

• ESOPHAGEAL CANCER •

Phase I/II enzyme gene polymorphisms and esophageal cancer risk: A meta-analysis of the literature

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Received: 2004-09-24 Accepted: 2004-11-19

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Key words: CYPs; GSTs; Gene polymorphisms; Esophageal cancer; Meta-analysis

Yang CX, Matsuo K, Wang ZM, Tajima K. Phase I/II enzyme gene polymorphisms and esophageal cancer risk: A meta-analysis of the literature. *World J Gastroenterol* 2005; 11(17): 2531-2538

<http://www.wjgnet.com/1007-9327/11/2531.asp>

Abstract

AIM: Phase I/II enzymes metabolize environmental carcinogens and several functional polymorphisms have been reported in their encoding genes. Although their significance with regard to esophageal carcinogenicity has been examined epidemiologically, it remains controversial. The present systematic review of the literature was performed to clarify associations.

METHODS: Eligible studies were case-control or cohort studies published until September 2004 that were written in any language. From PubMed and a manual review of reference lists in relevant review articles, we obtained 16 studies related to the *CYP1A1* Ile-Val substitution in exon 7, *CYP1A1* *MspI* polymorphisms, *CYP2E1* *RsaI* polymorphisms, *GSTM1* null type, *GSTT1* null type and *GSTP1* Ile104Val. All were of case-control design. Summary statistics were odds ratios (ORs) comparing heterozygous-, homozygous-non-wild type or these two in combination with the homozygous wild type, or the null type with the non-null type for *GSTM1* and *GSTT1*. A random effect model was used to estimate the summary ORs. A meta-regression analysis was applied to explore sources of heterogeneity.

RESULTS: Individuals with the Ile-Val substitution in *CYP1A1* exon 7 had increased esophageal cancer risk, with ORs (95%CI) compared with *Ile/Ile* of 1.37 (1.09-1.71), 2.52 (1.62-3.91) and 1.44 (1.17-1.78) for *Ile-Val*, *Val/Val* genotype and the combined group. No significant association was found between esophageal cancer risk and the other genetic parameters.

CONCLUSION: A significant association exists between the *CYP1A1* Ile-Val polymorphism and risk of esophageal cancer. Polymorphisms that increase the internal exposure to activated carcinogens may increase the risk of esophageal cancer.

INTRODUCTION

Most environmental chemical carcinogens undergo activation by phase I enzymes, often in an oxidation reaction, and detoxication by phase II enzymes. The cytochrome P450 enzyme superfamily constitutes the majority of phase I enzymes, while the glutathione-S-transferases (GSTs) and N-acetyltransferase are primarily responsible for the detoxication of xenobiotics. The drug-metabolizing enzymes often display genetic polymorphisms, which may alter the enzyme activity and thus impact on the risk of cancer.

The enzyme CYP1A1 is involved in the activation of major classes of tobacco procarcinogens, like polycyclic aromatic hydrocarbons and aromatic amines, and is present in many epithelial tissues^[1]. *CYP1A1* Ile-Val substitution in the heme-binding region results in a two-fold increase in microsomal enzyme activity and is in complete linkage disequilibrium in Caucasians with the *CYP1A1* *MspI* polymorphism, which has also been associated experimentally with increased catalytic activity^[2]. *CYP2E1* is primarily responsible for the metabolic activation of many low molecular weight carcinogens^[3], including certain nitrosamines, which may be involved in carcinogenesis of the esophagus. This enzyme is also believed to participate in the oxidation of other compounds, such as ethanol, to produce reactive free radicals that may initiate lipid peroxidation and consequently influence carcinogenesis^[4]. The variant $\alpha 2$ allele recognized by *RsaI* digestion in the 5'-flanking region of the gene appears to be associated with decreased enzyme activity^[5].

