Case–control study and meta-analysis of SULT1A1 Arg<sup>213</sup>His polymorphism for gene, ethnicity and environment interaction for cancer risk

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Cytosolic sulphotransferase SULT1A1 plays a dual role in the activation of some carcinogens and inactivation of others. A functional polymorphism leading to Arg<sup>213</sup>His substitution (SULT1A1*<sup>2</sup>) affects its catalytic activity and thermostability. To study the association of SULT1A1*<sup>2</sup> polymorphism with tobacco-related cancers (TRCs), a case–control study comprising 132 patients with multiple primary neoplasm (MPN) involving TRC and 198 cancer-free controls was carried out. One hundred and thirteen MPN patients had at least one cancer in upper aerodigestive tract including lung (UADT-MPN). SULT1A1*<sup>2</sup> showed significant risk association with UADT-MPN (odds ratio (OR) = 5.50, 95% confidence interval (CI): 1.09, 27.7). Meta-analysis was conducted combining the data from 34 published studies that included 11,962 cancer cases and 14,673 controls in diverse cancers. The SULT1A1*<sup>2</sup> revealed contrasting risk association for UADT cancers (OR = 1.62, 95% CI: 1.12, 2.34) and genitourinary cancers (OR = 0.73, 95% CI: 0.58, 0.92). Furthermore, although SULT1A1*<sup>2</sup> conferred significant increased risk of breast cancer to Asian women (OR = 1.91, 95% CI: 1.08, 3.40), it did not confer increased risk to Caucasian women (OR = 0.92, 95% CI: 0.71, 1.18). Thus risk for different cancers in distinct ethnic groups could be modulated by interaction between genetic variants and different endogenous and exogenous carcinogens.

Keywords: multiple primary neoplasm; SULT1A1; meta-analysis; single nucleotide polymorphism; tobacco related cancer; upper aerodigestive tract cancer.

Tobacco-related cancer (TRC) accounts for almost half the global burden of cancer and arises from a complex gene–environment interaction. Traditionally, only lung, oesophageal, and head and neck cancers were described as TRCs. However, based on several studies, the International Association of Research in Cancer (IARC) has broadened the definition of TRC to include carcinoma of the cervix, bladder, stomach, kidney, pancreas, liver and myeloid leukaemia (Doll, 1999; IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, 2004; Hung et al., 2005). Although prolonged exposure to tobacco with or without other carcinogens plays a central role in the genesis of these cancers, various host genetic factors could significantly modulate the risk of developing TRC.

Several genetic alterations in the genes coding for xenobiotic-metabolising enzymes (XMEs), DNA repair, cell cycle regulation and apoptotic pathway confer a low-penetrance genetic susceptibility to tobacco carcinogens (Kotnis et al., 2005). There is a large body of evidence, including meta-analyses to support the association of various isoforms of glutathione-S transferases (GSTs), cytochrome P450 and N-acetyl transferase with tobacco carcinogenesis. Although sulphotransferase (SULT) enzymes could play an equally important role in detoxifying tobacco carcinogens, there are very few, mostly inconclusive, studies examining the association of genetic alteration in the genes coding for this superfamily of multifunctional enzymes with TRC (Seth et al., 2000; Hung et al., 2004; Sellers et al., 2005; Dandara et al., 2006).

Sulphotransferase enzymes catalyse sulphation by transferring sulphonate (sulphuryl) group from cofactor 3'-phosphoadenosine 5'-phosphosulphate to a nucleophilic acceptor substrate to form either a sulphate ester or a sulphamate. These sulphate conjugates are more polar and less reactive than the parent compound and facilitate their excretion (Glatt et al., 2001). However, some sulpho conjugates are strong electrophiles and may covalently bind to DNA and proteins (Glatt et al., 2001).

