *COMT* gene, 5 were present in the 5′ flanking region, 21 in introns, 4 in exons, and 3 in the 3′ flanking region. Two of the exonic SNPs would cause substitution of amino acids (Ala72Ser in exon 3 and Val158Met in exon 4); both of these variants have been reported previously in the National Center for Biotechnology Information (NCBI) dbSNP database (rs6267 and rs165688, respectively). An insertion/deletion polymorphism was found in exon 6 (rs362204) corresponding to the 3′ untranslated region (3′ UTR). Of the 34 genetic variations we found in this gene, 17 were novel.

Of the six SNPs found in the *GAMT* gene, four were present in introns and two in the 3′ flanking region; none caused amino-acid substitutions. We were unable to find any SNPs in the 5′ flanking region or in the regions between exons 2 and 5.

As regards the *HNMT* gene, one of the 41 SNPs was present in the 5′ flanking region, 31 were in introns (20 of them in intron 5), 3 in exons, and 6 in the 3′ flanking region. Of the three exonic SNPs, the one at codon 105 in exon 4 would cause a substitution of isoleucine for threonine. This variant had been reported previously in the NCBI dbSNP database (rs1801105). Two known SNPs were identified in the 3′ UTR in exon 6 (rs1050891 and rs1050900).

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

No.	Location	Position[a]	Genetic variation	NCBI SNP ID
1	5′ Flanking	−1287	G/A	
2	5′ Flanking	−1217	G/A	
3	5′ Flanking	−503	G/A	
4	5′ Flanking	−425	C/T	
5	5′ Flanking	−277	C/T	
6	Intron 1	12058	G/A	
7	Intron 1	12070	A/G	
8	Intron 1	12074	G/A	rs1055503
9	Intron 1	17883	T/G	rs174694
10	Intron 1	17963	T/C	rs174695
11	Intron 1	18831	C/T	
12	Intron 2	51	C/G	rs165656
13	Intron 2	201	C/T	rs165722
14	Intron 2	832	G/C	
15	Intron 2	1140	A/G	rs6269
16	Exon 3	186	T/C(His62His)	rs4633
17	Exon 3	214	G/T(Ala72Ser)	rs6267
18	Intron 3	90	A/G	
19	Intron 3	425	T/G	
20	Intron 3	671	G/A	
21	Intron 3	676	G/T	
22	Exon 4	119	C/G(Leu136Leu)	rs4818
23	Exon 4	183	G/A(Val158Met)[b]	rs165688
24	Intron 5	75	G/C	
25	Intron 5	310	C/T	
26	Intron 5	346	G/A	
27	Intron 5	679	C/T	rs165737
28	Intron 5	739	G/A	rs165774
29	Intron 5	2634	T/C	rs174699
30	Intron 5	3023	G/A	
31	Exon 6	212	C/del(3′ UTR)	rs362204
32	3′ Flanking	231	A/G	rs165599
33	3′ Flanking	473	T/C	rs165728
34	3′ Flanking	1081	C/A	rs9265

COMT, catechol-*O*-methyltransferase; NCBI, National Center for Biotechnology Information; SNP, single-nucleotide polymorphism; UTR, untranslated region

[a]For SNPs in the 5′ flanking, intron, or 3′ flanking regions, nucleotide positions are counted from the first intronic nucleotide at the exon–intron junction; for SNPs in the exon region, from the first exonic nucleotide

[b]SNP previously reported by Lotta et al. (1995)

