Relationship between the rs333 Polymorphism in the CC Chemokine Receptor Type Five (CCR5) Gene and Immunological Disorders: Data from a Meta-Analysis

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Abstract: *Introduction*: Inflammatory Bowel Disease (IBD), periodontitis and Systemic Lupus Erythematous (SLE) are multifactorial diseases, one of the factors in the course of these diseases is the rs333 polymorphism in the CC chemokine receptor type five (CCR5) *gene*. However, the results remain contradictory. Therefore, we aimed to perform a meta-analysis evaluating the relation between this polymorphism and the aforementioned conditions.

Material and Methods: A search in the literature was performed in diverse scientific and medical databases for studies published before June 22, 2020. The data were extracted from the studies and the statistical evaluation was performed by the calculations of statistical heterogeneity (I^2), Odds Ratio (OR) with 95% of Confidence Intervals (CI) and publication bias. The values of $P<0.05$ were considered as significant for all calculations.

Results: 19 articles with 21 case/control studies in 4,304 case patients and 3,492 controls were included. The meta-analysis showed a non-significant association among the rs333 polymorphism and IBD (OR = 1.05, 95% CI: 0.91-1.20, P = 0.51), periodontitis (OR = 0.86, 95% CI: 0.64-1.17, P = 0.34) or SLE (OR = 1.00, 95% CI: 0.56-1.80, P = 1.00) under the allelic model or for any other performed calculation. There were no obvious publication bias in the analyses.

Conclusion: In conclusion, this current meta-analysis evidenced the non-significant relation among the rs333 polymorphism and the risk of IBD, periodontitis or SLE. Further studies are required to validate our data.

Keywords: Cytokine, inflammation, autoimmune disease, genetic variation.

INTRODUCTION

Inflammatory Bowel Disease (IBD), periodontitis and Systemic Lupus Erythematous (SLE) are complex, multifactorial and relevant immunological disorders in the clinical field for the contribution of environmental, genetics and epigenetics factors that contribute to their pathogeneses [1-3].

IBD reached the burden of 5 million globally [4] with considerable prevalence in Netherlands [5] and the value of 4.3% per year between 2008 and 2018 in Scotland [6]. The main feature of IBD is the long-standing relapsing inflammation that affects various parts of gastrointestinal and it comprises two commonly diseases: the Cronh's Disease (CD) and Ulcerative Colitis (UC) [7]. Both clinical types share the same clinical course and outcome; however, it was observed that confined differences to the site of the lesions which CD affects the wall of the gut and UC is represented by lesions in the colon where it extends from the rectum to proximal parts of the large intestine [8].

Periodontal diseases are a group of inflammatory disorders that range the supporting structures around the teeth resulting in possible teeth loss [9]. The aetiology of periodontal diseases is well hallmarked by the dysbiotic states between the oral microbial populations and the host immune response [10]. Besides, periodontal diseases receive several classifications, out of which two forms are commonly found in the clinic, the chronic and the aggressive periodontitis forms (CP and AgP, respectively). The main feature of CP is the slow and swift progression of disease that reaches subjects with an increased mean

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of age [11] and AgP has the faster linear pattern of tissue destruction with early age of onset [12]. The disease has increased prevalence in the United States affecting around 42.2% of the population [13] and young populations ranging from 0.66% in Argentina to 5.9% in Israel [14].

On the other hand, SLE is an autoimmune immune disease with multifactorial aetiology. The exact pathogenesis of SLE is still unclear. However, the apoptosis/autophagy plus clearance defects are appointed as events that result in increased exposure of nuclear self-antigens, with the activation of type 1 interferon pathway and altered immune cells signalling and autoimmune response [3]. The prevalence of the disease increased in Korea in 2015 (from 21.25/100,000 people to 35.45/100,000 people), by the contrary the incidence has been decreased in this population [15] what these findings are according to data of the prevalence rate from United States between 2009 and 2016 [16].

These diseases are complex and multifactorial, which one of the influencer genetic factor in their pathogeneses is the rs333 polymorphism in the CC chemokine receptor type five (CCR5). CCR5 is responsible for the events of migration immune cells and the mediation of inflammatory responses [17]. The CCR5 *gene* is located at the short arm position 21 on chromosome 3, denoted as 3p21 and has been described by Report *et al*. [18].

Diverse types of cells express this G-protein coupled chemokine receptor such as dendritic cells, macrophages as well as memory T-cells. Besides, epithelial cells, endothelial cells; fibroblasts, vascular smooth muscle and microglia, astrocytes and neurons also express CCR5 in their surface [19]. In addition, CCR5 is a co-receptor recognized by the Human Immunodeficiency Virus-1 (HIV-1) for entry into the cells.

