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A meta-analysis of the association between CTLA-4 +49 A/G, −318 C/T, and IL-1 polymorphisms and susceptibility to cervical cancer

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Our aim was to explore whether cytotoxic T lymphocyte antigen-4 (CTLA-4) and interleukin-1 (IL-1) polymorphisms are associated with cervical cancer.

A meta-analysis was conducted on the associations between the CTLA-4 +49 A/G, −318 C/T, IL-1B −511 C/T, and IL-1 receptor antagonist (IL-1RN) polymorphisms and cervical cancer.

We included 15 studies on cervical cancer. The meta-analysis showed a significant association between cervical cancer and the CTLA-4 +49 G allele when all studies were considered (OR = 0.822, 95% CI 0.731–0.924, $p = 0.001$). Stratification by ethnicity indicated an association between the CTLA-4 +49 GG+GA genotype and cervical cancer in East Asians (OR = 0.708, 95% CI 0.532–0.943, $p = 0.018$). However, no association was found between cervical cancer and the CTLA-4 −318 C/T polymorphism. Meta-analysis showed an association between cervical cancer and the IL-1B −511 T allele (OR = 1.380, 95% CI 1.048–1.816, $p = 0.022$), and stratification by ethnicity indicated an association between the IL-1B −511 CC+CT genotype in East Asians (OR = 1.622, 95% CI 1.227–2.43, $p = 0.001$). An association was found between the IL-1RN*2 allele and cervical cancer in Indians, but not in Europeans (OR = 2.154, 95% CI 1.547–2.948, $p = 1.6 \times 10^{-7}$; OR = 1.269, 95% CI 0.969–1.661, $p = 0.083$).

The meta-analysis suggests that the CTLA-4 +49 A/G and IL-1B −511 C/T polymorphisms are associated with cervical cancer in East Asians, and that the IL-1RN VNTR polymorphism is associated with cervical cancer in Indians.

Key words: cervical cancer, cytotoxic T lymphocyte antigen-4, interleukin-l, polymorphism, meta-analysis

Cervical cancer is a malignant neoplasm arising from cells originating in the cervix uteri. It is the second most common cancer in women worldwide and a major cause of mortality in developing countries [1]. Although its etiology is not fully understood, it has been suggested that cervical cancer results from the interaction between a susceptible genome and various environmental factors.

Cytotoxic T lymphocyte associated antigen-4 (CTLA-4) is a critical down-regulatory molecule, expressed in T cells, that plays a major role in inhibiting T cell activation and peripheral tolerance [2]. It has been suggested that CTLA-4 may play a role in the regulation of self-tolerance by the immune system and in the pathogenesis of cancers and autoimmune disorders [3]. Therefore it is highly likely that defective CTLA-4 expression and function are associated with cancers [4]. More than 100 polymorphisms have been identified in the CTLA-4 gene. Of these, CTLA-4 +49 A/G (rs231775) and −318 C/T (rs5742909) have been studied the most in association with cervical cancer. CTLA-4 +49 A/G causes a threonine-to-alanine change at amino acid 17 of CTLA-4, which may reduce its cell surface expression [5]. Carriers of the CTLA-4 −318 T allele show significantly increased expression of CTLA-4 mRNA and protein [6].

Interleukin-1 (IL-1) initiates the recruitment of immune cells and inflammation [7]. Interleukin-1A (IL-1A) and interleukin-1B (IL-1B) are pro-inflammatory cytokines that bind to the IL-1 receptor, resulting in signal transduction. IL-1 receptor antagonist (IL-1RN) is a competitive inhibitor that does not elicit intracellular signaling when bound to the same receptor [8]. IL-1RN is a member of the IL-1 cytokine family; it inhibits the activity of IL-1A and IL-1B and modulates the immune response [9]. IL-1RN has a variable number tandem repeat (VNTR) polymorphism in intron 2. IL1RN*2 is associated with increased activity of IL1B, which plays a key role as a pro-inflammatory cytokine [10, 11]. The IL-1B −511 C/T (rs16944) and IL-1RN VNTR polymorphisms have been studied most frequently in relation to cervical cancer.

However, previous studies considering the relations between the CTLA-4 and IL-1 polymorphisms and cervical

cancer have produced inconsistent results [12-25]. These inconsistencies may be due to the sample sizes, racial/ethnic differences in allele frequencies, or publication bias [26]. Therefore, in order to overcome the limitations of individual studies, resolve inconsistencies, and reduce the likelihood that random errors are responsible for false-positive or false-negative associations, we turned to meta-analysis [26-28]. The aim of the present study was to determine using meta-analysis whether the CTLA-4 and IL-1 polymorphisms are associated with cervical cancer.

Materials and methods

Identification of eligible studies and data extraction. We performed a search of studies that examined the associations between the CTLA-4 and IL-1 polymorphisms and cervical cancer. The MEDLINE and EMBASE citation databases were used to identify articles published up to June 2013 in which these two polymorphisms were analyzed in patients with cervical cancer. We used words and phrases such as 'cytotoxic T lymphocyte associated antigen,' 'CTLA-4,' 'interleukin-1,' 'IL-1,' 'polymorphism,' and 'cervical cancer' as Medical Subject Headings (MeSH) and search text. References in studies that were identified were investigated, to find additional studies that were not indexed by MEDLINE or EMBASE. Genetic association studies were also eligible for inclusion if they analyzed the distributions of the CTLA-4 +49 A/G, −318 C/T, IL-1B −511C/T, or IL1-RN VNTR polymorphisms in individuals who had cervical cancer and controls who did not. Inclusion criteria were: (1) case–control study design; (2) original data; and (3) sufficient genotype data to calculate odds ratios (ORs). No language restriction was applied. Exclusion criteria were: (1) overlapping data; (2) inability to ascertain the numbers of null and wild genotypes; (3) family members studied, because analyses that include family members include linkage considerations; and (4) genotype distribution in controls not consistent with Hardy–Weinberg equilibrium, because this suggests the possibility of genotyping errors or bias during control-selection. Information on methods and results was extracted from the original publications by two people working independently. Discrepancies were resolved by consensus or a third person. The following information was extracted from each study: author(s), year of publication, ethnicity of the study population, demographics, and numbers of cases and controls for each genotype of the CTLA-4 +49 A/G, −318 C/T, IL-1B −511C/T, and IL-1RN VNTR polymorphisms. Frequencies of alleles were calculated from the corresponding genotype distributions.

Evaluation of publication bias. Funnel plots are often used to detect publication bias. However, this method requires a range of studies of varying sizes and involves subjective judgments, so we evaluated publication bias using Egger's linear regression test [29], which measures the asymmetry of the funnel plot using odds ratios on a natural logarithm scale.