GSTs are a family of multifunctional enzymes which metabolize a variety of xenobiotics with a large overlap in the substrate specificity. Individuals who are homozygous for the null *GSTM1* or null *GSTT1* alleles lack the respective enzyme functions^[6,7]. *GSTP1* is a major GST isoform expressed in human esophagus^[8], which can eliminate DNA oxidative products of thymidine or uracil propenal^[9]. After induction by cytochrome P450, some cigarette-related carcinogens, such as benzo[a]pyrene diol epoxide and acrolein,

can also be eliminated by GSTP1^[10]. The Ile-Val substitution at residue 104 may be associated with a higher level of DNA adducts^[11], thus increasing the susceptibility to cancer induction.

Therefore, the CYP1A1 *Val* allele, the CYP1A1 *MspI* non-wild allele, the null type of GSTM1 and GSTT1 as well as the GSTP1 *Val* allele may increase the risk of esophageal cancer, while the CYP2E1 c2 allele (recognized by *RsaI* digestion) may decrease the risk. Based on the possible biological significance of *CYP1A1*, *CYP2E1*, *GSTM1*, *GSTT1* and *GSTP1* polymorphisms on cancer susceptibility, several epidemiologic studies have been conducted to assess their association with esophageal cancer. However, most studies featured only small samples and the results were not always consistent. To obtain a better understanding of the significance of gene polymorphisms with regard to esophageal cancer risk, we performed a systematic review of all the relevant studies published in the literature.

MATERIALS AND METHODS

Selection of studies

Before the study, we defined inclusion criteria as follows: (1) any study design giving relative risk (an OR or a risk ratio) for candidate gene (*CYP1A1*, *CYP2E1*, *GSTM1*, *GSTT1*, and *GSTP1*) polymorphisms regarding the risk of esophageal cancer (including both squamous cell carcinomas and adenocarcinomas); (2) inclusion of non-cancer or disease-free subjects as a control group; (3) already published in any language but cited in PubMed.

All the studies were obtained via PubMed using key words “CYP1A1”, “CYP2E1”, “GSTM1”, “GSTT1” and “GSTP1” in combination with “esophageal cancer” to identify potentially relevant articles. A total of 45 articles were captured, and 21, 28, 20, 12, and 12 were related to CYP1A1, CYP2E1, GSTM1, GSTT1, and GSTP1, respectively. We selected all the studies, which provided a relative risk with the candidate gene.

We examined abstracts of all the candidate articles to decide whether to include/exclude in the further detailed review. Thereby, we excluded a total of 22 studies due to inappropriate study design; among them, 8, 18, 5, 4, and 4 were related to *CYP1A1*, *CYP2E1*, *GSTM1*, *GSTT1*, and *GSTP1*, respectively. Among the 22 excluded articles, five were reviews^[1, 12-15], three concerned the expression of cytochrome P450 (CYPs) in esophageal mucosa^[16-18], four covered animal experiments^[19-22], three compared gene polymorphism frequencies among different populations^[23-25], one focused on the metabolism of *N*-nitrosobenzylmethylamine by human cytochrome P450 enzyme^[26], one was related to gastric cancer, not esophageal cancer^[27], and other five were incompatible with the inclusion criteria^[28-32].

Other studies were further excluded based upon detailed review because in three cases^[33-35] they were the same study as in two other papers^[36]. The newest two studies were retained for the analysis. Three more studies were excluded because they did not provide relevant information required for our analysis. Most of them did not apply subjects with other diseases as control groups and did not provide relative risk of esophageal cancer for candidate gene polymorphisms^[37-39]. One study was excluded since it was the only example which examined associations between a tandem

repeat polymorphism of *CYP2E1* and the risk of cancer^[40].

Finally, a total of 16 case-control studies were included in the meta-analysis (Table 1), 9, 5, 12, 6 and 7 concerning *CYP1A1*, *CYP2E1*, *GSTM1*, *GSTT1* and *GSTP1*, respectively. All the potentially relevant articles were reviewed by two independent investigators (Y.CX. and M.K.).

We also tried to use “esophageal” combined with candidate genes as keywords to search for much more relevant articles as well as check the reference lists in the reviews and selected original investigations and found no additional eligible articles.