The rs333 polymorphism in the CCR5 *gene*, also known as CCR5Δ32 32-base-pair deletion, is able to create a disruption in the CCR5 protein consequently be important to prevent the HIV-1 infection [20]. This genetic variant also was considered as protective factor against the Hepatitis B Virus (HBV)/HIV co-infection [21] and has no influence on the risk of Hepatitis C Virus (HCV) and HCV/HIV co-infection [22].

The rs333 polymorphism has been related to the pathogenesis of several conditions beyond viral diseases such as celiac disease [23] and breast cancer [24]. Regarding the results on the rs333 polymorphism in the CCR5 *gene* and IBD, periodontitis and SLE, the data showed contradictories, which a better evaluation is required.

Therefore, when we considered the heterogeneous prevalence profile of the rs333 polymorphism among the populations [25] and the contrary results on the relation among this genetic variant and the risk of IBD, periodontitis and SLE, we attempted to perform a meta-analysis to gather these contrary results and bring a new evaluation on these data.

MATERIAL AND METHODS

In order to delineate and performing of this study, the authors have followed the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines [26].

This meta-analysis is registered in PROSPERO database with the following assigned number: CRD42020170583.

Eligibility Criteria

The following inclusion criteria were respected to the collection process in the literature search: (1) case-control studies or replication genetic studies; (2) the case-patients have been diagnosed by a rigorous clinical evaluation for [I] Inflammatory Bowel Disease [27], Periodontitis (and its diverse clinical manifestations) [28] or Systemic Lupus Erythematosus [29]; (3) the control patients have presented healthy periodontal evaluation or non-systemic sign of auto-immune disease; (4) the genotypic frequency have been completely documented; (5) the studies have strictly included human beings; (6) the included participants (case and control patients) have not presented cardiovascular diseases or pregnancy.

Strategy Search

The strategy search in the literature has been performed by two authors independently in the Google Scholar, MedLine, Pubmed, WAFANG and Web of Science. The literature retrieves in these medical and scientific databases occurred for studies published before June 22, 2020. The following combination of keywords was used in the search: “periodontitis or periodontal disease or chronic periodontitis or aggressive periodontitis or inflammatory bowel disease or ulcerative colitis or Crohn’s disease or systemic

lupus erythematosus" and "polymorphism or genetic variation" and "CCR5" and "rs333 or Δ32 deletion". There was no language restriction in the strategy search. In addition, there was a screening on the references from the included articles to detect any potential additional study.

Data Collection Process

Three investigators collected the data following a standardized formulary that composed the table of characteristics of included studies presented in the results. The data were extracted by author, year of publication, ethnicity/country of the included participants, study design, clinical diagnosis of the diseases, subject type evaluated in the included studies, sample size, mean of age of the participants, respect to the Hardy-Weinberg Equilibrium (HWE), method of genotyping and the data for risk of bias assessment that will be described at soon.

Risk of Bias Assessment

To verify the methodological quality the New-Castle Ottawa Scale (NOS) was used for evaluation of included genetic studies on the aforementioned polymorphism and the relation with these diseases, which the study that received less than 7 points has been excluded.

Statistical Analysis

For the calculations applied in the meta-analysis two statistical software were used: the Review Manager Software version 5.3 (RevMan, Nordic Cochrane Centre, The Cochrane Collaboration, 2012) for Odds Ratio (OR) and heterogeneity (I^2) calculations. The publication bias has been evaluated by the use of the Comprehensive Meta-analysis statistical software version 3.3.070 (2014).