Evaluations of statistical association. Chi-square tests were used to determine whether observed frequencies of genotypes conformed to Hardy–Weinberg expectations. Meta-analyses were performed using (1) allelic contrast, (2) contrast of homozygotes, and (3) recessive and (4) dominant models. Point estimates of risks, ORs, and 95% confidence intervals (CIs) were estimated for each study. Cochran's Q-statistic was used to assess within- and between-study variation or heterogeneity. This test assesses the null hypothesis that all studies were evaluating the same effect. I^2 values were used to quantify the effect of heterogeneity. Values of I^2 range between 0% and 100% and represent the proportion of between-study variability that can be attributed to heterogeneity rather than chance [30]. I^2 values of 25%, 50%, and 75% were regarded as low, moderate, and high respectively. The fixed effects model assumes that a genetic factor has the same effect on the risk of cervical cancer across all studies, and that the variations observed between studies are caused by chance alone. The random effects model assumes that different studies show substantial diversity and assesses both within-study sampling error and between-study variation. When study groups are homogeneous, the two models are similar, but if this is not the case the random effects model usually provides wider CIs than the fixed effects model. The random effects model is used in the presence of significant between-study heterogeneity [31]. Statistical manipulations were undertaken using Comprehensive Meta-Analysis software (Biostat, Englewood, NJ, USA).

Results

Studies included in the meta-analysis. Electronic and manual searching identified 30 articles, and 17 of these were selected for a full review based on title and abstract details [12-25, 32, 33]. Two were excluded because one was about a different disease [32] and one was about a different polymorphism [33]; thus, 15 studies in total met our inclusion criteria. These consisted of 8 studies on the CTLA-4 polymorphism [12-18] and 7 on the IL-1 polymorphism [19-25] (Fig. 1). Studies on the CTLA-4 polymorphism used a total of 2,841 patients and 2,558 controls (5,399 study subjects). Two of the study populations were European, four East Asian, one Indian, and one Arab. The CTLA-4 +49 G/A polymorphism was examined by 5 studies and the CTLA-4 −318 C/T polymorphism by 6.

Studies on the IL-1 polymorphism used a total of 1,327 patients and 1,492 controls (2,819 study subjects). Two of the populations were European, two East Asian, two Indian, and one Arab. The IL-1B −511 C/T and IL-1RN VNTR polymorphisms were examined by 4 studies each. Selected characteristics of the studies we used are summarized in Table 1.

Meta-analysis of association between CTLA-4 +49 A/G and −318 C/T polymorphisms and cervical cancer. A summary of the findings of our meta-analyses on the association between the CTLA-4 +49 A/G and −318 C/T polymorphisms and cervical cancer is shown in Table 2. Meta-analysis of the CTLA-4 +49 A/G polymorphism showed a significant

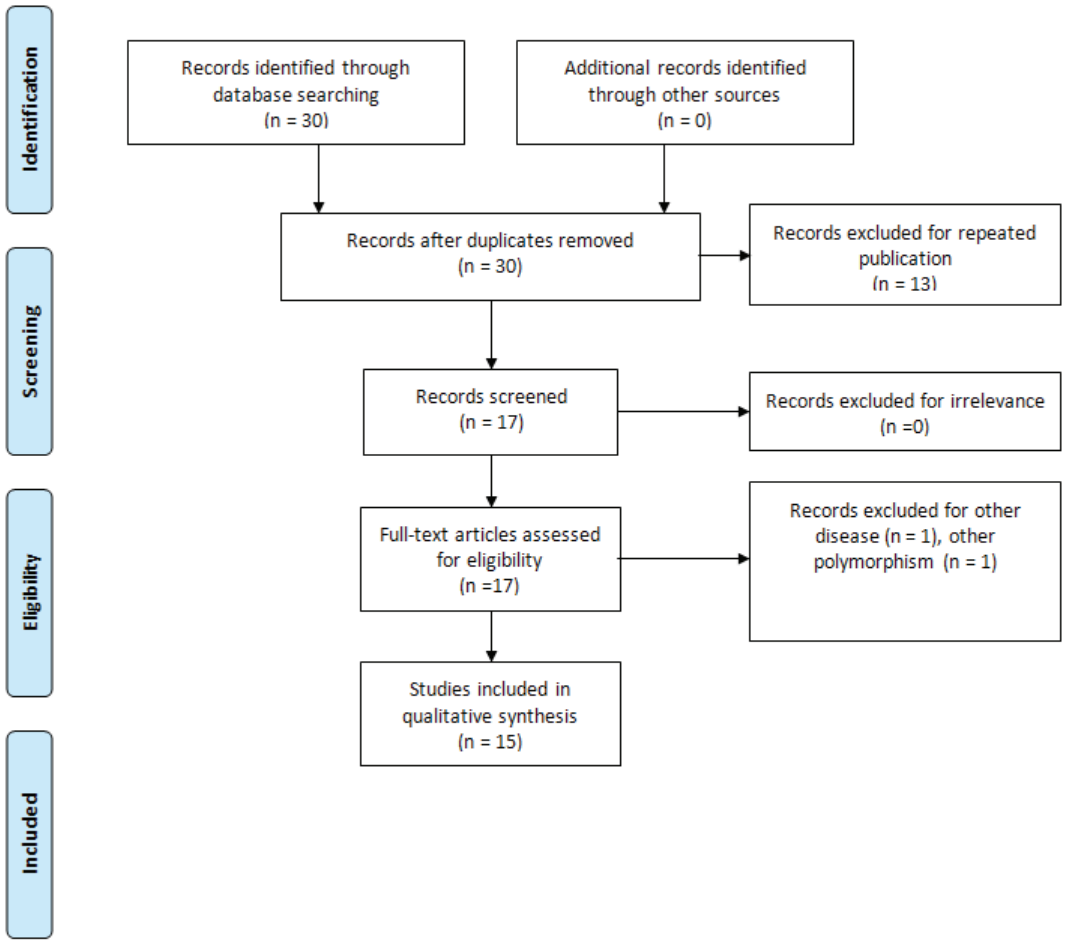


Figure 1. Flow diagram of literature search strategy and selection of studies in the meta-analysis.

Table 1. Characteristics of the individual studies included in the meta-analysis.