Data abstraction

Two investigators using a standard information extraction form independently abstracted data. Characteristics abstracted from the articles included the name of the first author, year of publication, location of the study, study design, mean age for all cases and controls, the percentage of males in the case and control groups, matched factors as well as adjusted factors; number of cases, number of controls, number of cases and controls with each genotype of candidate polymorphisms, and overall crude or adjusted odds ratios (ORs) with their 95%CI. For one study^[41], which supplied the result for both present controls and total controls (including historic control and the present control), total control data were selected for our meta-analysis.

Statistical analysis

The STATA statistical package (version 8, stata, College Station, TX) was used for the meta-analysis. The homozygous wild type was used as the reference group for *CYP1A1*, *CYP2E1* and *GSTP1*, and the non-null type for *GSTM1* and *GSTT1*. With four papers^[36, 44, 47, 48] whose reference groups were defined in the opposite way, the ORs were inverted for our analysis. Adjusted ORs were employed for the present meta-analysis if available in the reports, otherwise, crude ORs were used. Since some of the original studies did not provide the ORs but the genotype frequencies were available, crude ORs were then calculated and employed for our meta-analysis. A random-effect model was applied to obtain summary ORs and their 95%CIs since the results with fixed-effect models are the same as with random-effect models if there is no heterogeneity across the studies. A random-effect model should be applied if heterogeneity exists. Publication bias was graphically assessed by funnel plots and statistically assessed by Egger's test. Meta-regression analysis was applied to explore potential sources of heterogeneity. The factors, study design, Chinese population (yes/no), Asian population (yes/no), publication year (after 2 000 or not), number of cases and controls (both greater than 100 or not) and matching (matched for sex and age or not) were examined. Statistical significance was defined as a *P*-value less than 0.05 except for meta-regression analyses, which used a *P*-value 0.10 because of the relatively weak statistical power.

RESULTS

In the final analysis, we had a total of 16 case-control studies consisting of 3 hospital-based (controls selected from non-cancer patients), 12 population-based (controls selected from the healthy population) and 1 without a clear type.

Among them, 9 were studies of the *CYP1A1* exon 7 Ile-Val substitution, 1^[53] without any Ile-Val substitution in either cases and controls, 5 concerned the *CYP1A1* *MspI* polymorphism, 5 the *CYP2E1* *RsaI* polymorphism, 12 the *GSTM1* null type, 6 the *GSTT1* null type and 7 the *GSTP1*

Ile-Val substitution (Table 1).

For *CYP1A1* exon 7 Ile-Val substitution, all ORs for the *Ile/Val* genotype and the combined group were larger than 1 when compared with the *Ile/Ile* genotype, although only one study demonstrated a significantly increased risk.

Table 1 Summary of studies included in the analysis of CYP1A1, CYP2E1, GASTM1, GSTT1 and GSTP1