The detection of true I^2 was measured by the Cochran's X^2 test or the chi-squared Q-based statistical test. The I^2 also was analyzed by visualization of the Funnel plot graphic to verify the heterogeneity. When the observed value of I^2 showed non-significant and was defined as mild or moderate (I^2<50%, P>0.05) the authors have performed the pooled OR calculation by the Fixed-effect statistical model [30]. When I^2 presented a statistically significant value and was defined as elevated (I^2>50%, P<0.05) the Random-effects statistical model [31] was used for obtaining the OR calculations. The P value <0.05 was considered as significant in all calculations. Based on mutant allele as Δ32 and the wild type allele as w, six genetic models were calculated. This calculation aimed to highlight the exact influence of the rs333 polymorphism in the CCR5 *gene* and the risk of the diseases. Then, the allele model (Δ32 *versus* w and w *versus* Δ32), the dominant homozygous model (Δ32Δ32 *versus* ww), the recessive homozygous model (ww *versus* Δ32Δ32) and the codominant models (Δ32Δ32 *versus* ww+wΔ32 and wΔ32 *versus* ww+Δ32Δ32) were evaluated. In addition, a sensitive analysis was performed omitting one study at a time in the pooled OR calculation for the mutant allele to detect any single interference. The authors have performed a sensitive analysis by omitting one single included study at the time to verify any possible significant change in the OR value. To detect the publication bias, the Begg's test [32] and Egger's linear regression test [33] were used as a statistical test to identify the potential publication bias (with P<0.05). In addition, asymmetry in the Funnel plot for publication bias was also considered to validate the results on Begg's test and Egger's test. The data from the included studies were dichotomous and have been expressed as OR with 95% of confidence intervals (CI) to verify the association among the rs333 polymorphism and IBD, periodontitis and SLE.

RESULTS

Characteristics of the Included Studies

At the final of the systematic search, 19 articles with 21 case/control studies have been included after the steps of identification, screening and eligibility as showed in Figure **1**. The studies were performed in IBD (n = 9 studies), periodontitis (n = 5 studies) and SLE (n = 7 studies) and have been published between 2001 and 2017 in diverse ethnical groups from different countries. In the studies, IBD was classified as UC CD sub-types. On the other hand, periodontitis was classified as Chronic Periodontitis (CP), Aggressive Periodontitis (AgP) or Generalized Aggressive Periodontitis (GAgP). All included studies have respected the Hardy-Weinberg Equilibrium. Additional and relevant details are available in Table **1**.

Meta-Analysis

rs333 Polymorphism and Inflammatory Diseases

The data available in the studies show a decreased Δ32 allele frequency, which several studies did not find double-positive participants for this allele in case or control groups (Table **1**). Overall, based on 19 papers composed by 21 studies [34-52] in 4,304 case patients

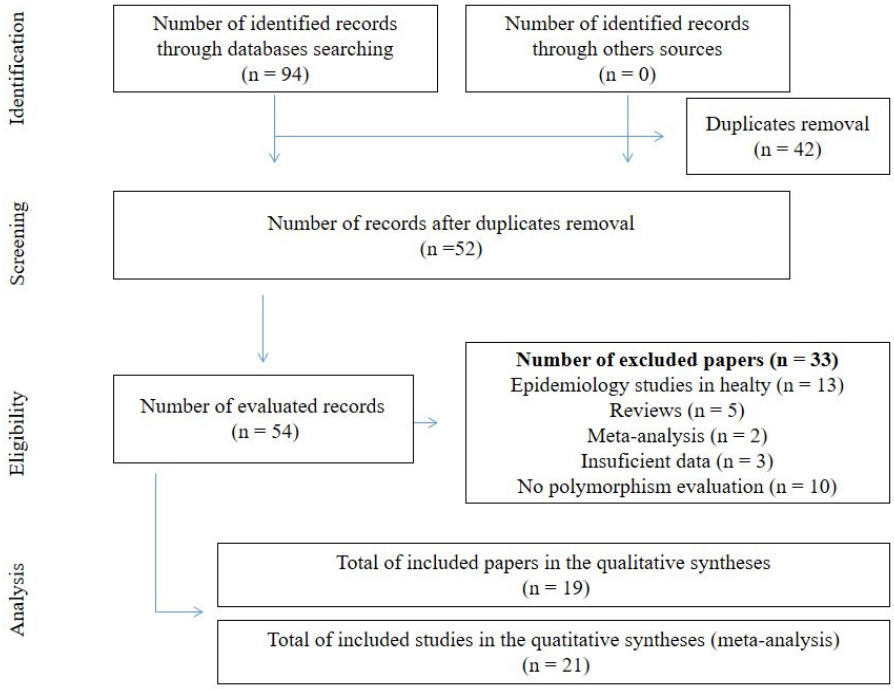

Figure 1:

and 3,492 controls, the rs333 polymorphism was not associated to any inflammatory condition approached in this systematic review and meta-analysis (Table **2** and Figure **2**).