SULT1A1 gene is one of the most important and well-studied members of the SULT family and is abundant in a wide variety of tissues. SULT1A1 plays a major role in biotransformation of numerous substrates including several carcinogens, neurotransmitters, steroid hormones and drugs (Raftogianis et al., 1999; Hildebrandt et al., 2007). Although sulphonation is an important property of SULT1A1 in the inactivation of carcinogens, it also

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plays an important role in toxification of dietary and environmental mutagens (Glatt et al, 2001; Al-Buheissi et al, 2006). Of the various polymorphisms in SULT1A1, the Arg→His polymorphism at position 213, in exon 7 (SULT1A1*2), has a twofold lower catalytic activity and thermo stability than its high-activity Arg213 counterpart as demonstrated in platelet cytosol (Raffogianis et al, 1997; Nowell et al, 2000; Nagar et al, 2006). Although several reports show risk association of the SULT1A1 variant with different cancers (Sun et al, 2005; Dandara et al, 2006; Pachouri et al, 2006; Fan et al, 2007; Lilla et al, 2007; Bardakci et al, 2008), others show either no effect (Sellers et al, 2005) or a protective effect (Cheng et al, 2005).

In this case–control study, we have examined the association of SULT1A1 Arg213His polymorphism with tobacco carcinogenesis using a unique group of individuals with multiple tobacco-related primary cancers and have used a meta-analysis approach to confirm our findings. Considering that His213 variant of SULT1A1 has a lower activity than Arg213 variant (Raffogianis et al, 1997; Nagar et al, 2006), we hypothesise that if tobacco carcinogenesis is significantly modulated by the Arg213His polymorphism, it would demonstrate a significant association with TRC and this association may be stronger in individuals with multiple primary TRC.

**MATERIALS AND METHODS**

**Study subjects**

A registry of patients with multiple primary cancers or familial cancers was established at the Tata Memorial Hospital, Mumbai, in 1996 by one of the authors (RS). From this registry, 132 consecutive multiple primary neoplasm (MPN) patients where one or both the primaries were tobacco related, and their genomic DNA and consent were available, were taken up for this study. Histological or cytological confirmation of each primary cancer was available and each of the cancers was classified as TRC or non-TRC as per the IARC criteria (IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, 2004). There was no restriction for age at diagnosis, gender or carcinogen exposure. For defining two cancers as distinct multiple primaries, modified Hong’s criteria (Hong et al, 1990) was used, which states that – (a) there is >2 cm of normal intervening mucosa between two primaries in head and neck region; (b) lung as second primary if present, should be of different histology, or be solitary and with characteristic radiology of lung cancer; and (c) there is no evidence of haematogenous spread. Bilateral cancers in paired organs such as breast, ovaries or kidneys were not classified as MPN.

Majority of the MPN cases in the registry hailed from the western and northern parts of India. The cancer-free controls (n = 198) were volunteers who consented to donate blood or buccal washes for the study. The controls were also from the same region and were free of any cancer or pre-cancerous condition. They were either visiting our hospital in the Preventive Oncology region and were free of any cancer or pre-cancerous condition.

**DNA extraction and genotyping**

Genomic DNA was extracted from peripheral blood/mouthwash samples using phenol chloroform method standardised in our laboratory (Koppikar and Mulherkar, 2006). PCR for SULT1A1 genotyping followed by RFLP using HaeII restriction enzyme was carried out as described by Wang et al (2002). The authenticity of the PCR products was confirmed by sequencing at least five PCR products at random on an automated DNA sequencer (ABI Prism 3100 Avant) using the Big Dye terminator kit (ABI Prism, Foster City, CA, USA) as per the manufacturer’s instructions.

**Identification and analysis of studies for meta-analysis**

PUBMED searches were conducted to identify studies on SULT using the search words ‘SULT1A1, SULT AND polymorphism’ and ‘SULT AND cancer’. The inclusion criteria were case–control studies examining associations of SULT1A1 Arg213His polymorphism either alone or in combination with other genes, published until July 2007. For every study, publication date, country of origin, demographics, genotyping methodology, ethnicity, source and genotype frequency of study subjects were reviewed. In case of missing information, the authors were contacted and requested to provide the data. One study was excluded as all the required information could not be obtained (Peng et al, 2003). Genotyping studies on only cancer cases or exclusively on healthy subjects were excluded as comparison of cancer patients with matched controls was a prerequisite for studying association of a particular genotype with cancer risk (Nowell et al, 2002b, 2005; Magagnotti et al, 2003; Sparks et al, 2004; Shatalova et al, 2005; Grabinski et al, 2006).