Study	Yr	Study design	Cases Mean age	Controls Mean age	Cases Man (%)	Controls Man (%)	Polymorphisms	Result	Adjusted factors	Reference
Hori H	1997	2	NA	NA	78 (83)	NA	<i>CYP1A1</i> , <i>MspI</i>	NS		42
							<i>CYP1A1</i> , Ile-Val	NS		
							<i>GSTM1</i>	NS		
Nimura Y	1997	1	NA	NA	67 (75)	76 (55)	<i>CYP2E1</i> , <i>RsaI</i>	NS		43
							<i>CYP1A1</i> , Ile-Val	S		
							<i>GSTM1</i>	NS		
Morita S	1997	2	62.2	50.5	45 (85)	112 (85)	<i>CYP1A1</i> , Ile-Val	NS		44
							<i>CYP2E1</i> , <i>RsaI</i>	NS		
							<i>GSTM1</i>	NS		
Morita S	1998	2	62.1	49.8	56 (85)	102 (62)	<i>GSTP1</i>	S		45
Lin DX	1998	NA	55.5	53.3	27 (60)	23 (50)	<i>CYP2E1</i> , <i>RsaI</i>	S	Adjusted by age and sex	36
							<i>CYP2E1</i> , <i>DraI</i>	NS		
							<i>GSTM1</i>	NS		
							<i>GSTT1</i>	NS		
							<i>GSTP1</i>	NS		
van Lieshout EM	1999	2	NA	52	27 (79)	98 (40)	<i>CYP1A1</i> , <i>MspI</i>	S		46
							<i>CYP1A1</i> , Ile-Val	NS		
							<i>GSTM1</i>	NS		
							<i>GSTT1</i>	NS		
							<i>GSTP1</i>	S		
Shao G	2000	2	55	53	74 (69)	80 (72)	<i>CYP1A1</i> , Ile-Val	NS		47
							<i>GSTM1</i>	S		
Lee JM	2000	2	NA	NA	82 (91)	228 (89)	<i>GSTP1</i>	NS	Adjusted by potential factors	48
Tan W	2000	2	54.5	53.6	99 (66)	99 (66)	<i>CYP2E1</i> , <i>RsaI</i>	S	Adjusted by age, sex and smoking	49
							<i>GSTM1</i>	S		
							<i>GSTT1</i>	NS		
							<i>GSTP1</i>	NS		
Wu M-T	2002	1	60.6	61.2	133 (91)	298 (92)	<i>CYP1A1</i> , <i>MspI</i>	NS	Adjusted by age, sex, smoking and alcohol, etc.	50
Gao CM	2002	2	NA	NA	55 (59)	131 (66)	<i>CYP1A1</i> , Ile-Val	S		
							<i>CYP2E1</i> , <i>RsaI</i>	NS	Adjusted by age, sex and potential factors	51
Yokoyama A	2002	2	61.7	58.8	234 (100)	634 (100)	<i>GSTM1</i>	NS		52
Gao CM	2002	2	NA	NA	78 (55)	149 (67)	<i>GSTM1</i>	S	Adjusted by age and sex	53
							<i>GSTT1</i>	NS		
Wang LD	2003	2	NA	NA	32 (53)	NA	<i>CYP1A1</i> , <i>MspI</i>	NS		54
							<i>CYP1A1</i> , Ile-Val	NS		
							<i>GSTM1</i>	NS		
							<i>GSTT1</i>	NS		
							<i>GSTP1</i>	NS		
Casson AG	2003	2	NA	NA	38 (84)	38 (84)	<i>CYP1A1</i> , <i>MspI</i>	NS	Adjusted by age, sex and smoking	55
							<i>CYP1A1</i> , Ile-Val	NS		
							<i>GSTM1</i>	NS		
							<i>GSTT1</i>	NS		
							<i>GSTP1</i>	S		
Wang AH	2004	1	NA	NA	97 (76)	78 (77)	<i>CYP1A1</i> , Ile-Val	S		37
							<i>GSTM1</i>	S		

1: Population-based case-control study; 2: hospital-based case-control study. NA, not available; NS, not significant ($P>0.05$); S, significant ($P<0.05$).

In three of eight cases, the *Val/Val* genotype was associated with significantly increased ORs (Table 2). The meta-analysis with a total of 754 cases and 1 563 controls showed significantly increased ORs of 1.37 (1.09-1.71), 2.52 (1.62-3.91) and 1.44 (1.17-1.78) for *Ile-Val* and *Val/Val* genotypes and the combined group, respectively. There was no heterogeneity across the studies, so that the results for the fixed-effect model were the same as for the random-effect model for *CYP1A1* exon 7 *Ile-Val* substitution. In contrast, no significantly increased risk of esophageal cancer was observed for the *CYP1A1 MspI* polymorphism.

For *CYP2E1*, two out of five investigations suggested that the *c2* allele may significantly decrease the risk with adjusted ORs (95%CI) of 0.31 (0.24-0.40) and 0.21 (0.08-0.56) for the homozygous and combined group, respectively. The meta-analysis showed non-significantly decreased ORs for the *c1/c2* and combined group (Table 2). For *GSTP1*, one of seven showed significantly increased risk with ORs (95%CI) of 3.44 (1.47-8.55), 3.65 (0.56-16.82) and 3.47 (1.51-8.46) for the hetero, homo and combined group,

respectively, while one indicated an opposite association. Another study showed a marginally increased OR for the hetero of 2.5 (1.0-6.3) but the meta-analysis generated a null result (Table 2). For *GSTM1*, 3 of 12 studies showed the null type to significantly increase the risk but the meta-analysis failed to confirm this result (Table 3). For *GSTT1*, all the studies were homogenous and both the fixed-effect and random-effect models generated the same result. All the studies and the meta-analysis found no relationship between this gene polymorphism and risk of esophageal cancer (Table 3).