Rs333 Polymorphism and Inflammatory Bowel Disease

Nine case/control studies with 2,622 patients with a diagnosis of IBD and 2,085 controls described the relationship between the rs333 polymorphism and IBD risk [34-42]. As showed in Figure **2** the statistical evaluation showed that the Δ32 allele was not significantly associated with IBD risk when compared with the wild type allele (OR = 1.05, 95% CI: 0.91-1.20, P = 0.51) with non-interference of obvious heterogeneity (I^2 = 0%, P*heterogeneity* = 0.71) (Figure **2A**). Non-significant associations have been observed in any calculations obtained in this current meta-analysis.

rs333 Polymorphism and Periodontitis

Four papers composed of five studies aimed to determine the association between the rs333 polymorphism in CCR5 *gene* and periodontitis [43-46]. Interesting to note is that the meta-analysis showed the Δ32 allele as a protecting factor for periodontitis risk which the variant allele was associated with the control group but with no statistical significance (OR = 0.86, 95% CI: 0.64-1.17, P = 0.34) (Figure **2B**). On the other hand, the wild type allele was not significantly associated with case-patients (OR = 1.16, 95% CI: 0.86-1.56, P = 0.34). The additional calculated genetic models are showed in Table **3**, in which there were non-significant associations; all these calculations were obtained by the fixed-effect model.

rs333 Polymorphism and SLE

To verify the association between the rs333 polymorphism and SLE risk, six papers with seven studies are included in the quantitative analyses [47-52]. The polymorphism was not significantly associated neither for the case-patients nor for the control group (Δ32 *versus* w – OR = 1.00, 95% CI: 0.56-1.80, P = 1.00, for both calculations) (Figure **2C**). The same results for the variant allele and the wild type are curious and should be noted, especially for the increased heterogeneity value (I^2 = 76%, P<0.0003) what led us to use the random-effects statistical model for these calculations. Other important calculations are showed in Table **4**.

Sensitive Analysis and Publication Bias

The sensitive analysis has indicated that no single study changed the pooled OR value what led us to take these data as accurate. Likewise, the results on the Begg's test and Egger's linear regression test did not find any evidence of possible publication bias (Table **5**), which is attested by the absence of obvious asymmetry in the funnel plots (Figure **3**), with consequent validation of our results.

Table 1: Characteristics of the Included Studies in this Current Meta-Analysis

Disease	Author	Year	Country/ Ethnical group	Sample size (Cc/Co)	Clinical sub-type	Clinical informations			Genotypic frequencies (CC/Co)			HWE (P_{value}Cc/Co)	NOS
						Mean of Age (years±SD)	Mean disease duration (years±SD)	Smoking status (%)	WW	WΔ32	Δ32Δ32		
Inflammatory Bowel Disease	Craggs	2001	England/ Caucasian	350/103	UC-CD	-	-	-	250/80	58/23	4/0	NI	7
	Eri	2004	England/ Caucasian	364/419	UC-CD	UC = 46±1.2 CD = 41±1.1	UC = 14±0.7 CD = 13±0.6	UC = 2 CD = 33	227/338	66/79	0/2	YES (P>0.05/P=0.93)	7
	Henckaerts	2006	Belgium/ Caucasian	400/362	UC-CD-IC	UC* = 29.11 (8-70) CD* = 24.50 (7-62)	UC = 14.6±6 CD = 17.5±8	NI	304/227	91/82	5/3	YES (NI)	7
	Herfarth	2001	Germany/ Caucasian	235/346	CD	NI	NI	NI	191/287	42/54	2/5	YES (NI)	8
	Hosek	2008	Czech Republic and Slovakia/ Caucasian	67/59	UC-CD	NI	NI	NI	49/41	17/18	1/0	YES (NI)	7
	Martin	2001	Germany/ Caucasian	200/120	UC-CD	NI	NI	NI	77/94	24/25	0/1	NI	8
	Paavola	2001	Finland/ Caucasian	435/172	UC-CD	NI	NI	NI	319/129	105/39	11/4	YES (P>0.10)	8
	Rector	2001	Belgium/ Caucasian	538/310	UC-CD	29* (8-70)	NI	NI	334/43	92/60	5/7	YES (NI)	9
	Satsangi	2009	England/ Caucasian	33/194	UC	NI	NI	NI	58/155	30/36	0/3	NI	7
						PD (mean±SD)	CAL (mean±SD)	PI (mean±SD)					
Periodontitis	Cavalla	2017	Brazil/ Mixed	288/197	CP-AgP	CP = 4.29±0.75 AgP = 3.75±1.38	CP = 3.92±0.64 AgP = 3.88±1.51	CP = 51.26±9.78 AgP = 46.41±8.51	256/186	31/31	1/1	YES (P>0.05)	8
	Fowaczny	2003	Germany/ Caucasian	81/121	-	54.0±12.4	-	-	67/96	12/24	2/1	NI	8
	Savarrio	2007	Scotland/ Caucasian	106/69	CP	-	-	-	53/35	23/15	2/0	YES (P>0.1)	7
	Shih	2014	China/ Asian	152/61	CP-GAgP	-	-	-	144/45	37/16	1/0	NI	7
						Mean of Age (years±SD)	Renal Nephritis (n)	Polyarthritis (n)					
Systemic Lupus Erythematosus	Aguillar	2003	Spain/ Caucasian	276/194	-	41.6 (14-91)	99	238	244/178	31/16	1/0	YES (NI)	7
	Baltus	2015	Brazil/ Mixed	169/132	-	40.0 (30.0-51.0)	-	-	199/194	18/34	2/2	YES (P>0.05)	7
	Carvalho	2014	European	219/230	-	-	-	-	199/194	18/34	2/2	YES (NI)	7
	Martens	2010	Netherlands/ Caucasian	97/431	-	44.0 (23-78)	38	61	78/333	17/86	2/12	YES (NI)	7
	Schauren	2013	Brazil/ Mixed	367/435	-	45.9±14.4	161	303	346/396	20/37	1/0	YES (NI)	7
	Yang	2003	China/ Asian	146/159	-	-	-	-	146/158	0/1	0/0	YES (NI)	7