**Statistical analysis**

The risk (odds ratio, OR) was estimated by comparison of the variant 213His genotype vs the wild-type 213Arg allele using dominant model ((Arg/His + His/His) vs Arg/Arg), recessive model (His/His vs (Arg/His + Arg/Arg)) as well as the extreme model (His/His vs Arg/Arg). The risk was adjusted for age and habit using unconditional logistic regression analysis using SPSS v14.0. Hardy–Weinberg equilibrium in the controls was evaluated for each study using χ2 test. For each genetic contrast, the between-study heterogeneity was estimated across all eligible comparisons using Q statistics. Funnel plots and Egger’s test were used to assess potential publication bias, which results from non-publication of small studies with negative results (Egger et al, 1997). This test detects funnel plot asymmetry by determining whether the intercept deviates significantly from zero in a regression of the standardised effect estimates against their precision. Influence analysis was also carried to assess whether summary OR was driven by any one study in the recessive model of meta-analysis (Sterne et al, 2002), Stratification by ethnicity (Asian, Caucasian and Others), total study size of cases and controls (up to 500 or more), Hardy–Weinberg equilibrium (yes/no), primary site (upper aerodigestive tract (UADT) and lung, breast, colorectal, genitourinary and other sites), source of control (population/hospital) and carcinogen exposure studied (yes/no) were pre-specified as characteristics for the assessment of heterogeneity. Meta-analysis was carried out using Review Manager Version 4.2 (Cochrane Collaboration) and STATA software for meta-regression analysis. P-values were two sided.

**RESULTS**

In this case–control study, we have examined the association of SULT1A1 Arg213His (SULT1A1*2) polymorphism in 132 patients with tobacco-related multiple primary cancers and 198 cancer-free controls. For selection of MPN patients, stringent modified Hong’s criteria were used to minimise the possibility of misclassifying...
metastasis, recurrences, skip lesions and radiation-induced cancers, as second primaries. Radiation therapy was used in the management of first primary in 60% of patients but none of the second primaries were classified as radiation-associated sarcomas (Huber et al., 2007) or meningiomas (King et al., 2007).

Using the IARC definition of TRC (IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, 2004), these 132 MPN patients with at least one TRC primary were further subclassified as those with at least one primary in the UADT (UADT-MPN, n = 113, Table 1) and those having none of the primaries in UADT (n = 19). Majority of the patients (n = 74) had both the primaries within the UADT region. The characteristics of these 113 patients with at least one primary in UADT and the healthy controls are shown in Table 1. Of the 113 patients with UADT-MPN, 96 (85%) reported tobacco use and majority (68%) had tobacco-chewing habit (Supplementary Table S1). The most common form of tobacco was chewing of tobacco quid with lime or with betel leaf or application of roasted tobacco (masheeri) over gums. Quanta and duration of tobacco chewing were not available for all the participants and hence were not included in the analysis.

The genotype distribution of SULT1A1*2 as His/His (homozygous variant), Arg/His (heterozygous) and Arg/Arg (homozygous wild type) was compared in cases and controls using dominant, recessive and extreme models. These models were based on the biological plausibility that His213 variant allele is risk conferring compared with Arg213 allele. Thus His/His213 genotype was considered risk conferring whereas Arg/Arg213 would confer protection. However, activity of the SULT1A1 enzyme in platelets from heterozygous (Arg/His) individuals has been reported to be only slightly lower than that from the Arg/Arg individuals but much higher than that from His/His individuals (Rafogianis et al., 1997; Nowell et al., 2000). Hence recessive model (Arg/Arg+Arg/His vs His/His) was considered in the study.

The risk association of SULT1A1*2 was evaluated in 113 MPN patients with at least one UADT TRC. The remaining 19 patients with both TRCs outside UADT were analysed as a separate group as well as a combined TRC group (n = 132) (Table 2). The results of the TRC outside UADT group (n = 19) and the combined group (n = 132) are not included in the meta-analysis due to small sample size (n = 19) with very diverse TRCs (cervix, bladder and stomach and so on). After adjusting for age and tobacco use, a significant risk association of SULT1A1*2 with UADT TRC was seen in dominant, recessive as well as extreme models. In all three models, there was a significant increased risk associated with His213 genotype (Table 2).