We also examined publication bias for each polymorphism and only the *GSTM1* polymorphism showed a significant existence. Regarding *CYP1A1 Ile-Val*, the test was far from statistically significant. In addition, the source of heterogeneity was examined by meta-regression analysis for potential factors such as Asian and Chinese population, publication year, study design, and matching. No obvious source of heterogeneity was identified except studies in Asian populations for the *GSTP1* polymorphism (Table 4).

Table 2 Summary of the meta-analysis of CYP1A1, CYP2E1, GSTP1 and esophageal cancer risk

Study	Country	Cases	Control	Cases wt/wt	Cases hetero ¹	Cases homo ¹	Control wt/wt ¹	Control hetero ¹	Control homo ¹	OR1 (95%CI) ^{2,3}	OR2 (95%CI) ^{2,3}	OR3 (95%CI) ^{2,3}
<i>CYP1A1 Ile-Val</i> :												
Hori H	Japan	91	428	52	37	2	275	133	20	1.47 (0.89-2.41)	0.53 (0.06-2.29)	1.35 (0.83-2.19)
Nimura Y	China	89	137	50	26	13	92	38	7	1.26 (0.65-2.41)	3.42 (1.17-10.72)	1.59 (0.89-2.87)
Morita S	Japan	53	132	32	20	1	80	49	3	1.02 (0.49-2.08)	0.83 (0.02-10.84)	1.0 (0.5-1.9)
van Lieshout EM	Netherlands	34	247	26	8	0	207	37	3	1.72 (0.62-4.30)	0 (0-10.62)	1.59 (0.58-3.95)
Shao G	China	107	111	43	56	8	55	51	5	1.40 (0.78-2.53)	2.05 (0.54-8.49)	1.46 (0.83-2.59)
Wu M-T	Taiwan	146	324	68	62	16	179	127	18	1.34 (0.86-2.07) ⁴	2.48 (1.15-5.34) ⁴	1.42 (0.94-2.14)
Wang LD	China	62	38	30	28	4	20	16	2	1.17 (0.47-2.93)	1.33 (0.17-15.97)	1.19 (0.49-2.88)
Casson AG	Canada	45	45	45	0	0	45	0	0	NA ⁶	NA ⁶	NA ⁶
Wang AH	China	127	101	21	56	50	31	48	22	1.72 (0.83-3.58)	3.35 (1.49-7.61)	2.24 (1.14-4.43)
Meta-analysis results		754	1 563	367	293	94	984	499	80	1.37 (1.09-1.71)	2.52 (1.62-3.91)	1.44 (1.17-1.78)
<i>CYP1A1 MspI</i> :												
Hori H	Japan	94	242	33	50	11	106	97	39	1.66 (0.96-2.88)	0.91 (0.38-2.06)	1.44 (0.86-2.44)