Legend: **CC** – Case patients; **Co** – Control patients; **HWE** – Hardy Weinberg Equilibrium; **NOS** – Newcastle-Ottawa Scale; **SD** – Standardized Deviation; **UC** – Ulcerative colitis; **CD** – Cronh's disease; **NI** – Not informed; **CP** – Chronic Periodontitis; **AgP** – Aggressive Periodontitis; **PD** – Probing depth; **CAL** – Clinical Attachment Loss; **PI** – Plaque Index; **n** – Number of patients.

Table 2: Meta-Analysis of Comparison to rs333 Polymorphism in CCR5 Gene and Inflammatory Bowel Disease Risk (Allelic and Genotypic Comparisons)

	Comparison (n)	OR (95% CI)	Z *value* test (P *value*)	I² (%)	Pheterogeneity	Statistical model used
	IBD (n = 9)					
Allelic models	Δ32 versus w	1.05 (0.91-1.20)	0.66 (0.51)	0	0.71	F
	w versus Δ32	0.96 (0.84-1.09)	0.66 (0.51)	0	0.71	F
Homozygous dominant model	Δ32Δ32 versus ww	0.81 (0.47-1.42)	0.73 (0.47)	0	0.91	F
Recessive homozygous model	ww versus Δ32Δ32	0.93 (0.81-1.08)	0.93 (0.35)	7	0.38	F
Codominant models	Δ32Δ32 versus ww+Δ32w	0.80 (0.46-1.39)	0.79 (0.43)	0	0.87	F
	Δ32w versus ww+ Δ32Δ32	1.09 (0.94-1.27)	1.16 (0.25)	31	0.17	F

IBD – Inflammatory Bowel Disease, OR – Odds Ratio, CI – Confidence Intervals, I² – Heterogeneity, Δ32 – mutant allele, m – wild type allele, F – Fixed-effect statistical model.