Study [Ref]	Country	Population	Numbers		Polymorphisms studied	Findings (p-value of association)
			Case	Control		
Gokhale, 2013 [12]	India	Indian	104	162	CTLA-4 +49 A/G, −318 C/T	CTLA-4 49 G/A ($p = 0.04$), −318 C/T ($p = 0.03$)
Li, 2011 [13]	China	East Asian	314	320	CTLA-4 +49 A/G	NS
Jiang, 2011 [14]	China	East Asian	100	100	CTLA-4 +49 A/G, −318 C/T	CTLA-4 49 G/A (NS), −318 C/T ($p = 0.001$)
Rahimifar, 2010 [15]	Iran	Arab	55	110	CTLA-4 +49 A/G, −318 C/T	CTLA-4 49 G/A (NS), −318 C/T ($p = 0.021$)
Hu, 2010 [16]	China	East Asian	696	709	CTLA-4 +49 A/G	CTLA-4 49 G/A ($p = 0.008$)
Ivansson, 2010 [17]	Sweden	European	1281	554	CTLA-4 −318 C/T	NS
Pawlak, 2010	Poland	European	147	225	CTLA-4 −318 C/T	CTLA-4 −318 C/T ($p = 0.003$)
Su, 2007 [18]	Taiwan	East Asian	144	378	CTLA-4 −318 C/T	NS
Sousa, 2012 [19]	Portugal	European	228	196	IL-1RN VNTR	IL-1RN VNTR ($p = 0.025$)
Al-Tahhan, 2011 [20]	Egypt	Arab	100	50	IL-1B −511 C/T	IL-1B −511 C/T ($p = 0.008$)
Qian, 2010 [21]	China	East Asian	404	404	IL-1RN VNTR, IL-1B −511 C/T	IL-1RN VNTR (NS), IL-1B −511 C/T ($p = 0.047$)
Singh, 2008 [22]	India	Indian	150	162	IL-1RN VNTR, IL-1B −511 C/T	IL-1RN VNTR ($p = 1.1 \times 10^{-5}$), IL-1B −511 C/T ($p = 0.0007$)
Tamandani, 2008 [23]	India	Indian	150	209	IL-1RN VNTR	NS
Kang, 2007 [24]	Korea	East Asian	182	364	IL-1B −511 C/T	IL-1B −511 C/T ($p < 0.001$)
Mustea, 2003 [25]	Germany	European	113	107	IL-1RN VNTR	NS

Ref reference, *NS* not significant

Table 2. Meta-analysis of the associations between the CTLA-4 +49 A/G and −318 C/T polymorphisms and cervical cancer

Polymorphism	Population	No. of studies	Test of association			Test of heterogeneity		
			OR	95% CI	*p*	Model	*p*	I^2
CTLA-4 +49 G vs. A	Overall	5	0.822	0.731–0.924	0.001	R	0.105	47.6
	East Asian	3	0.881	0.690–1.124	0.327	R	0.060	64.5
GG vs. GA+AA (recessive)	Overall	5	0.846	0.722–0.992	0.039	R	0.202	32.8
	East Asian	3	0.856	0.705–1.010	0.066	F	0.104	55.7
GG + GA vs. AA (dominant)	Overall	5	0.652	0.512–0.831	0.001	F	0.232	28.4
	East Asian	3	0.708	0.532–0.943	0.018	F	0.155	46.2
GG vs. AA	Overall	5	0.632	0.480–0.830	0.001	F	0.122	45.0
	East Asian	3	0.732	0.425–1.262	0.262	R	0.076	61.1
CTLA-4 −318 T vs. C	Overall	6	1.146	0.698–1.881	0.590	R	0.000	79.3
	European	2	1.412	0.772–2.584	0.263	R	0.0021	81.2
	East Asian	2	1.525	0.310–7.516	0.604	R	0.000	91.7
TT vs. TC+CC (recessive)	Overall	6	1.449	0.606–3.462	0.404	F	0.744	0
	European	2	1.361	0.477–3.884	0.565	F	0.228	31.2
	East Asian	2	2.210	0.369–13.23	0.385	F	0.170	0
TT + TC vs. CC (dominant)	Overall	6	1.133	0.660–1.946	0.650	R	0.000	80.2
	European	2	1.441	0.767–2.707	0.256	R	0.027	79.6
	East Asian	2	1.500	0.257–8.743	0.652	R	0.000	92.5
TT vs. CC	Overall	6	1.470	0.615–3.516	0.386	F	0.669	0
	European	2	1.420	0.497–4.056	0.513	F	0.192	41.3
	East Asian	2	2.174	0.362–13.04	0.396	F	0.700	0

CTLA-4 cytotoxic T lymphocyte antigen-4, *OR* odds ratio, *CI* confidence interval, *F* fixed effects model, *R* random effects model

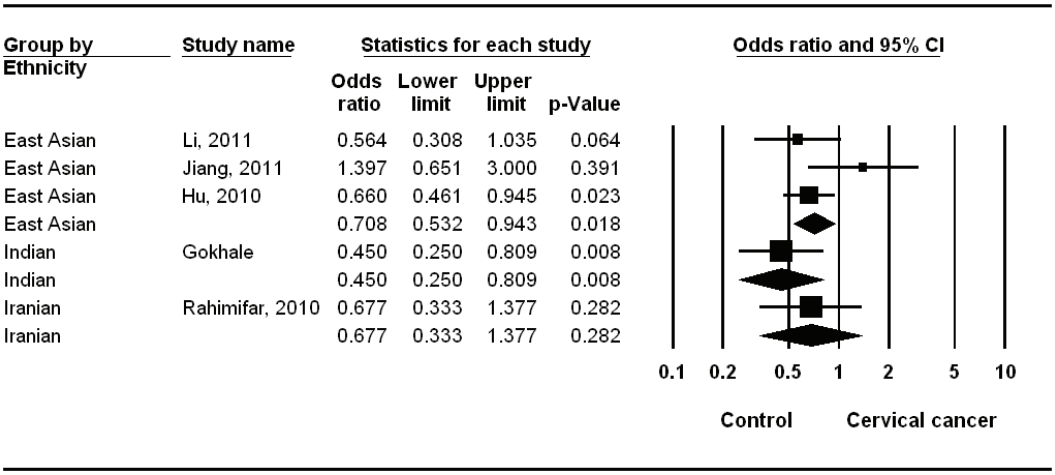
association between cervical cancer and the CTLA-4 +49 G allele when all studies were considered (OR = 0.822, 95% CI 0.731–0.924, $p = 0.001$). The association between cervical cancer and the CTLA-4 +49 A/G polymorphism was found to be significant using the recessive model, the dominant model, and contrast of homozygotes (these three models gave respectively OR = 0.846, 95% CI 0.722–0.992, $p = 0.039$; OR = 0.652, 95% CI 0.512–0.831, $p = 0.001$; and OR = 0.632, 95% CI 0.480–0.830, $p = 0.001$). Stratification by ethnicity indicated an association between the CTLA-4 +49 GG+GA genotype and cervical cancer in East Asians (OR = 0.708, 95% CI 0.532–0.943, $p = 0.018$; see Figure 2). When all studies were considered, meta-analysis revealed no association between cervical cancer and the CTLA-4 −318 C allele (OR = 1.146, 95% CI 0.698–1.881, $p = 0.590$; Table 2). Stratification by ethnicity indicated no association between