van Lieshout EM	Netherlands	34	247	22	12	0	207	37	3	3.05 (1.26-7.08)	0 (0-12.63)	2.82 (1.17-6.51)
Wu M-T	Taiwan	146	324	60	65	21	136	146	42	0.98 (0.63-1.53) ⁴	1.24 (0.65-2.36) ⁴	1.04 (0.68-1.57)
Casson AG	Canada	45	45	38	NA	NA	33	NA	NA	NA	NA	0.6 (0.2-1.8) ⁴
Wang LD	China	62	38	33	25	4	12	22	4	0.41 (0.16-1.08)	0.36 (0.06-2.33)	0.41 (0.16-1.02)
Meta-analysis results		381	896	186	152	36	494	302	88	1.21 (0.64-2.32)	1.02 (0.62-1.68)	1.07 (0.64-1.80)
<i>CYP2E1</i> :												
Hori H	Japan	79	633	49	24	6	412	202	19	1.00 (0.57-1.72)	2.66 (0.83-7.33)	1.14 (0.68-1.89)
Morita S	Japan	53	132	34	18	1	85	42	5	1.07 (0.51-2.22)	0.50 (0.01-4.72)	1.0 (0.5-2.0)
Lin DX	China	45	45	36	6	3	20	22	3	0.15 (0.04-0.48)	0.56 (0.07-4.59)	0.21 (0.08-0.56) ^{4,5}
Tan W	China	150	150	107	31	12	66	77	7	0.25 (0.14-0.43)	1.06 (0.36-3.34)	0.31 (0.24-0.40) ^{4,5}
Gao CM	China	93	196	55	31	7	121	62	13	1.13 (0.60-2.13) ⁴	1.23 (0.40-3.77) ⁴	1.15 (0.64-2.07) ⁴
Meta-analysis results		420	1 156	281	110	29	704	405	47	0.59 (0.28-1.23)	1.33 (0.72-2.44)	0.63 (0.30-1.30)
<i>GSTP1</i> :												
Morita S	Japan	66	164	61	5	0	113	48	3	0.19 (0.07-0.52)	0 (0-4.6)	0.13 (0.04-0.45) ⁵
Lin DX	China	42	36	29	12	1	22	11	3	0.83 (0.28-2.51)	0.25 (0.005-3.48)	0.7 (0.3-1.8)
van Lieshout EM	Netherlands	34	247	10	21	3	146	89	12	3.44 (1.47-8.55)	3.65 (0.56-16.82)	3.47 (1.51-8.46)
Lee JM	Taiwan	90	254	65	NA	NA	160	NA	NA	NA ⁶	NA ⁶	0.65 (0.39-1.11) ^{4,5}
Tan W	China	150	150	93	48	9	91	53	6	0.89 (0.53-1.48)	0.95 (0.58-1.55)	1.0 (0.8-1.3)
Wang LD	China	62	38	29	30	3	24	13	1	1.91 (0.76-4.89)	2.48 (0.18-135.66)	1.95 (0.79-4.87) ⁴
Casson AG	Canada	45	45	19	22	4	26	12	7	2.5 (1.0-6.3)	0.8 (0.2-3.1)	1.8 (0.8-4.3) ⁴
Meta-analysis results		489	934	306	138	20	582	226	32	1.17 (0.55-2.49)	1.02 (0.65-1.58)	1.01 (0.60-1.70)