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

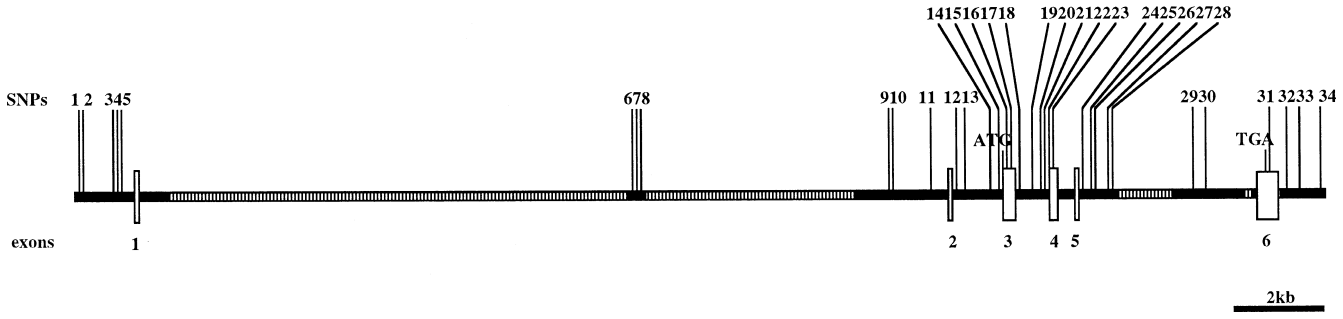
No.	Location	Position[a]	SNP	NCBI SNP ID
1	Intron 1	429	G/A	
2	Intron 1	529	T/C	rs266806
3	Intron 1	616	T/C	rs266807
4	Intron 5	1411	G/A	
5	3′ Flanking	136	A/G	rs266813
6	3′ Flanking	626	G/A	rs740446

GAMT, guanidinoacetate *N*-methyltransferase

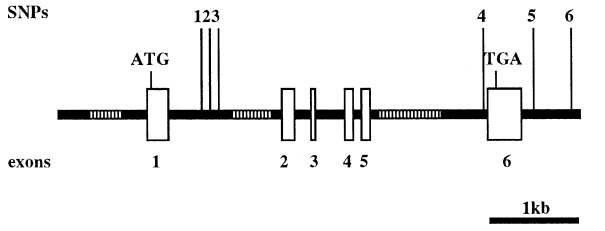
Among the eight SNPs we found in the *NNMT* gene, one was in the 5′ flanking region, six were in introns, and one was in the 3′ flanking region.

In the *PEMT* gene, of the 98 SNPs found, 93 were intronic; 3 were in exons, and 2 were in the 3′ flanking region. Two of the exonic SNPs would cause amino acid substitutions (Val58Leu in exon 5 and Ala175Thr in exon