Table 3. Meta-analysis of associations between the IL-1B −511 C/T and the IL-1RN VNTR polymorphisms and cervical cancer

Polymorphism	Population	No. of studies	Test of association			Test of heterogeneity		
			OR	95% CI	*p*	Model	*p*	I^2
IL-1B −511 T vs. C	Overall	4	1.380	1.048–1.816	0.022	R	0.018	70.1
	East Asian	2	1.131	0.968–1.321	0.120	F	0.447	0
TT vs. TC+CC (recessive)	Overall	4	1.321	0.779–2.242	0.302	R	0.001	81.5
	East Asian	2	0.896	0.687–1.167	0.415	F	0.129	56.6
TT+TC vs. CC (dominant)	Overall	4	1.720	1.336–2.214	2.6×10^{-6}	F	0.339	10.8
	East Asian	2	1.622	1.227–2.143	0.001	F	0.151	51.6
TT vs. CC	Overall	4	1.741	1.281–2.366	3.9×10^{-5}	F	0.206	34.
	East Asian	2	1.459	1.017–2.073	0.035	F	0.637	0
IL1RN*2 vs. others	Overall	4	1.415	0.984–2.034	0.061	F	0.009	70.4
	European	2	1.269	0.969–1.661	0.083	F	0.940	0
	Indian	2	2.154	1.547–2.948	1.6×10^{-7}	F	0.361	0

OR odds ratio, *CI* confidence interval, *R* random effects model, *F* fixed effects model

A



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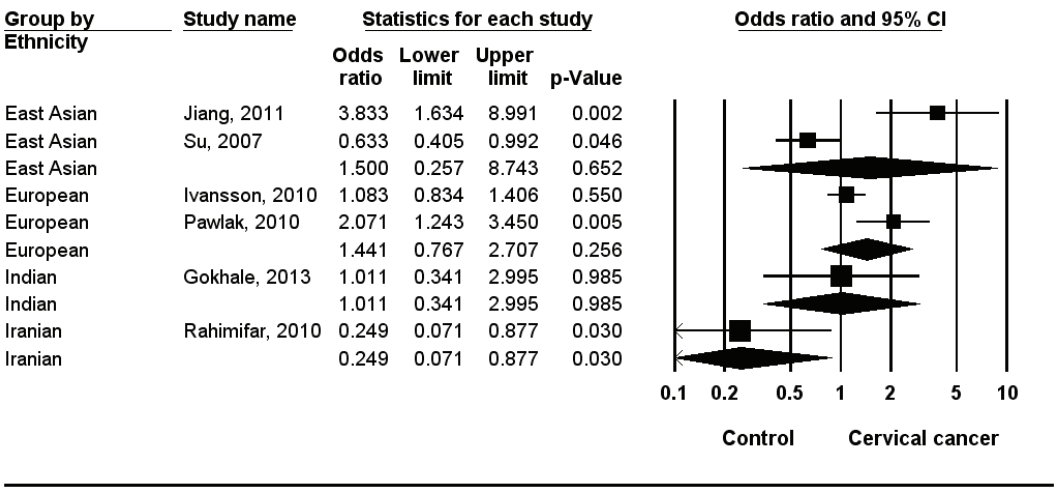
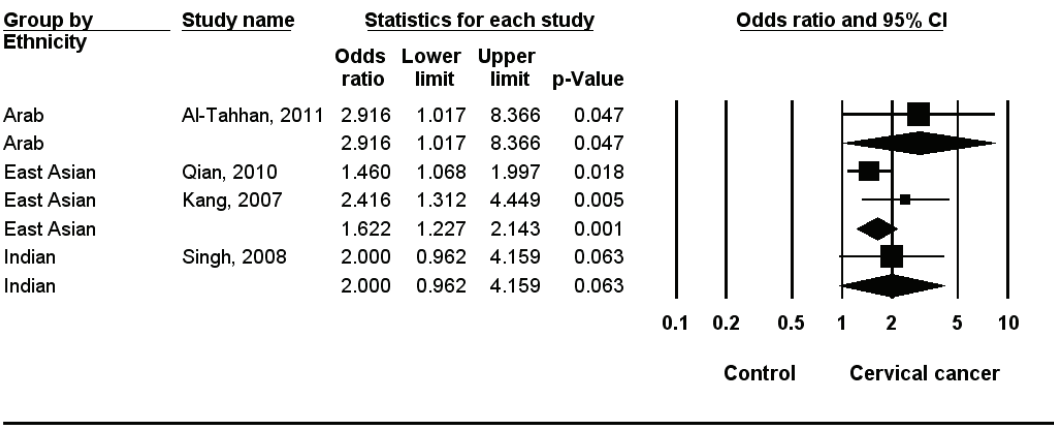


Figure 2. Odds ratios and 95% CIs for studies and pooled data on the association between the CTLA-4 +49 GG+GA (A) and CTLA-4 −318 TT+TC (B) genotypes and cervical cancer in each ethnic group.

the CTLA-4 −318 C allele and cervical cancer in Europeans and Asians (OR = 1.412, 95% CI 0.772–2.584, $p = 0.263$; OR = 1.525, 95% CI 0.310–7.516, $p = 0.604$). No association was found between cervical cancer and the CTLA-4 −318 C/T polymorphism using the recessive model, the dominant model, or homozygote contrast (see Figure 2).