To compare these observations with the published studies, a meta-analysis of studies evaluating association of SULT1A1 Arg213His with different cancers was performed. From the Medline search using the search terms described earlier, we identified 34 case–control studies for SULT1A1 Arg213His polymorphism (Seth et al., 2000; Steiner et al., 2000; Bamber et al., 2001; Zheng et al., 2001, 2003; Ozawa et al., 2002; Wang et al., 2002; Nowell et al., 2002a, 2004; Tang et al., 2003; Wu et al., 2003; Chacko et al., 2004; Hung et al., 2004; Langensleben et al., 2004; Liang et al., 2004; Tiemersma et al., 2004; Tsukino et al., 2004; Boccia et al., 2005, 2006; Cheng et al., 2005; Choi et al., 2005; Han et al., 2005; Jerevall et al., 2005; Marchand et al., 2005; Lilla et al., 2005; Moreno et al., 2005; Pereira et al., 2005, 2006; Sun et al., 2005; Yang et al., 2005; Dandara et al., 2006; Kellen et al., 2006; Mikhailova et al., 2006; Pachouri et al., 2006). Including the 113 patients with UADT-MPN from this study, there were 11 962 cancer cases and 14 673 cancer-

### Table 1 Demographics of study subjects

| Category | UADT TRC<sup>a</sup> (n = 113) (%) | Cancer-free controls<sup>b</sup> (n = 198) (%) |
|----------|---------------------------------|---------------------------------|
| Gender   |                                 |                                 |
| Males    | 74 (65)                         | 129 (65)                        |
| Females  | 39 (35)                         | 69 (35)                         |
| Age<sup>c</sup> |                                |                                 |
| Median   | 50                              | 46                              |
| Range    | (26–75)                         | (20–84)                         |
| Type of MPN |                                |                                 |
| Synchronous<sup>d</sup> | 32 (28)                   | —                               |
| Metachronous | 79 (70)                  | —                               |
| Primary cancer sites |                             |                                 |
| Oral – 90 | —                               |                                 |
| Oesophagus – 28 | —                           |                                 |
| Larynx/hypopharynx – 24 | —                         |                                 |
| Tobacco habit |                                |                                 |
| No habit | 14 (12)                         | 14 (7)                          |
| Only T   | 70 (62)                         | 156 (79)                        |
| T+A      | 26 (23)                         | 27 (14)                         |
| No information | 3 (3)                | 1 (<1)                          |
| SULT1A1 genotypes |                             |                                 |
| Arg/Arg  | 60 (53)                         | 135 (68)                        |
| Arg/His  | 47 (42)                         | 61 (31)                         |
| His/His  | 6 (5)                           | 2 (1)                           |

<sup>A</sup> = alcohol; <sup>MPN</sup> = multiple primary neoplasm; <sup>T</sup> = tobacco; <sup>TRC</sup> = tobacco-related cancer; <sup>UADT</sup> = upper aerodigestive tract. Tobacco-related cancers were defined by IARC (2004) and included UADT (including nasopharynx), cervix, bladder, stomach, kidney, liver; pancreas and myeloid leukaemia. <sup>d</sup>At least one primary in the UADT. <sup>1</sup>Controls were enrolled mainly from the Preventive Oncology Department, Tata Memorial Hospital and Government Dental College, Mumbai. <sup>c</sup>Age (years) at the diagnosis of the index cancer of the patients or age at accrual for the controls. <sup>d</sup>Synchronous – cancers occurring within 6 months of diagnosis of first primary site.