¹"wt" allele was defined for each polymorphism as follows: the *Ile* allele for *CYP1A1 Ile-Val* and *GSTP1 Ile-Val*, the *1 allele for *CYP1A1 MspI*, and *c1* for *CYP2E1*. Non-"wt" allele was defined as follows: the *Val* allele for *CYP1A1 Ile-Val* and *GSTP1 Ile-Val*, the *2 allele for *CYP1A1 MspI* and *c2* for *CYP2E1*. "wt/wt", "hetero", and "homo" indicate homozygous for "wt" allele, heterozygous, and homozygous for non-"wt" allele, respectively. ²wt/wt was defined as the reference group and OR1, OR2, and OR3 for heterozygous, non-"wt" homozygous and the combination of heterozygous and non-"wt" homozygous groups. ³All the ORs were crude values calculated from the genotype distribution except in places denoted by ^{4,5}. ⁴indicates cases where adjusted OR in the report was used and ⁵indicates where the OR value was inverted. ⁶NA: not available.

Table 3 Summary of the meta-analysis of GSTM1, GSTT1 and esophageal cancer risk

Study	Country	Cases	Controls	Case	Case	Control	Control	OR (95%CI)
GSTM1:				Non-null	Null	Non-null	Null	
Hori H	Japan	94	428	53	41	232	196	0.92 (0.57-1.47)
Nimura Y	China	89	137	42	47	74	63	1.31 (0.74-2.32)
Morita S	Japan	53	132	30	23	77	55	1.1 (0.6-2.0)
Lin DX	China	45	45	25	20	24	21	1.0 (0.4-2.3) ³
van Lieshout EM	Netherlands	34	247	17	17	119	128	0.93 (0.42-2.04)
Shao G	China	107	111	68	39	56	55	1.76 (1.03-2.74)
Tan W	China	150	150	104	46	74	76	0.43 (0.33-0.56) ^{3,4}
Yokoyama A	Japan	234	634	131	103	313	321	0.77 (0.56-1.05)
Gao CM	China	141	223	35	106	90	133	2.17 (1.35-3.50) ³
Wang LD	China	62	38	35	27	19	19	0.77 (0.32-1.88)
Casson AG	Canada	45	45	19	26	20	25	1.1 (0.5-2.7) ³
Wang AH	China	127	101	53	74	57	44	1.81 (1.03-3.18)
Meta-analysis results		1 181	2 291	612	569	1 155	1 136	1.07 (0.76-1.51)
GSTT1:								
Lin DX	China	45	45	26	19	22	23	0.7 (0.3-1.5) ³
van Lieshout EM	Netherlands	34	247	28	6	198	49	0.87 (0.28-2.29)
Tan W	China	150	150	90	60	91	59	1.11 (0.83-1.43) ^{3,4}
Gao CM	China	141	223	67	74	104	119	0.90 (0.59-1.39) ³
Wang LD	China	62	38	28	34	18	20	1.09 (0.45-2.65)
Casson AG	Canada	45	45	37	8	33	12	0.6 (0.2-1.7) ³
Meta-analysis results		477	748	276	201	466	282	0.99 (0.80-1.22)

¹Non-null genotype as the reference group. ²All the ORs were crude values calculated from the genotype distribution except in places denoted by ^{3,4}. ³indicates cases where the adjusted OR in the report was used and ⁴where the OR value was inverted.

DISCUSSION

In this systematic review, we found a significant association between the *CYP1A1* Ile-Val polymorphism and the risk of esophageal cancer, while failing to detect links with other gene polymorphisms examined.

The CYPs superfamily, which plays a central part in the metabolism of carcinogens through activating oxidation reactions, may be expressed in esophageal mucosa^[16-18]. The *CYP1A1* Ile-Val substitution in exon 7 results in a two-fold increase in microsomal enzyme activity^[2] and therefore the *Val* allele would be expected to increase the susceptibility to esophageal cancer. In fact, our results are in line with eight of the studies previously published, although five of them failed to find a significant association, possibly because of small sample sizes (Table 2). One meta-(OR for *Val/Val* genotype, 1.62 (0.93-2.82)) and one pooled analysis (OR for *Val/Val* genotype, 1.54 (0.97-1.46)) of lung cancers and another of head and neck cancer (OR for *Val/Val* type, 1.35 (0.95-1.82)) also showed that the *CYP1A1 Val* allele may increase cancer risk, although this was not significant^[54-56].

MspI polymorphisms in the 3'-flanking region of the *CYP1A1* are completely linked with the Ile-Val substitution in exon 7 in Caucasians, which has also been associated experimentally with increased catalytic activity^[2]. However, this complete linkage between *MspI* and Ile-Val substitution could not be found in Asian population^[41,49]. The previous five studies on this polymorphism and esophageal cancer risk showed different results. Only one study in Caucasians showed the *MspI* non-wild allele, which was completely linked with the *Val* allele in control group to significantly increase the risk of esophageal cancer (Table 2). The meta-analysis showed no significance with ORs around unity (Table 2). This may be because the *MspI* polymorphism itself does not alter activity of the *CYP1A1* enzyme. Increased enzyme activity^[2] and susceptibility to esophageal cancer^[45] in Caucasians may be because of the high association between the *MspI* polymorphism and the Ile-Val substitution in exon 7. This should be clarified in further studies.