Meta-analysis of association between IL-1B −511C/T and IL-1RN VNTR polymorphisms and cervical cancer. Meta-analysis showed a significant association between cervical cancer and the IL-1B −511 T allele when all studies were considered (OR = 1.380, 95% CI 1.048–1.816, $p = 0.022$). (See Table 3 for the meta-analyses in this section.) An association was also found between cervical cancer and the IL-1B −511 C/T polymorphism using the dominant model or contrast of homozygotes (OR = 1.720, 95% CI 1.336–2.214, $p = 2.6 \times 10^{-6}$; OR = 1.741, 95% CI 1.281–2.366, $p = 3.9 \times 10^{-5}$). Stratification by ethnicity indicated an association between the IL-1B −511 C/T polymorphism in East Asians, using the dominant model or contrast of homozygotes (OR = 1.622, 95% CI 1.227–2.43 $p = 0.001$; OR = 1.459, 95% CI 1.017–2.073, $p = 0.035$; see Figure 3). When all studies were considered, meta-analysis revealed a trend of association between cervical cancer and the IL-1RN*2 allele (OR = 1.415, 95% CI 0.984–2.034, $p = 0.061$). Ethnicity-specific meta-analysis revealed an association between the IL-1RN*2 allele and cervical cancer in Indians, but not in Europeans (OR = 2.154, 95% CI 1.547–2.948, $p = 1.6 \times 10^{-7}$; OR = 1.269, 95% CI 0.969–1.661, $p = 0.083$; see Figure 3).

A



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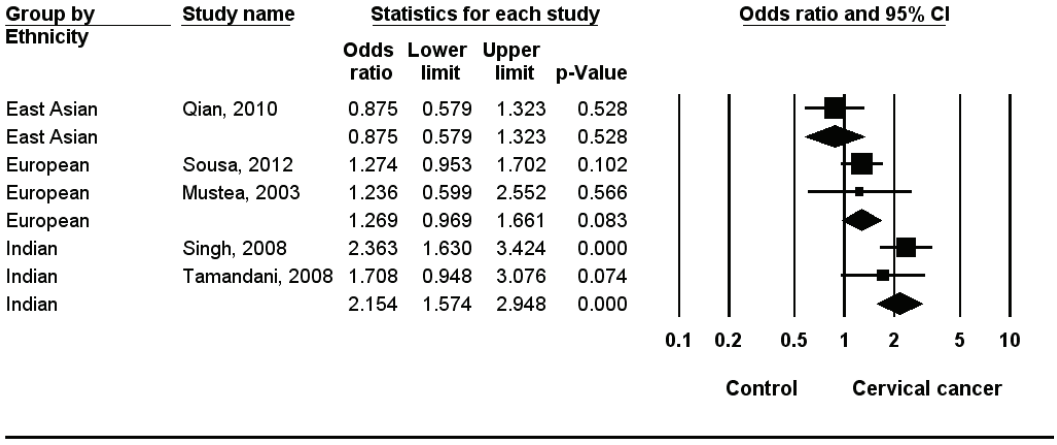

Figure 3. Odds ratios and 95% CIs for studies and pooled data on the association between the IL-1B −511 TT+TC genotype (A) and IL-1RN*2 allele (B) and cervical cancer in each ethnic group.

Heterogeneity and publication bias. Some heterogeneity was found in the meta-analyses of the CTLA-4 and IL-1B −511 C/T polymorphisms, but no between-study heterogeneity was found for the IL-1RN VNTR polymorphisms. Funnel plots are commonly used to detect publication bias, but in our meta-analyses it was difficult to interpret the funnel plots because the number of studies was relatively small. Publication bias causes a disproportionate number of positive studies, and poses a problem for meta-analyses, but Egger's regression test showed no evidence of publication bias in these meta-analyses (all *p*-values were greater than 0.1; see Figure 4).

Discussion

The multifactorial natures of cervical cancers are well recognized, but genetic factors are considered strong determinants of these diseases, and this has encouraged researchers to search for the genes responsible. CTLA-4 is a member of the same family of cell surface molecules as CD28, and it is also responsible for its involvement in the regulation of T cells [2]. CTLA-4 signaling mediates a negative regulator in both cellular and humoral responses, and mediates antigen-specific apoptosis [34]. Negative signaling via CTLA-4 plays an active role in the regulation of autoreactive T cells, and the disruption of the normal physiologic control provided by CTLA-4 can contribute to the pathogenesis of cancers. CTLA-4 polymorphisms have been found to be associated with cancers as well as several autoimmune disorders [3, 35, 36]. IL-1 is a potent inflammatory cytokine that plays a key role in immune regulation [8] and has been associated with pathogenesis of cancer [37].

This study addresses the association between the CTLA-4 and IL-1 polymorphisms and cervical cancer. Although our