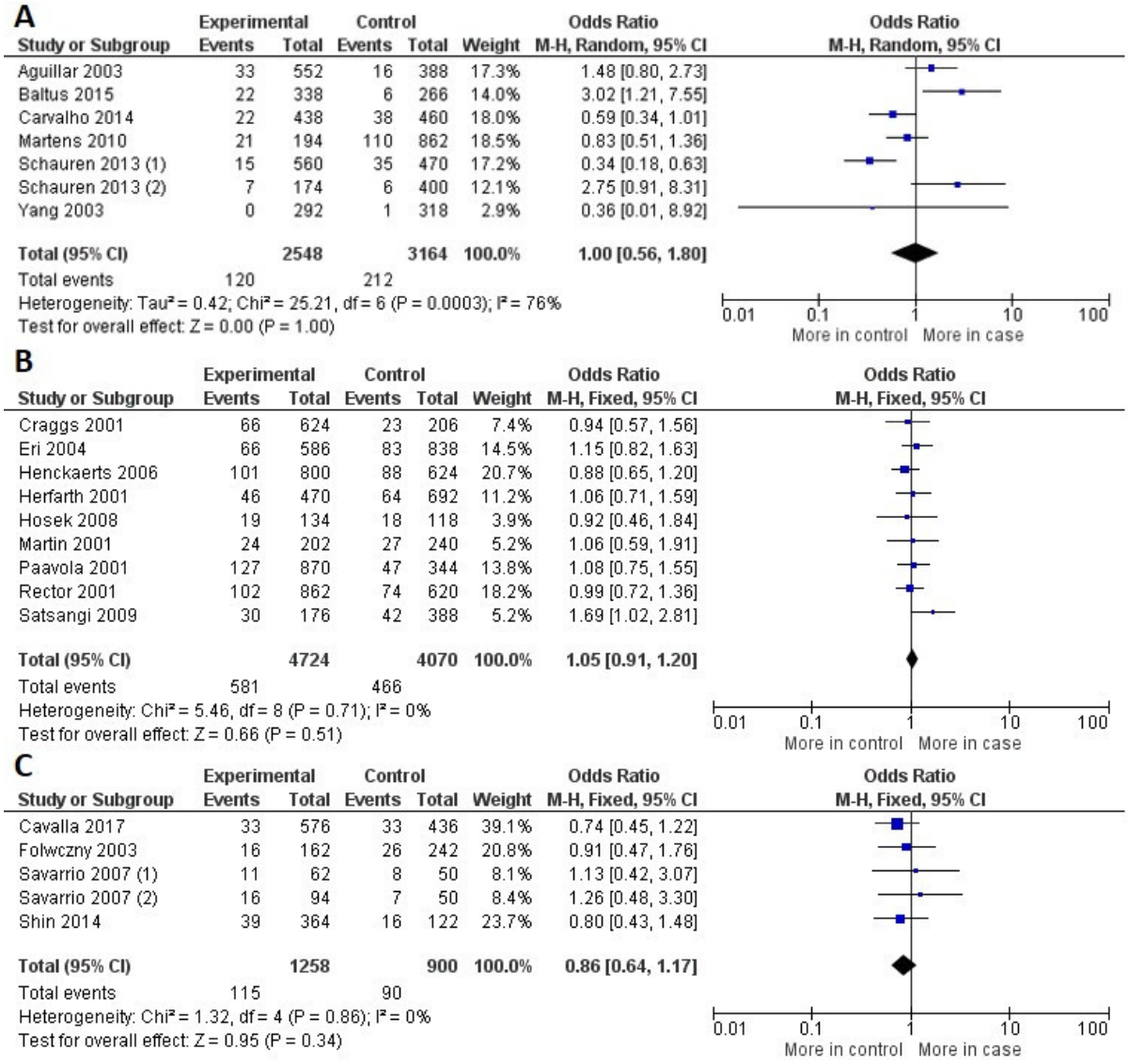

Figure 2:

Table 3: Meta-Analysis of Comparison to rs333 Polymorphism in CCR5 Gene and Periodontitis risk (Allelic and Genotypic Comparisons)

	Comparison (n)	OR (95% CI)	Z *value* test (P *value*)	I² (%)	Pheterogeneity	Statistical model used
	Periodontitis (n = 5)					
Allelic models	Δ32 versus w	0.86 (0.64-1.17)	0.95 (0.34)	0	0.86	F
	w versus Δ32	1.16 (0.86-1.56)	0.95 (0.34)	0	0.86	F
Homozygous dominant model	Δ32Δ32 versus ww	1.67 (0.42-6.64)	0.73 (0.47)	0	0.86	F
Recessive homozygous model	ww versus Δ32Δ32	0.60 (0.15-2.39)	0.73 (0.47)	0	0.86	F
Codominant models	Δ32Δ32 versus ww+Δ32w	1.74 (0.44-6.93)	0.79 (0.43)	0	0.86	F
	Δ32w versus ww+ Δ32Δ32	0.77 (0.55-1.07)	1.59 (0.11)	0	0.95	F

OR – Odds Ratio, CI – Confidence Intervals, I² – Heterogeneity, Δ32 – mutant allele, m – wild type allele, F – Fixed-effect statistical model.

Table 4: Meta-Analysis of Comparison to rs333 Polymorphism in CCR5 Gene Systemic Lupus Erythematous Risk (Allelic and Genotypic Comparisons)

	Comparison (n)	OR (95% CI)	Z *value* test (P *value*)	I² (%)	Pheterogeneity	Statistical model used
	SLE (n = 7)					
Allelic models	Δ32 versus w	1.00 (0.56-1.80)	0.00 (1.00)	76%	0.0003	R
	w versus Δ32	1.00 (0.56-1.80)	0.00 (1.00)	76%	0.0003	R
Homozygous dominant model	Δ32Δ32 versus ww	0.96 (0.41-2.28)	0.08 (0.93)	0	0.60	F
Recessive homozygous model	ww versus Δ32Δ32	1.04 (0.44-2.45)	0.08 (0.93)	0	0.60	F
Codominant models	Δ32Δ32 versus ww+Δ32w	0.99 (0.42-2.34)	0.02 (0.98)	0	0.64	F
	Δ32w versus ww+ Δ32Δ32	0.93 (0.53-1.63)	0.25 (0.81)	70	0.003	R