### Table 2 Analysis of risk association in tobacco-related MPN patients using genetic models

| Category | Genotype | Cancer-free controls<sup>a</sup> (n = 198) | TRC outside UADT<sup>a</sup> (n = 19) | At least one in UADT<sup>a</sup> (n = 113) | All TRCs (n = 132) |
|----------|----------|---------------------------------|---------------------------------|---------------------------------|-------------------|
| Dominant | Arg/Arg  | 135                             | 6 (ref)                         | 60 (ref)                        | 66 (ref)          |
|          | His/His/His/Arg | 2, 61                   | 0, 13 (7.91 (2.06, 30.39)) | 6, 47 (1.94 (1.20, 3.14)) | 6, 60 (2.12 (1.3, 3.44)) |
|          | Recessive | His/Arg/Arg/Arg | 61, 135 | 13, 6 (ref) | 47, 60 (ref) | 60, 66 (ref) |
|          | Arg/Arg  | 2                                | 0                               | 6 (6.07 (1.20, 30.66)) | 6 (7.43 (1.42, 38.820) |
|          | His/His  | 6 (ref)                         | 60 (ref)                        | 66 (ref)                        | 6 (8.92 (1.71, 46.66)) |
|          | Extreme  | Arg/Arg | 135                         | 6 (ref)                         | 6 (7.84 (1.53, 40.15)) | 6 (8.92 (1.71, 46.66)) |

<sup>MPN</sup> = multiple primary neoplasm; <sup>TRC</sup> = tobacco-related cancer; <sup>UADT</sup> = upper aerodigestive tract. *Odds ratio (OR) adjusted for age and tobacco habit; 95% CI – 95% confidence interval.
free controls. To specifically examine the risk association of His213 with different types of cancers including UADT TRC, which is biologically more plausible, the studies included in meta-analysis were categorised according to the site of primary cancer as UADT TRC, genitourinary, breast, colorectal and other cancer sites. All the studies were further analysed with respect to genotypes, source of controls, ethnicity and carcinogen exposure. The study characteristics (Supplementary Table s2) showed that 13 studies had accrued controls from general population whereas 20 studies had hospital-based controls and one had mixed source of controls. In 23 studies, the distribution of genotypes in controls was consistent with Hardy–Weinberg equilibrium (Supplementary Table s3). It was noteworthy that the His213 allele occurred at a significantly lower frequency amongst Asians (13%; 95% confidence interval (CI): 7 – 19) as compared with Caucasians (33%; 95% CI: 30 – 37) and other ethnic groups (31%; 95% CI: 23 – 31) (Figure 1). However, it is possible that the variation is even larger between different Asian populations.

All the studies were analysed using the three models, namely recessive (Figure 2), which was biologically more plausible (Raftogianis et al., 1997), as well as dominant and extreme models (Supplementary Figure s1, s2 and Supplementary Table s3). All the three models showed a high degree of statistical heterogeneity among the 35 studies, including this study. Meta-regression analysis was performed to investigate the source for statistical heterogeneity (Supplementary Table s3). No obvious source of heterogeneity was identified except for ethnicity in the dominant model.

Symmetrical funnel plot suggested the absence of publication bias for all the three models (Egger’s test P-value > 0.05; Supplementary Figure s3). Influence analysis was carried out to study the effect of individual studies in the meta-analysis on the overall outcome (Supplementary Figure s4). None of the studies affected the outcome of the meta-analysis significantly. When different ethnic groups were analysed separately irrespective of cancer site, the Asians showed a significant increased risk (OR = 1.84, 95% CI: 1.20, 2.83) as compared with Caucasians (OR = 1.03, 95% CI: 0.82, 1.29) (Supplementary Table s3) in the recessive model.

The effect of ethnicity for specific cancer sites could be examined separately only for breast cancer where ethnicity was analysed separately only for breast cancer where ethnicity was recessive model (OR = 2008 Cancer Research UK). The Asians showed a significant increased risk of breast cancer (OR = 1.91, 95% CI: 1.08, 3.40), it did not confer increased risk to Caucasian women (OR = 0.92, 95% CI: 0.71, 1.18). Stratified meta-analysis according to cancer site, irrespective of ethnicity or any other factor, showed a 1.46- to 1.62-fold risk for UADT cancer in all the three models, whereas the cancers in the genitourinary site showed a significant protection with an OR of 0.67–0.81 in the three models (Figure 2, Supplementary Figure s1 and s2). Other tumour sites, however, did not show any significant association.

**DISCUSSION**

Despite decades of public health programmes, TRCs remain the leading cause of cancer morbidity and mortality worldwide. Epidemiological studies over the past 50 years have clearly established how tobacco contributes to cancer risk not only in the directly exposed and anatomically related regions of the upper aerodigestive tract and lung but also in distant organs such as cervix, bladder, kidney, pancreas and so on (IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, 2004). Tobacco is implicated as the single most important environmental factor for several TRCs (lung, head and neck, oesophagus, bladder, kidney and pancreas). It also confers significant risk for cancers even where viral or bacterial oncogenesis plays a predominant role (e.g., cervix, stomach, liver and nasopharynx). Weak genetic susceptibility in tobacco-exposed population is conferred by a large number of low-penetrance genes. However, there is paucity of systematic studies of all the important genes that may predispose to tobacco carcinogenesis.