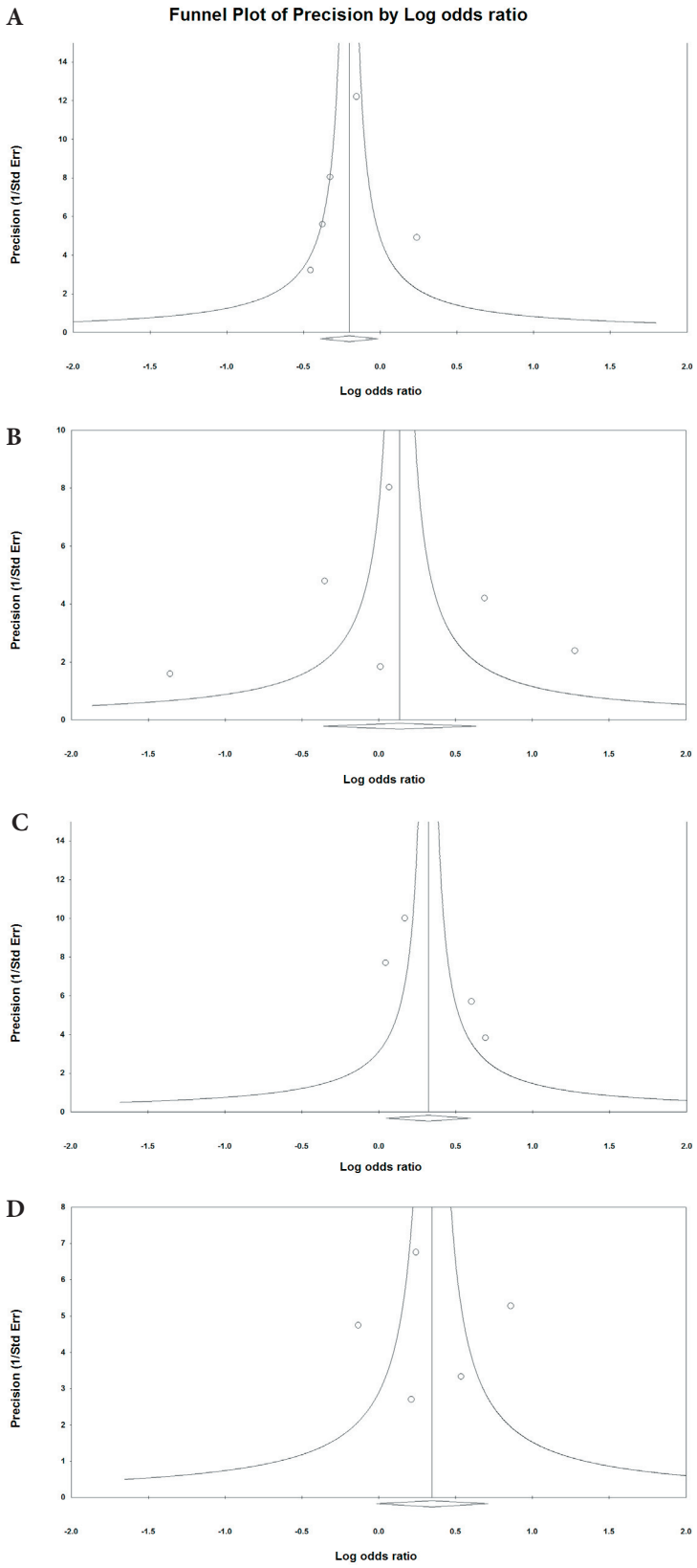

Figure 4. Funnel plot of studies of association between the CTLA-4 +49G (A), −318C (B), IL1B −511T (C), and IL-1RN*2 (D) alleles and cervical cancer in all studies (Egger's regression test *p*-values: 0.882, 0.940, 0.200, 0.988 respectively).

findings do not provide evidence for an association between the CTLA-4 −318 C/T polymorphisms and cervical cancer, the meta-analysis revealed that the CTLA-4 +49 A/G and IL-1B −511 C/T polymorphisms are associated with cervical cancer risk in East Asians. Another meta-analysis showed an association between the IL-1RN VNTR polymorphism and cervical cancer in Indians, but not in Europeans.

Our finding that the CTLA-4 +49 G allele plays a protective role against the development of cervical cancer is consistent with results from functional studies of the CTLA-4 +49 A/G polymorphism. A G allele at exon-1 +49, which causes a threonine-to-alanine change at amino acid 17, leads to less effective CTLA-4 export to the membrane and enhanced T cell proliferation, reducing CTLA-4 expression [5]. Subjects with the CTLA-4 +49 GG genotype have reduced CTLA-4 expression upon T cell activation [5]. Immune responses mediated by T cells are important in controlling HPV-associated cancer [38]. Reduced CTLA-4 expression or function may be associated with cervical cancer risk. The CTLA-4 +49 GG genotype may decrease susceptibility to persistent human papillomavirus (HPV) infection, thus conferring a protective effect against cervical cancer. The CTLA-4 −318 polymorphism may affect the expression of CTLA-4. The −318T allele is associated with higher levels of promoter activity than the −318C allele [6]. Therefore, the presence of the −318T allele may contribute to up-regulation of the expression of CTLA-4, and consequently represents one mechanism to inhibit excessive immune activity.

The present study supports the view that the changes in CTLA-4 expression due to functional polymorphism in CTLA-4 may increase susceptibility to cervical cancer. However, the result of the meta-analysis of the CTLA-4 −318 C/T polymorphism is not consistent with the results of functional studies of CTLA-4. It may not be uncommon that epidemiologic results disagree with the results of functional studies, because cervical cancer is a complex disease, and multiple genes, genetic backgrounds, and environmental factors contribute to its development.

Polymorphisms in cytokine genes can modify the risk of cervical cancer by influencing the immune response to HPV infection [39]. IL-1 reduces apoptosis by changing the ratio of BCL-2/BAX proteins [40] and may lead to increased p53 mutation load [41]. Therefore, increased levels of IL-1 may play a role in cervical cancer susceptibility. The IL-1 −511 C/T polymorphism has been found to be associated with increased intracellular IL-1B production [42]. IL-1RN regulates IL-1 activity by binding to the IL-1 receptor [43]. It has been suggested that IL1RN*2 may have functional importance, because the IL1RN VNTR contains possible binding sites for transcription factors [10, 11, 43]. The relative levels of IL-1RN and IL-1 may determine whether a proinflammatory response will be initiated and persist or instead be terminated. IL1RN*2 is associated with enhanced IL1B levels [44], results in the lowest IL1RN/IL1B ratio, and was associated with a heightened proinflammatory immune response [45, 46]. The results of the meta-analysis of the IL-1B −511 C/T and IL-1RN VNTR polymorphisms are consistent with the results of functional studies of the IL-1 polymorphisms. The present study supports the view that the changes in IL-1 function due to functional polymorphism may increase susceptibility to cervical cancer. However, we cannot rule out the possibility that the −511 C/T polymorphism may be affecting the cancer susceptibility due to its linkage with the other polymorphism at the IL-1 locus, which can directly influence the expression of the IL-1B gene.

Our study has some limitations that require consideration. First, heterogeneity and confounding factors may have distorted the analysis. Analysis of data stratified by age, smoking, parity, or HPV status would have provided more information, but these variables were not available. Second, haplotype analysis might have provided more information and would have been more powerful than single polymorphism analysis. However, meta-analysis of haplotypes was not possible due to inadequate haplotype data. Third, the polymorphisms might be associated with the stage of cervical cancer as well as the subject's susceptibility. However, the small amount of data did not allow us to examine possible associations with the stage of cancer. Fourth, the number of studies was too small to enable us to draw conclusions that hold across all the ethnic groups, and it was too small to enable conclusions on certain specific ethnic groups.

In conclusion, this meta-analysis demonstrates that the CTLA-4 +49 A/G and IL-1B −511 C/T polymorphisms are associated with cervical cancer in East Asians, and the IL-1RN VNTR polymorphism is associated with it in Indians. These findings suggest the need for further investigation of the associations between the CTLA-4 and IL-1 polymorphisms and cervical cancer. In particular, larger studies in populations with different ethnicities are necessary to explore the roles played by these two polymorphisms during the pathogenesis of cervical cancer.