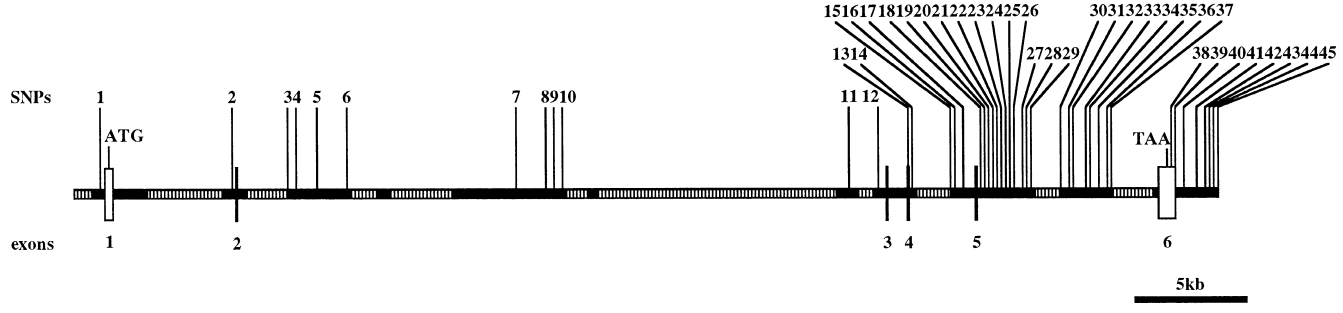
A *Catechol-O-methyltransferase (COMT)*



B *Guanidinoacetate N-methyltransferase (GAMT)*



C *Histamine N-methyltransferase (HNMT)*



D *Nicotinamide N-methyltransferase (NNMT)*

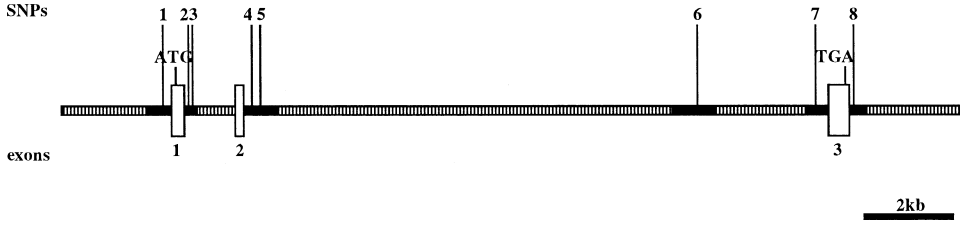
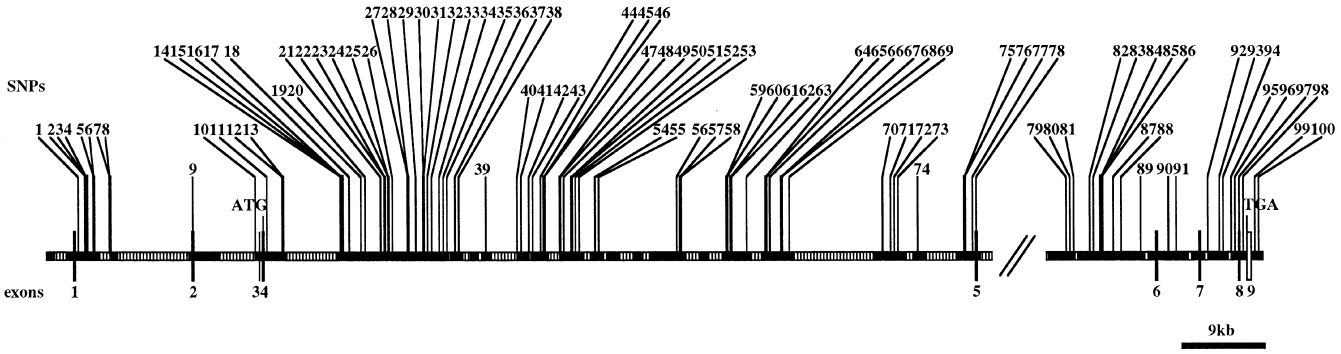


Fig. 1. Locations of single-nucleotide polymorphisms (*SNPs*) in the *COMT* (**A**), *GAMT* (**B**), *HNMT* (**C**), *NNMT* (**D**), *PEMT* (**E**), and *PNMT* (**F**) genes, indicated by *vertical lines*. *Open boxes* represent exons; *hatching* on the chromosomes indicates regions of repetitive elements; (*ATG*) and (*TGA* or *TAA*), initiation and stop codons, respectively

E ***Phosphatidylethanolamine N-methyltransferase (PEMT)***



F ***Phenylethanolamine N-methyltransferase (PNMT)***

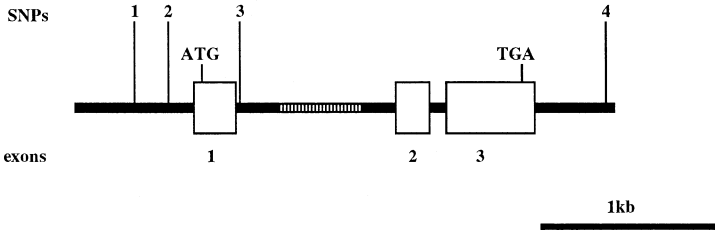


Fig. 1. Continued

8). Both of those variants have been reported in the NCBI dbSNP database (rs897453 and rs1918248, respectively).

Among the four SNPs detected in the *PNMT* gene, two were located in the 5′ flanking region, one in an intron, and one in the 3′ flanking region. Both SNPs in the 5′ flanking region were reported previously (Wu and Comings 1999).

Discussion

We identified 197 genetic variations (190 SNPs and 7 insertion/deletion polymorphisms) by screening the entire genomic regions, except for repetitive sequences, encoding six methyltransferase genes (*COMT*, *GAMT*, *HNMT*, *NNMT*, *PEMT*, and *PNMT*) in the Japanese population. Of the 197 genetic variations detected, 144 (73%) have not been reported before (17 in the *COMT* gene, 2 in *GAMT*, 32 in *HNMT*, 6 in *NNMT*, 86 in *PEMT*, and 1 in *PNMT*).