The focus of research to elucidate genetic susceptibility to tobacco carcinogenesis has been on phase I and phase II detoxifying enzymes and to a lesser extent on the genes that regulate DNA repair, apoptosis and other relevant pathways. In contrast to the GST super family of enzymes that have been studied extensively for tobacco carcinogenesis (Nakajima et al., 1995; Cheng et al., 1999; Buch et al., 2002; Jhavar et al., 2004, 2005), other phase II metabolising enzymes such as the SULT have been less extensively studied (Hung et al., 2004; Dandara et al., 2006; Pachouri et al., 2006). There also has not been any collation of published data or a meta-analysis of case-control studies evaluating SULT enzyme in cancers.

Yasuda et al. (2007) have reported that of the 11 known human cytosolic sulphotransferases, SULT1A1 is one of the four major SULT enzymes responsible for sulphation of tobacco carcinogens. The role of SULT1A1 in the biotransformation of tobacco carcinogens and its association with lung cancer has been previously reported (Liang et al., 2004). There are reports of SULT1A1*2 association with increased risk for oesophageal cancers (Dandara et al., 2006) as well as gastric cancer (Boccia et al., 2005) in individuals who consume alcohol and smoke tobacco.

To elucidate the effect of Arg213His polymorphism of SULT1A1 in tobacco users, we have studied a group of patients with multiple primary cancers, where at least one of the primary cancers was a TRC. We have postulated that as opposed to patients with a single primary TRC, those who develop multiple primary cancers are likely to show more pronounced gene–environment interactions (Kotnis et al., 2005). Hence, this may be a better clinical model to detect significant association of low-penetrance genes, even in smaller number of patients. In this case–control study, we show a strong association of the Arg213His polymorphism of SULT1A1 with the development of tobacco-related UADT cancers. These findings are further supported by the results of the meta-analysis examining the association of this polymorphism with cancer risk.
**SULT1A1 Arg<sup>213</sup>His polymorphism in tobacco-related cancers**

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**Review:** SULT1A1 Meta Analysis

**Comparison:** Cancer risk and SULT1A1Arg213 polymorphism - Recessive model (His/His vs His/Arg+Arg/Arg)

**Outcome:** Cancer risk

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| Study or subcategory | Cases | Controls | OR (random) | Weight | OR (random) | Year |
|----------------------|-------|----------|-------------|--------|-------------|------|
| **Genitourinary**    |       |          |             |        |             |      |
| Boccia (H&N)         | 8/197 | 9/247    | 2.08        | 1.12   | 0.42, 2.96  | 2006 |
| Liang (lung)         | 7/805 | 3/809    | 1.30        | 2.36   | 0.61, 9.15  | 2004 |
| Pachouri (lung)      | 12/103| 9/122    | 2.27        | 1.66   | 0.67, 4.10  | 2006 |
| Wang Y (lung)        | 67/643| 63/465   | 4.58        | 1.13   | 0.78, 1.64  | 2002 |
| Dandara C (oesoph)   | 74/236| 48/236   | 4.35        | 2.37   | 1.37, 4.15  | 2006 |
| Wu MT (oesoph)       | 0/187 | 0/308    | Not estimable |       |             |      |
| Present study        | 6/113 | 2/198    | 0.98        | 5.50   | 1.09, 27.70 | 2007 |
| **UADT & Lung**      | 2104  | 2435     | 15.55       | 1.62   | 1.12, 2.34  |      |
| Total events: 174 (cases), 134 (controls) | | | | | |
| **Test for heterogeneity:** $\chi^2 = 14.04$, df = 9, $P = 0.12$, $I^2 = 35.9\%$ | | | | | |
| **Test for overall effect:** $Z = 2.56$ ($P = 0.01$) | | | | | |