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References

[1] PARKIN DM. Global cancer statistics in the year 2000. The lancet oncology 2001; 2: 533–43. http://dx.doi.org/10.1016/S1470-2045(01)00486-7

[2] GREENWALD RJ, OOSTERWEGEL MA, VAN DER WOUDE D, KUBAL A, MANDELBROT DA et al. CTLA-4 regulates cell cycle progression during a primary immune response. Eur J Immunol 2002; 32: 366–73. http://dx.doi.org/10.1002/1521-4141(200202)32:2<366::AID-IMMU366>3.0.CO;2-5

[3] LEE YH, HARLEY JB, NATH SK. CTLA-4 polymorphisms and systemic lupus erythematosus (SLE): a meta-analysis. Human genetics 2005; 116: 361–7. http://dx.doi.org/10.1007/s00439-004-1244-1

[4] QUEZADA SA, PEGGS KS. Exploiting CTLA-4, PD-1 and PD-L1 to reactivate the host immune response against cancer. British journal of cancer 2013; 108: 1560–5. http://dx.doi.org/10.1038/bjc.2013.117

[5] UEDA H, HOWSON JM, ESPOSITO L, HEWARD J, SNOOK H et al. Association of the T-cell regulatory gene CTLA4 with susceptibility to autoimmune disease. Nature 2003; 423: 506–11. http://dx.doi.org/10.1038/nature01621

[6] WANG XB, ZHAO X, GISCOMBE R, LEFVERT AK. A CTLA-4 gene polymorphism at position -318 in the promoter region affects the expression of protein. Genes and immunity 2002; 3: 233–4. http://dx.doi.org/10.1038/sj.gene.6363869

[7] DINARELLO CA. The IL-1 family and inflammatory diseases. Clinical and experimental rheumatology 2002; 20: S1–13.

[8] STYLIANOU E, SAKLATVALA J. Interleukin-1. The international journal of biochemistry & cell biology 1998; 30: 1075–9. http://dx.doi.org/10.1016/S1357-2725(98)00081-8

[9] CARTER DB, DEIBEL MR, JR., DUNN CJ, TOMICH CS, LABORDE AL et al. Purification, cloning, expression and biological characterization of an interleukin-1 receptor antagonist protein. Nature 1990; 344: 633–8. http://dx.doi.org/10.1038/344633a0

[10] TARLOW JK, BLAKEMORE AI, LENNARD A, SOLARI R, HUGHES HN et al. Polymorphism in human IL-1 receptor antagonist gene intron 2 is caused by variable numbers of an 86-bp tandem repeat. Human genetics 1993; 91: 403–4. http://dx.doi.org/10.1007/BF00217368

[11] STEINKASSERER A, KOELBLE K, SIM RB. Length variation within intron 2 of the human IL-1 receptor antagonist protein gene (IL1RN). Nucleic acids research 1991; 19: 5095. http://dx.doi.org/10.1093/nar/19.18.5095

[12] GOKHALE P, KERKAR S, TONGAONKAR H, SALVI V, MANIA-PRAMANIK J. CTLA-4 gene polymorphism at position +49 A>G in exon 1: a risk factor for cervical cancer in Indian women. Cancer genetics 2013; 206: 154–61. http://dx.doi.org/10.1016/j.cancergen.2013.04.003

[13] LI H, ZHOU YF, GUO HY, SUN T, ZHANG WH et al. [Association between CTLA-4 gene polymorphism and susceptibility to cervical cancer]. Zhonghua zhong liu za zhi [Chinese journal of oncology] 2011; 33: 681–4.

[14] JIANG L, LUO RY, ZHANG W, WANG LR, WANG F et al. [Single nucleotide polymorphisms of CTLA4 gene and their association with human cervical cancer]. Zhonghua yi xue yi chuan xue za zhi = Zhonghua yixue yichuanxue zazhi = Chinese journal of medical genetics 2011; 28: 313–7.

[15] RAHIMIFAR S, ERFANI N, SARRAF Z, GHADERI A. ctla-4 gene variations may influence cervical cancer susceptibility. Gynecologic oncology 2010; 119: 136–9. http://dx.doi.org/10.1016/j.ygyno.2010.06.006

[16] HU L, LIU J, CHEN X, ZHANG Y, LIU L et al. CTLA-4 gene polymorphism +49 A/G contributes to genetic susceptibility to two infection-related cancers-hepatocellular carcinoma and cervical cancer. Human immunology 2010; 71: 888–91. http://dx.doi.org/10.1016/j.humimm.2010.05.023

[17] IVANSSON EL, JUKO-PECIREP I, GYLLENSTEN UB. Interaction of immunological genes on chromosome 2q33 and IFNG in susceptibility to cervical cancer. Gynecologic oncology 2010; 116: 544–8. http://dx.doi.org/10.1016/j.ygyno.2009.10.084

[18] PAWLAK E, KARABON L, WLODARSKA-POLINSKA I, JEDYNAK A, JONKISZ A et al. Influence of CTLA-4/CD28/ICOS gene polymorphisms on the susceptibility to cervical squamous cell carcinoma and stage of differentiation in the Polish population. Human immunology 2010; 71: 195–200. http://dx.doi.org/10.1016/j.humimm.2009.11.006

[19] SOUSA H, SANTOS AM, CATARINO R, PINTO D, MOUTINHO J et al. IL-1RN VNTR polymorphism and genetic susceptibility to cervical cancer in Portugal. Molecular biology reports 2012; 39: 10837–42. http://dx.doi.org/10.1007/s11033-012-1979-z

[20] AL-TAHHAN MA, ETEWA RL, EL BEHERY MM. Association between circulating interleukin-1 beta (IL-1beta) levels and IL-1beta C-511T polymorphism with cervical cancer risk in Egyptian women. Molecular and cellular biochemistry 2011; 353: 159–65. http://dx.doi.org/10.1007/s11010-011-0782-9

[21] QIAN N, CHEN X, HAN S, QIANG F, JIN G et al. Circulating IL-1beta levels, polymorphisms of IL-1B, and risk of cervical cancer in Chinese women. Journal of cancer research and clinical oncology 2010; 136: 709–16. http://dx.doi.org/10.1007/s00432-009-0710-5

[22] SINGH H, SACHAN R, GOEL H, MITTAL B. Genetic variants of interleukin-1RN and interleukin-1beta genes and risk of cervical cancer. BJOG : an international journal of obstetrics and gynaecology 2008; 115: 633–8. http://dx.doi.org/10.1111/j.1471-0528.2007.01655.x

[23] TAMANDANI DM, SOBTI RC, SHEKARI M, KAUR S, HURIA A. Impact of polymorphism in IL-1RA gene on the risk of cervical cancer. Archives of gynecology and obstetrics 2008; 277: 527–33. http://dx.doi.org/10.1007/s00404-007-0504-4

[24] KANG S, KIM JW, PARK NH, SONG YS, PARK SY ET AL. Interleukin-1 beta-511 polymorphism and risk of cervical cancer. Journal of Korean medical science 2007; 22: 110–3. http://dx.doi.org/10.3346/jkms.2007.22.1.110

[25] MUSTEA A, SEHOULI J, KONSGEN D, STENGEL D, SOFRONI D et al. Interleukin 1 receptor antagonist (IL-1RA) polymorphism in women with cervical cancer. Anticancer research 2003; 23: 1099–102.