In contrast to *CYP1A1*, no association was found in the present meta-analysis with the *CYP2E1* *c2* allele. *CYP2E1*

Table 4 Results of meta-regression analysis and Egger's test for publication bias

	Number of studies	Egger's test For publication bias ¹ P	Results of meta-regression test				
			Asian Yes/no Coefficient ³	Chinese Yes/no Coefficient ³	Publication Year Coefficient ³	Design 1 or 2 ² Coefficient ³	Matching Yes/no Coefficient ³
<i>CYP1A1</i> Ile-Val	8	0.96	-0.1	0.19	0.11	-0.2	-0.22
<i>CYP1A1 MspI</i>	5	0.96	-0.42	-0.67	-0.9	0.01	-0.01
<i>CYP2E1 RsaI</i>	5	0.32	1	-0.89	0.76	1.3	-0.13
<i>GSTM1</i>	12	0.04	0.06	0.21	0.25	-0.34	-0.16
<i>GSTT1</i>	6	0.11	0.34	0.34	-0.16	0.37	0.16
<i>GSTP1</i>	7	0.99	-1.25 ⁴	-0.07	0.88	0.4	0.02

¹P-value of Egger's test for publication bias. ²Study design: 1: population-based case-control study, 2: hospital-based case-control study. ³Coefficient in the meta-regression analysis indicates the summary OR change in the value for that factor. For example, the OR for studies examining *CYP1A1* Ile-Val polymorphism only among Asian population is the value of (summary OR-0.1). ⁴Indicates statistical significance at the level of *P*<0.10.

is primarily responsible for metabolic activation of many low molecular weight carcinogens^[3], including certain nitrosamines, which may be involved in carcinogenesis of the esophagus. The variant $\alpha 2$ allele appears to be associated with decreased enzyme activity^[5]. Possible explanations for the lack of any association found here include (1) a small number of studies, (2) greater influence of other polymorphisms in *CYP2E1* such as *Dra1* and tandem repeat polymorphisms and (3) difference in exposure level to xenobiotics across the study populations. These issues must be considered in future investigations.

We also failed to find any association with GST gene polymorphisms. *GSTM1* and *GSTT1* null type cannot encode functional enzymes and therefore affected individuals would be expected to be more vulnerable to carcinogens. The *GSTP1* Ile104Val substitution may also change the enzyme activity of GSTP1 and modulate susceptibility. A meta- and pooled analyses on head and neck cancer showed *GSTM1* (OR = 1.32, 95%CI, 1.07-1.62) and *GSTT1* (OR = 1.25, 95%CI, 1.00-1.57) to modestly increase susceptibility^[54], but most previous studies on esophageal cancer and our meta-analysis failed to find any relationship. Possible explanations include (1) significance of these enzymes may vary with the cancer site; (2) GSTs metabolize a variety of xenobiotics with a large overlap in the substrate specificity and individuals lacking only one functional enzyme also can metabolize the carcinogens by other GST enzymes; and (3) publication bias may exist together with heterogeneity across studies, which may decrease the statistical power.

As is often the case with meta-analyses, there were several limitations to the present study. Possible sources of heterogeneity, such as differences in study design, publication year and countries/ethnicities, must be considered although meta-regression did not demonstrate the existence of any significant variation except in ethnicity for *GSTP1*. Possible publication bias is another threat for our summary ORs, although it was detected only for *GSTM1*. In addition, as adjusted ORs are much more accurate than crude ORs but not available for certain studies, and adjusted and matching factors differed across the studies, residual confounding might have influenced our analysis. Finally, literature-based meta-analysis rather than individual data-based meta-analysis could be a potential source of bias.

In conclusion, we found here a significant association between the *CYP1A1* Ile-Val polymorphism and the risk of esophageal cancer by systematic review. Harboring the *Val* allele, expected to increase the internal exposure to activated carcinogens, thus appears to elevate the risk of esophageal cancer.

ACKNOWLEDGMENTS

The first author, Chun-Xia Yang, was the recipient of a "Special Japan-China Sasakawa Medical Fellowship" during the period of research for and compilation of this paper.

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Science Editor Guo SY Language Editor Elsevier HK