**BREAST**

| Study | Cases | Controls | OR (random) | Weight | OR (random) | Year |
|-------|-------|----------|-------------|--------|-------------|------|
| Charao  | 8/140 | 4/140 | 1.52 | 2.06 | 0.61, 7.01 | 2004 |
| Cheng TC | 2/468 | 0/740 | 0.32 | 7.94 | 0.38, 165.68 | 2005 |
| Choi J | 0/986 | 0/1045 | Not estimable |       |             |      |
| Han | 8/209 | 10/426 | 2.16 | 1.66 | 0.64, 4.26 | 2004 |
| Jerevall P | 28/229 | 38/228 | 3.79 | 0.66 | 0.41, 1.18 | 2005 |
| Langsenlehner U | 47/498 | 63/499 | 4.43 | 0.72 | 0.48, 1.08 | 2004 |
| Le Marchand L | 114/1339 | 104/1370 | 5.03 | 1.13 | 0.86, 1.49 | 2005 |
| Lilla C | 52/419 | 107/884 | 4.66 | 1.03 | 0.72, 1.47 | 2005 |
| Seth P | 39/444 | 23/222 | 3.71 | 0.85 | 0.50, 1.47 | 2000 |
| Tang D | 11/130 | 7/133 | 2.05 | 2.15 | 0.80, 5.76 | 2003 |
| Yang | 0/1102 | 0/1147 | Not estimable |       |             |      |
| Zheng | 29/155 | 44/326 | 3.85 | 1.48 | 0.88, 2.47 | 2001 |
| **Subtotal (95% CI)** | 8092 | 7165 | 31.51 | 1.06 | 0.85, 1.32 |      |
| **Total events:** 338 (cases), 400 (controls) | | | | | |
| **Test for heterogeneity:** $\chi^2 = 14.04$, df = 9, $P = 0.12$, $I^2 = 35.9\%$ | | | | | |
| **Test for overall effect:** $Z = 0.50$ ($P = 0.62$) | | | | | |

**Colorectum**

| Study | Cases | Controls | OR (random) | Weight | OR (random) | Year |
|-------|-------|----------|-------------|--------|-------------|------|
| Bamber DE | 26/226 | 32/293 | 3.68 | 1.06 | 0.61, 1.84 | 2001 |
| Moreno V | 23/229 | 23/272 | 3.43 | 0.92 | 0.50, 1.69 | 2005 |
| Nowell S | 15/130 | 55/301 | 3.38 | 0.58 | 0.32, 1.08 | 2002 |
| Sun XF | 39/109 | 97/666 | 4.19 | 3.27 | 2.09, 5.11 | 2005 |
| Teinserma EW | 40/348 | 58/373 | 4.26 | 0.71 | 0.46, 1.09 | 2004 |
| **Subtotal (95% CI)** | 1106 | 1905 | 18.94 | 1.07 | 0.56, 2.05 |      |
| **Total events:** 143 (cases), 265 (controls) | | | | | |
| **Test for heterogeneity:** $\chi^2 = 31.40$, df = 4, $P < 0.00001$, $I^2 = 87.3\%$ | | | | | |
| **Test for overall effect:** $Z = 0.20$ ($P = 0.84$) | | | | | |

**Others**

| Study | Cases | Controls | OR (random) | Weight | OR (random) | Year |
|-------|-------|----------|-------------|--------|-------------|------|
| Boccia (stomach) | 9/76 | 13/260 | 2.32 | 2.55 | 1.05, 6.23 | 2005 |
| Mihailova (Endo+Ov) | 28/166 | 50/170 | 3.81 | 0.49 | 0.29, 0.82 | 2005 |
| Pereira W (mixed) | 17/123 | 11/100 | 2.59 | 1.27 | 0.57, 2.86 | 2005 |
| Sellers T (ovary) | 63/454 | 69/542 | 4.60 | 1.10 | 0.77, 1.59 | 2005 |
| **Subtotal (95% CI)** | 821 | 1072 | 13.31 | 1.07 | 0.58, 1.97 |      |
| **Total events:** 117 (cases), 143 (controls) | | | | | |
| **Test for heterogeneity:** $\chi^2 = 12.11$, df = 3, $P = 0.007$, $I^2 = 75.2\%$ | | | | | |
| **Test for overall effect:** $Z = 0.23$ ($P = 0.82$) | | | | | |

| Total (95% CI) | 11962 | 14673 | 100.00 | 1.08 | 0.91, 1.29 |      |
| Total events: 911 (cases), 1152 (controls) | | | | | |
| **Test for heterogeneity:** $\chi^2 = 86.47$, df = 31, $P < 0.00001$, $I^2 = 64.1\%$ | | | | | |
| **Test for overall effect:** $Z = 0.90$ ($P = 0.37$) | | | | | |