[26] LEE YH, WITTE T, MOMOT T, SCHMIDT RE, KAUFMAN KM, HARLEY JB et al. The mannose-binding lectin gene polymorphisms and systemic lupus erythematosus: two case-control studies and a meta-analysis. Arthritis and rheumatism 2005; 52: 3966–74. http://dx.doi.org/10.1002/art.21484

[27] NATH SK, HARLEY JB, LEE YH. Polymorphisms of complement receptor 1 and interleukin-10 genes and systemic lupus erythematosus: a meta-analysis. Human genetics 2005; 118: 225–34. http://dx.doi.org/10.1007/s00439-005-0044-6

[28] LEE YH, HARLEY JB, NATH SK. Meta-analysis of TNF-alpha promoter -308 A/G polymorphism and SLE susceptibility. European journal of human genetics : EJHG 2006; 14: 364–71. http://dx.doi.org/10.1038/sj.ejhg.5201566

[29] EGGER M, DAVEY SMITH G, SCHNEIDER M, MINDER C. Bias in meta-analysis detected by a simple,

graphical test. BMJ 1997; 315: 629–34. http://dx.doi.org/10.1136/bmj.315.7109.629

[30] HIGGINS JP, THOMPSON SG. Quantifying heterogeneity in a meta-analysis. Stat Med 2002; 21: 1539–58. http://dx.doi.org/10.1002/sim.1186

[31] DERSIMONIAN R, LAIRD N. Meta-analysis in clinical trials. Control Clin Trials 1986; 7: 177–88. http://dx.doi.org/10.1016/0197-2456(86)90046-2

[32] GRIMM C, WATROWSKI R, BAUMUHLNER K, NATTER C, TONG D et al. Genetic variations of interleukin-1 and -6 genes and risk of cervical intraepithelial neoplasia. Gynecologic oncology 2011; 121: 537–41. http://dx.doi.org/10.1016/j.ygyno.2011.02.019

[33] SOBTI RC, KORDI TAMANDANI DM, SHEKARI M, KAUR P, MALEKZADEH K et al. Interleukin 1 beta gene polymorphism and risk of cervical cancer. International journal of gynaecology and obstetrics: the official organ of the International Federation of Gynaecology and Obstetrics 2008; 101: 47–52. http://dx.doi.org/10.1016/j.ijgo.2007.10.014

[34] GRIBBEN JG, FREEMAN GJ, BOUSSIOTIS VA, RENNERT P, JELLIS CL et al. CTLA4 mediates antigen-specific apoptosis of human T cells. Proc Natl Acad Sci U S A 1995; 92: 811–5. http://dx.doi.org/10.1073/pnas.92.3.811

[35] HAN S, LI Y, MAO Y, XIE Y. Meta-analysis of the association of CTLA-4 exon-1 +49A/G polymorphism with rheumatoid arthritis. Hum Genet 2005; 118: 123–32. http://dx.doi.org/10.1007/s00439-005-0033-9

[36] ZHENG J, YU X, JIANG L, XIAO M, BAI B et al. Association between the Cytotoxic T-lymphocyte antigen 4 +49G > A polymorphism and cancer risk: a meta-analysis. BMC cancer 2010; 10: 522. http://dx.doi.org/10.1186/1471-2407-10-522

[37] HE B, ZHANG Y, PAN Y, XU Y, GU L et al. Interleukin 1 beta (IL1B) promoter polymorphism and cancer risk: evidence from 47 published studies. Mutagenesis 2011; 26: 637–42. http://dx.doi.org/10.1093/mutage/ger025

[38] STEELE JC, MANN CH, ROOKES S, ROLLASON T, MURPHY D et al. T-cell responses to human papillomavirus type 16 among women with different grades of cervical neoplasia. British journal of cancer 2005; 93: 248–59. http://dx.doi.org/10.1038/sj.bjc.6602679

[39] STANCZUK GA, SIBANDA EN, PERREY C, CHIRARA M, PRAVICA V et al. Cancer of the uterine cervix may be significantly associated with a gene polymorphism coding for increased IL-10 production. International journal of cancer Journal international du cancer 2001; 94: 792–4. http://dx.doi.org/10.1002/ijc.1543

[40] SIMONART T, VAN VOOREN JP. Interleukin-1 beta increases the BCL-2/BAX ratio in Kaposi's sarcoma cells. Cytokine 2002; 19: 259–66. http://dx.doi.org/10.1006/cyto.2002.1964

[41] ASSCHERT JG, VELLENGA E, HOLLEMA H, VAN DER ZEE AG, DE VRIES EG. Expression of macrophage colony-stimulating factor (M-CSF), interleukin-6, (IL-6), interleukin-1 beta (IL-1 beta), interleukin-11 (IL-11) and tumour necrosis factor-alpha (TNF-alpha) in p53-characterised human ovarian carcinomas. European journal of cancer 1997; 33: 2246–51. http://dx.doi.org/10.1016/S0959-8049(97)00240-2

[42] HALL SK, PERREGAUX DG, GABEL CA, WOODWORTH T, DURHAM LK et al. Correlation of polymorphic variation in the promoter region of the interleukin-1 beta gene with secretion of interleukin-1 beta protein. Arthritis and rheumatism 2004; 50: 1976–83. http://dx.doi.org/10.1002/art.20310

[43] DRIPPS DJ, VERDERBER E, NG RK, THOMPSON RC, EISENBERG SP. Interleukin-1 receptor antagonist binds to the type II interleukin-1 receptor on B cells and neutrophils. The Journal of biological chemistry 1991; 266: 20311–5.

[44] VAMVAKOPOULOS J, GREEN C, METCALFE S. Genetic control of IL-1beta bioactivity through differential regulation of the IL-1 receptor antagonist. European journal of immunology 2002; 32: 2988–96. http://dx.doi.org/10.1002/1521-4141(2002010)32:10<2988::AID-IMMU2988>3.0.CO;2-9

[45] HURME M, SANTTILA S. IL-1 receptor antagonist (IL-1Ra) plasma levels are co-ordinately regulated by both IL-1Ra and IL-1beta genes. European journal of immunology 1998; 28: 2598–602. http://dx.doi.org/10.1002/(SICI)1521-4141(199808)28:08<2598::AID-IMMU2598>3.0.CO;2-K

[46] WITKIN SS, GERBER S, LEDGER WJ. Influence of interleukin-1 receptor antagonist gene polymorphism on disease. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America 2002; 34: 204–9. http://dx.doi.org/10.1086/338261