SLE – Systemic Lupus Erythematous, OR – Odds Ratio, CI – Confidence Intervals, I² – Heterogeneity, Δ32 – mutant allele, m – wild type allele, R – Random-effects statistical model, F – Fixed-effect statistical model.

Table 5: P-Value for the Begg's Test and Egger's Linear Regression Test for the Allelic Dominant Model Evaluation in this Current Meta-Analysis

Disease	Comparison	Begg's test	Egger's linear regression test
IBD	Δ32 *versus* w	0.754	0.479
Periodontitis		0.082	0.074
SLE		0.763	0.503

IBD – Inflammatory Bowel Disease, SLE – Systemic Lupus Erythematous, Δ32 – Mutant allele, w – Wild-type allele.

DISCUSSION

This current meta-analysis is the first to attempt to verify the relationship between the rs333 polymorphism in CCR5 *gene* and the risk of these inflammatory conditions. Despite the statistical results showed non-significant associations in any allelic or genotypic evaluations our results are accurate by the decreased heterogeneity value in most calculations, the absence of publication bias and the evaluation of adjustment factors for the evaluated inflammatory illness.

The CCR5 is a relevant receptor described in the literature, as the driver of cells for inflammatory sites

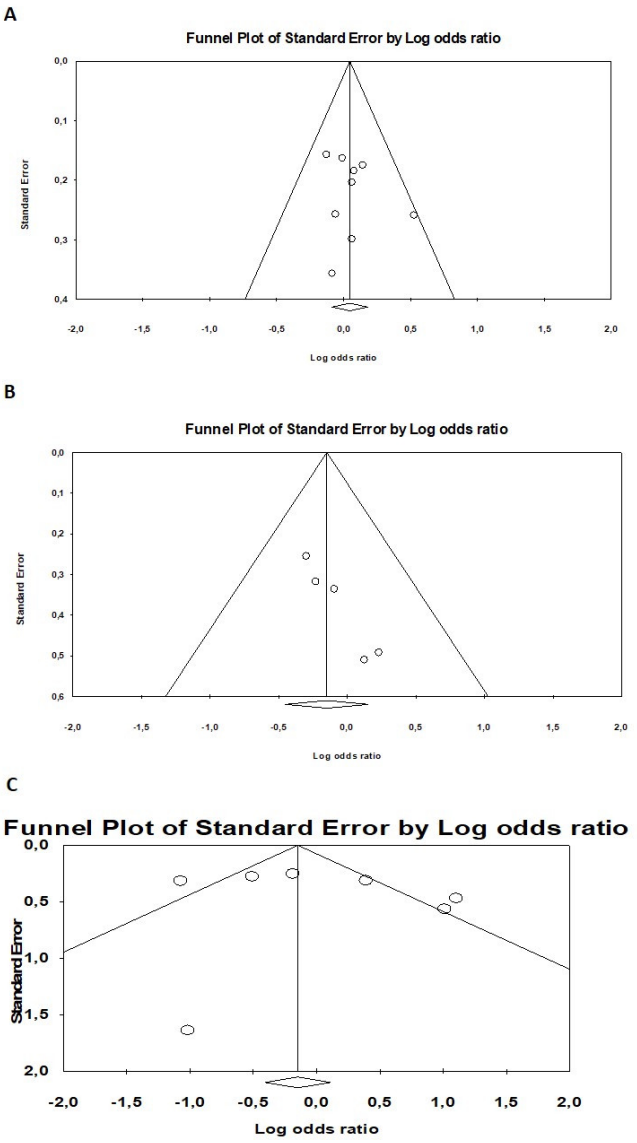

Figure 3:

[53] and, in assembly with other chemokine receptors, be able to enhance the T-cells functions [54]. Besides, CCR5 plays an important role in macrophage action, either by the increased responsiveness of monocyte-derived macrophages [55] or by the induction of macrophage survival during viral infection by anti-apoptotic signals [56]. It is well established that macrophages are critical cells for the initiation of the inflammatory process in IBD [57], periodontitis [58] and SLE [59].