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**Figure 2** Meta-analysis recessive model.
The meta-analysis also brings out the markedly lower mean frequency of *SULT1A1* His213 in the Asian population as compared with the Caucasian population. However, it is possible that the variation is even larger if different Asian populations are taken in the study separately. We (Jhavar et al., 2004, 2005) and others from India (Buch et al., 2002; Anantharaman et al., 2007) have reported a markedly lower frequency of GSTT1 null genotype in the Indian population as compared with that in the Japanese, Chinese and Korean population (Raimondi et al., 2006). Although marked geoethic variation in the incidence of different cancers is attributed largely to the differences in carcinogenic exposure and diet, marked differences in the population frequency of the risk-confering genotype of some XMEs could also influence cancer risk. The results of this meta-analysis are intriguing as they demonstrate opposite effects of *SULT1A1* polymorphism on two distinct anatomical sites of TRCs. Thus in the meta-analysis, in contrast to the seven studies where UADT and lung cancers showed an increased risk association with *SULT1A1*2, seven studies on genitourinary cancers showed a protective effect. This could perhaps be explained by the dual role of *SULT1A1* in the bioactivation as well as detoxification of carcinogens (Glatt, 2000). Thus, detoxification of exogenous and endogenous carcinogens confers a protective effect for cancer (Glatt et al., 2001), whereas bioactivation of promutagens could increase the risk of certain cancers (Zheng et al., 2003; Tiemersma et al., 2004). The risk association of *SULT1A1*2 with cancers of the UADT and lung is expected from its known role in tobacco detoxification (Al-Bueheissi et al., 2006). Nowell et al. (2004) has reported that *SULT1A1* could contribute to prostate cancer risk, and the magnitude of the association may depend on ethnicity and meat consumption. It has been reported that the carcinogens are transferred to the kidney and ureter (Meinl et al., 2006) although their levels are substantially lower in the kidney than in the liver. However, it is difficult to explain the protective role of *SULT1A1*2 His213 variants with cancer in the genitourinary cancers. Detailed biochemical studies in different human tissues, especially in the genitourinary vs UADT region, might explain the opposing tissue-specific effects of *SULT1A1*.

Another important aspect that has emerged from the meta-analysis is the difference in the risk of breast cancer conferred by *SULT1A1*2 variant to Asian women compared with Caucasian women. A similar phenomenon has been reported for GSTM1 polymorphism. Carlsten et al. (2008) have reported that although GSTM1 null status conferred a significantly increased risk of lung cancer to East Asians it did not confer increased risk to Caucasians. Thus, in distinct ethnic groups, risk for different cancers could be modulated by interaction between genetic variants and different endogenous and exogenous carcinogens.

There are several limitations in the present meta-analysis as is often the case. Contribution of possible sources of heterogeneity such as site of cancer, ethnicity, Hardy–Weinberg equilibrium, source of controls, sample size/power and carcinogen exposure were considered. However, meta-regression analysis demonstrated a significant heterogeneity due to ethnicity alone. This was also reflected in the allele frequency where Asians and Caucasians showed a striking difference. Hence, the actual source of heterogeneity could not be investigated due to the complexities of the confounding variables. In addition, meta-analysis in general looks at the crude OR instead of adjusted OR as the adjustment and matching factors differ across the studies. The residual confounders might have influenced our analysis.

This study encourages detailed biochemical investigation on the tissue-specific influence of *SULT1A1* Arg213His enzyme in metabolism of tobacco carcinogens. This is the first meta-analysis that provides significant and contrasting association of *SULT1A1* Arg213His polymorphism on cancer risk in distinct sites of TRCs namely UADT and genitourinary and an increased risk for breast cancer in Asians.

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