When we investigated SNPs in these six genes using the NCBI dbSNP database, we noticed that 18 exonic SNPs in the archive were absent from our 96-chromosome Japanese population sample. Of those "missing" SNPs, eight had been reported in the *COMT* gene: rs1805052 (5′ UTR) in exon 1; rs6270 (Cys34Ser) and rs740602 (Gln73Gln) in exon 3; rs769223 (Ala134Ala) in exon 4; rs769224 (Pro199Pro) and rs165631 (Leu203Leu) in exon 5; and rs12814 (3′ UTR) and rs14968 (3′ UTR) in exon 6. Three were in the *GAMT* gene: rs1050914 (Val165Phe; exon5) and rs659455 and rs659460 in the 3′ UTR of exon 6. The archive also showed one [rs1050207 (Thr245Pro)] in exon 3 of the *NNMT* gene and six in exon 3 of the *PNMT* gene [rs5638 (Lys152Llys), rs5639 (Ser188Cys), rs5640 (Leu211His), rs5641 (Leu217Gln), rs5642 (Arg254His), and rs5643 (Trp276Arg)]. Since Yan et al. (1999) detected no SNPs or insertion/deletion events within either exons or 5′ flanking regions of *NNMT* and Smith et al. (1998) reported the same result, the archived SNPs may be very rare substitutions or limited to some ethnic group, or they may reflect sequencing errors.

In the *COMT* gene, we identified two SNPs that caused amino acid substitutions, Ala72Ser in exon 3 and Val158Met in exon 4. Lachman et al. (1996) demonstrated that the Val158Met polymorphism in exon 4 was associated with a three- to fourfold difference in the enzyme's activity; however, it is unclear whether the Ala72Ser polymorphism in exon 3 would alter enzymatic activity. Transcription of the *COMT* gene is controlled by two distinct promoter regions (Tenhunen et al. 1994); the proximal promoter (P1), located between the S- and MB-*COMT* ATG start codons, regulates expression of S-*COMT* mRNA; MB-*COMT* transcription is controlled by the distal promoter (P2). We

Table 1C. Summary of genetic variations detected in the *HNMT* gene

No.	Location	Position[a]	Genetic variation	NCBI SNP ID
1	5′ Flanking	−211	C/T	
2	Intron 1	5409	A/G	
3	Intron 2	2561	A/G	
4	Intron 2	2895	A/C	
5	Intron 2	3977	G/A	
6	Intron 2	5296	T/C	
7	Intron 2	13317	C/T	
8	Intron 2	14682	A/del	
9	Intron 2	15047	C/T	rs1020678
10	Intron 2	15406	G/A	
11	Intron 2	28943	A/G	
12	Intron 2	30390	T/A	rs973013
13	Exon 4	16	C/T(Thr105Ile)[c]	rs1801105
14	Intron 4	49	A/G	
15	Intron 4	1404	G/T	rs1455167
16	Intron 4	1942–1943	A/ins	
17	Intron 4	2405	C/A	
18	Intron 5	80–81	TT/ins	
19	Intron 5	235	T/C	
20	Intron 5	702–703	AT/ins	
21	Intron 5	749	T/G	
22	Intron 5	1101	T/G	
23	Intron 5	1137	G/A	
24	Intron 5	1348	C/G	
25	Intron 5	1517	A/G	rs1378321
26	Intron 5	1673	C/G	
27	Intron 5	2022	C/T	
28	Intron 5	2253	A/G	rs1455162
29	Intron 5	2285	G/C	
30	Intron 5	4159	C/T	
31	Intron 5	4501	C/G	
32	Intron 5	4729	G/T	rs993891
33	Intron 5	5251	C/G	
34	Intron 5	5296	A/C	rs1455159
35	Intron 5	5802	C/T	
36	Intron 5	6189	G/C	
37	Intron 5	6297	T/A	
38	Exon 6	416	A/G(3′ UTR)	rs1050891
39	Exon 6	574	A/T(3′ UTR)	rs1050900
40	3′ Flanking	458	C/T	
41	3′ Flanking	993	G/A	
42	3′ Flanking	1402	C/T	rs1455158
43	3′ Flanking	1516	T/C	rs1455157
44	3′ Flanking	1575	C/G	rs1455156
45	3′ Flanking	1793	G/A	