In genetic levels, the rs333 polymorphism is another factor that influences the role of CCR5 in inflammatory conditions. Previous authors suggested that this gene variation is generated by the occurrence of stem-loop structure in DNA [60] which the Δ32 deletion leads to a synthesis of a truncated protein that fails to be inserted into the cell membrane, becomes nonfunctional [61]. This genetic variation is exhaustively associated with HIV-1 resistance by the decreased or absent expression of this co-receptor for virus-cell infection [62-64] and also with others virus type infections that were approached in previous studies [65,66].

On inflammatory conditions risk, the rs333 polymorphism was related to the risk of rheumatoid arthritis [67], multiple sclerosis [68] and psoriasis [69]. The data on IBD risk, studies bring contrary results, which some authors did not find the correlation between the delta32 allele with UC or CD [34] and other authors identified a significant association of this polymorphism with control groups with a possible protective effect [36]. The results from our meta-analysis are in disagreement with this previous finding and showed the non-significant association between the variant allele with the IBD risk ($P>0.05$). One explanation for this finding is the pooled data from the meta-analytic calculations that increase the number of case/control elements and increases the power of association.

The non-significant between the polymorphism and IBD risk also was found for periodontitis and SLE (Table **2**). It is relevant to denote two contradictory points in these results. First, the number of included studies for the diseases (n = 5 and n = 7, respectively) might promote a bias in the interpretation of results due to limited statistical power of noteworthiness association or biased value of I^2 [70].

On the other hand, periodontitis and SLE showed as two-way relationship diseases [71]. This two-way relationship may be explained by the constant interaction between the oral human microbiome and the host, which an imbalance leads to bone resorption with a periodontal dysbiosis that goes beyond the oral compartment influencing the SLE pathogenesis [72]. Although there was a non-significant association between the rs1333 polymorphism in the CCR5 *gene* for these two conditions, the results may be lead with caution.

Although to the best of our knowledge, this is the first meta-analysis that aimed to determine the association among the rs1333 polymorphism in CCR5 *gene* and IBD, periodontitis and SLE, which results in bringing the relevant data and the meta-analysis showed important limitations that should be appointed.

First, the decreased number of selected studies did not show robustness in the results what may be

considered as a source of bias. To avoid the potential bias of this limitation we have used accurate and robust statistical methods to validate our results. Second, important features from the evaluated patients were not available in the included articles. A complete evaluation of adjusting factors such as familiar history, gender, smokers and non-smokers, stratified age data and others conditions that influence the development of the diseases was not possible due to the limited information in the papers. Third, these three inflammatory diseases are clinical conditions that receive several classifications. An evaluation based on these clinical variations could not be performed due to limited data available in the studies. Fourth, a large number of the meta-analysis calculations were influenced by a significant I^2 and by the use of Random-effects as a statistical model. The value of I^2 indicates how the studies are inconsistent in a statistical manner. It may be a relevant factor in the meta-analysis because the true heterogeneity may affect directly the statistical model used on the results. The Random-effects statistical model provides an increased weight to studies containing a smaller sample size, which is not considered as totally trustworthy. Fifth, diverse non-significant associations were found in the meta-analysis calculations. However, the non-significant P-value does not always reflect the absence of clinical relevance. Sixth, we have evaluated one single polymorphism, other genetic variations may influence the rs1333 role in these inflammatory conditions. Previous studies available in the literature already showed the considerable impact of genetic polymorphisms and IBD [73], periodontitis [74-79] and SLE [80,81] further studies are required to validate our results.

CONCLUSION

In conclusion, this current meta-analysis composed of 21 case-control studies showed the non-significant association among the rs1333 polymorphism in CCR5 *gene* and IBD, periodontitis and SLE.

FUNDING

None.

LIST OF ABBREVIATIONS

rs333 = Reference Sequence polymorphism in the CCR5 *gene*

CCR5 = CC chemokine receptor type five

IBD = Inflammatory Bowel Disease

SLE = Systemic Lupus Erythematous

I^2 = Heterogeneity

OR = Odds Ratio

CI = Confidence Intervals

CP = Chronic Periodontitis

AgP = Aggressive Periodontitis

CD = Cronh's Disease

UC = Ulcerative Colitis

HIV-1 = Human Immunodeficiency Virus-1

HBC = Hepatitis B Virus

HCV = Hepatitis C Virus

PRISMA = Preferred Reporting Items for Systematic Reviews and Meta-Analyses

HWE = Hardy-Weinberg Equilibrium

NOS = New-Castle Ottawa Scale

GAgP = Generalized Aggressive Periodontitis

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