HNMT, histamine *N*-methyltransferase; del, deletion; ins, insertion
[c]SNP previously reported by Preuss et al. (1998)

Table 1D. Summary of SNPs detected in the *NNMT* gene

No.	Location	Position[a]	SNP	NCBI SNP ID
1	5′ Flanking	−228	A/T	
2	Intron 1	44	T/C	
3	Intron 1	149	A/G	
4	Intron 2	158	G/A	rs1941404
5	Intron 2	433	T/C	rs1894030
6	Intron 2	10826	C/T	
7	Intron 2	13630	T/C	
8	3′ Flanking	71	A/G	

NNMT, nicotinamide *N*-methyltransferase

found five SNPs in the region corresponding to the P2 promoter, and one (intron 2 +1140, rs6269) in the P1 promoter region. These SNPs may have some effect on transcription of the MB-*COMT* and S-*COMT* mRNAs. Since COMT is a major conjugation enzyme for the catechol iatrogenes and its genetic polymorphism is thought to correlate with its enzymic activity, a number of investigators have examined potential associations of *COMT* genotypes with altered susceptibilities to various diseases, for example breast cancer (Lavigne et al. 1997; Millikan et al. 1998; Thompson et al. 1998; Huang et al. 1999) and ovarian cancer (Goodman et al. 2000). Some of these investigators have reported an association of the *COMT* genotype with risk for breast cancer, but others have found no relation between genotype and risk of either breast or ovarian cancer. The novel SNPs in *COMT* published here should contribute to further explorations of potential relationships between *COMT* allelic status and susceptibility to certain diseases.

In the *GAMT* gene, we found no SNP that would cause an amino acid substitution. Although mutations causing severe GAMT deficiency have been reported (Stöckler et al. 1994, 1996a, 1996b, 1997; Carducci et al. 2000; Ilas et al. 2000), no SNPs that may correlate with disease-susceptibility phenotypes have been reported to date.

In the *HNMT* gene, one SNP we detected in exon 4 would substitute an amino acid, Thr105Ile. This polymorphism, which was reported previously (Preuss et al. 1998), has been associated with altered HNMT activity. In fact, potential associations between *HNMT* genotypes and susceptibility to asthma (Yan et al. 2000a) and schizophrenia (Yan et al. 2000b), have been investigated, but without conclusive results. Hence, the novel SNPs reported here may serve as additional tools for further exploration of possible relationships between an individual's *HNMT* genotype and susceptibility to certain diseases.

As regards the *NNMT* gene, we found no SNP causing an amino acid substitution, as previous reports have indicated as well (Smith et al. 1998; Yan et al. 1999). However, we detected one SNP in the 5′ flanking region that might be capable of influencing transcriptional efficiency.

In the *PEMT* gene, we found two SNPs that would substitute amino acids, Val58Leu in exon 5 and Ala175Thr in exon 8, but it is unclear whether these changes would alter the enzyme's activity. However, the eight SNPs and two insertion/deletion polymorphisms we identified in intron 1, exon 2, and intron 2 might influence transcriptional efficiency.

Although we found no SNP in the *PNMT* gene that would cause amino acid substitution, we did identify two in the 5′ flanking region. Since PNMT may be an important component of the mammalian stress response and is considered to be a candidate for a wide range of psychiatric disorders, the two SNPs in the putative promoter region may be useful for investigating a possible role of this gene in mental illnesses.

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

No.	Location	Position[a]	Genetic variation	NCBI SNP ID	No.	Location	Position[a]	Genetic variation	NCBI SNP ID
1	Intron 1	297–299	TGT/del		51	Intron 4	23627	C/T	
2	Intron 1	817	A/G		52	Intron 4	23941	G/A	
3	Intron 1	830	G/A		53	Intron 4	24091	G/T	
4	Intron 1	1035	T/C		54	Intron 4	25348	G/A	
5	Intron 1	1573	C/T		55	Intron 4	25603	G/A	
6	Intron 1	1759	A/G		56	Intron 4	31540	T/C	
7	Intron 1	2768	C/A		57	Intron 4	31637	G/A	
8	Intron 1	2785	T/C		58	Intron 4	31642	G/A	
9	Exon 2	162	C/T(5′ UTR)		59	Intron 4	35593	G/A	
10	Intron 2	4598	T/del		60	Intron 4	35647	C/A	
11	Intron 4	39	C/T		61	Intron 4	35862	C/T	
12	Intron 4	1317	G/A		62	Intron 4	35882	T/G	
13	Intron 4	1355	A/C		63	Intron 4	37141	T/C	
14	Intron 4	5925	C/T		64	Intron 4	38862	C/G	
15	Intron 4	6028	G/C		65	Intron 4	38872	G/T	
16	Intron 4	6078	C/T		66	Intron 4	39140	C/T	
17	Intron 4	6089	A/G		67	Intron 4	39635	G/T	
18	Intron 4	6379	G/A		68	Intron 4	39713	C/T	
19	Intron 4	7339	C/T		69	Intron 4	40436	G/A	rs936108
20	Intron 4	7619	A/G		70	Intron 4	47485	C/T	
21	Intron 4	8858	T/G		71	Intron 4	48131	G/A	
22	Intron 4	9029	G/A		72	Intron 4	48558	C/G	
23	Intron 4	9056	C/T		73	Intron 4	48702	G/A	
24	Intron 4	9512	A/G		74	Intron 4	50302	T/C	
25	Intron 4	9523	T/C		75	Intron 4	54102	A/T	rs746900
26	Intron 4	9622	G/A		76	Intron 4	54220	G/A	rs746899
27	Intron 4	10776	G/A		77	Intron 4	54371	G/A	
28	Intron 4	10912	G/C		78	Exon 5	79	G/C(Val58Leu)	rs897453
29	Intron 4	11590	G/C		79	Intron 5	−6796	A/C	rs748196
30	Intron 4	12090	G/C		80	Intron 5	−6636	T/C	rs897456
31	Intron 4	12263	G/A		81	Intron 5	−6448	G/A	
32	Intron 4	12448	G/A		82	Intron 5	−5218	C/G	
33	Intron 4	12730	C/T	rs744925	83	Intron 5	−4824	G/A	
34	Intron 4	13240	T/C	rs897451	84	Intron 5	−4249	C/A	
35	Intron 4	13494	T/C		85	Intron 5	−4230	C/T	
36	Intron 4	13817	A/G		86	Intron 5	−4182	G/A	
37	Intron 4	14773	C/T		87	Intron 5	−3369	G/C	
38	Intron 4	14951	G/A	rs1531098	88	Intron 5	−2625	C/T	
39	Intron 4	16896	T/C		89	Intron 5	−1200	A/T	
40	Intron 4	19439	G/A		90	Intron 6	606	A/G	
41	Intron 4	19557	C/T		91	Intron 6	1229	G/A	rs1242390
42	Intron 4	20051	A/G		92	Intron 7	716	C/G	
43	Intron 4	20816	C/T		93	Intron 7	1537	G/A	
44	Intron 4	21196	C/G		94	Intron 7	1718	T/C	rs1020697
45	Intron 4	21528	G/T		95	Intron 7	2695	C/T	
46	Intron 4	21596	C/T		96	Intron 7	3039	A/T	rs1918249
47	Intron 4	22672	C/T		97	Exon 8	56	G/A(Ala175Thr)	rs1918248
48	Intron 4	22713	A/T		98	Intron 8	140	C/T	
49	Intron 4	23010	G/A		99	3′ Flanking	179	C/T	
50	Intron 4	23588	C/T		100	3′ Flanking	394	A/G	rs1020696

PEMT, phosphatidylethanolamine *N*-methyltransferase

Table 1F. Summary of SNPs detected in the PNMT gene

No.	Location	Position[a]	SNP	NCBI SNP ID
1	5′ Flanking	−367	G/A[d]	
2	5′ Flanking	−161	G/A[d]	rs876493
3	Intron 1	35	G/T	
4	3′ Flanking	438	G/A	rs407307

PNMT, phenylethanolamine *N*-methyltransferase
[d]SNPs previously reported by Wu et al. (1999)

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