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Alcohol consumption is associated with decreased risk of rheumatoid arthritis; Results from two Scandinavian case-control studies

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Abstract

Objectives—The aim of the present study is to determine the association between risk of rheumatoid arthritis (RA) and alcohol consumption in combination with smoking and HLA-DRB1 shared epitope (SE).

Methods—Data from two independent case-control studies of RA, the Swedish EIRA (1204 cases and 871 controls) and the Danish CACORA- (444 cases and 533 controls) were used to estimate odds ratios (OR) of developing RA for different amounts of alcohol consumed.

Results—Alcohol consumption was, more common in controls ($p<0.05$) and dose-dependently associated with reduced risk of RA (p-trend<0.001) in both studies. Among alcohol consumers, the quarter with highest consumption had a decreased risk of RA in the order of 40-50% compared with the half with the lowest consumption EIRA: (OR=0.5 (95% confidence interval (CI) 0.4-0.6) and CACORA: OR=0.6 (95% CI 0.4-0.9)). For the subset of RA that is seropositive for antibodies to citrullinated peptide antigens, alcohol consumption was observed to reduce the risk the most in smokers carrying HLA-DRB1 SE alleles.

Conclusions—The observed inverse association between alcohol intake and risk of RA and the recent demonstration of a preventive effect of alcohol in experimental arthritis, indicates that alcohol may protect against RA. This highlights the potential role of life-style in determining the risk to develop RA, and emphasises the advice to stop smoking, but not necessarily to abstain from alcohol

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in order to diminish risk of RA. More generally, the evidence of potential RA prevention, urges for additional studies on how this can be achieved.

Keywords

Alcohol; Rheumatoid Arthritis; Smoking; HLA-DRB1; Case-control studies; Epidemiology

Introduction

Rheumatoid arthritis (RA) is a common, complex disease which seems to develop as a result of an interplay between inducing and protective environmental and genetic factors.[1-5] Recently, evidence for a possible interaction between lifestyle factors and genetic factors was provided from studies showing how smoking interacts with the shared epitope (SE) alleles of the HLA-DRB1 gene, in providing a very high risk to develop RA. Furthermore, the effect of both these risk factors was confined to one subset of RA, characterised by the presence of antibodies to citrullinated peptide antigens (ACPA).[2-4,6] These findings are of interest not only for public health, but also from a biological perspective, as they provide leads to a possible etiology of RA.[2] In a more general sense, they illustrate how investigations of life style factors can benefit from being done in the context of genetics, after appropriate subdivision of the disease into subsets.

Our interest in the role of alcohol consumption was triggered from several reports suggesting that alcohol influences inflammation in general and arthritis in particular; alcohol has thus been shown to diminish the response to immunogens in animals as well as in humans.[7-10] Alcohol can down-regulate production of pro-inflammatory molecules via influence on innate immunity.[11] Notably, addition of alcohol to the drinking water of mice was recently shown to reduce clinical signs of arthritis as well as joint destruction.[12] An indication that alcohol consumption may influence the risk also for human RA has come from four studies on environmental factors in RA development [5-6,13-15], whereas other studies did not found any alcohol-RA association.[16-17]. No studies have been published, to quantify the possible influence of alcohol consumption on the risk of RA and relate the influence of alcohol to genetic risk factors, and to the effects of smoking.

We combined information from two independent population based case-control studies on environmental and genetic risk factors for RA, to determine the influence of alcohol consumption on RA taking into account potential interactions between alcohol consumption, smoking and presence of HLA-DRB1 SE alleles.

METHODS

Two population-based case-control studies on environmental and genetic risk factors for RA were utilised: One was the Swedish EIRA (Epidemiological Investigation of Rheumatoid Arthritis) investigation. The part of the EIRA material that is used in the current study comprises 1419 incident RA cases, 18-70 years of age, recruited from 19 clinics in the south and middle of Sweden during the period between May 1996 and December 2003. All cases were diagnosed by rheumatologists according to the criteria given by the American College of Rheumatology (ACR) in 1987.[18] A total of 1674 controls were randomly selected from the general population with consideration taken to age, sex and residential area among the RA cases. More details on the study design and methods used are given elsewhere.[2,4,19] The second material was from the Danish CACORA (CAse-COntrol study on Rheumatoid Arthritis). This material comprises 515 prevalent RA cases fulfilling the ACR 1987 classification criteria for RA (mean disease duration 2.3 years, range 0-5 years) and 769 controls recruited between August 1998 and July 2003. Patients with RA were identified in

rheumatology and internal medicine departments throughout Denmark, and controls, frequency-matched by gender and birth year to the RA cases, were randomly selected from the Danish population by means of the Danish Civil Registration System, as detailed elsewhere.[5-6]

Ethical permit was obtained from relevant ethics committees and all the participants consented to contribute to the study.

Data collection and biological analysis

In EIRA, information on alcohol, smoking and other environmental exposures was obtained by means of an extensive self-administered questionnaire that was given to the cases shortly after they had been informed about their RA diagnosis, and mailed to the controls. The questionnaires were supposed to be answered at home and incomplete questionnaires were completed, by telephone or mail, by purpose-trained persons. Each case and control was asked to contribute with blood samples. The participation rate was 96 % for the cases and 82 % for the controls (for questionnaires) and 92 % and 63 % of participating cases and controls donated blood. Concomitant genetic and questionnaire information on alcohol and smoking was present for 1204 cases (879 female and 325 male) and 871 (645 female and 226 male) controls.

In CACORA, information on environmental exposures was obtained by means of a structured telephone interview. All participants were asked to give a blood sample for genotyping and serological analysis. The participation rate was 83 % for cases and 64 % for controls. Concomitant genetic and questionnaire information was present for 444 (312 female, 132 male) cases and 533 (327 female, 206 male) controls.

We genotyped participants that contributed with a blood sample for shared epitope (SE) alleles, defined as DRB1*01, DRB1*04 and DRB1*10 in the HLA-DRB1 gene, by using SSP-PCR (DR low resolution analysis).[20-21] Subjects with SE alleles were classified as having single or double SE alleles. For simplicity of evaluation, we assumed a dominant SE allele model. More details regarding the genotyping procedure is described elsewhere.[2,5-6,22]

Cases were sub grouped according to whether or not they had antibodies to citrullinated peptide antigens (ACPA) (ACPA-positive RA and ACPA-negative RA), as described elsewhere.[5, 23]

In EIRA, alcohol consumption was detailed by questions concerning present alcohol consumption during the last week as well as previous habitual consumption. In CACORA, the questions regarding alcohol consumption detailed the weekly consumption ten years before inclusion in the study. The questions in EIRA and CACORA allowed quantification of average alcohol consumption in drinks per week (1 drink= 16 gram alcohol). We categorized alcohol consumption in four different categories, based on the distribution of the sum of consumed alcoholic beverages per week among the controls in each study. The categories were non-drinkers (12.5 % of all in EIRA, 10.1 % of all in CACORA), low consumption (more than zero but below or equal to the median), moderate consumption (more than median but below or equal to the 75^{th} percentile), high consumption (above the 75^{th} percentile). In both studies, information on tobacco consumption was collected in ways allowing detailed quantification of smoking habits before disease onset.[5-6,19]

Statistical analysis

We calculated odds ratios (OR) for RA associated with each category of alcohol consumption together with 95 % confidence intervals (CI) by means of unconditional logistic regression models. We did separate analyses based on sex as well as on ACPA status among cases. Biologic interaction, defined by departure from additivity of effects as described by Rothman

and others, was evaluated between alcohol and smoking and between alcohol and HLA-DRB1 SE alleles. To quantify the amount of interaction, the attributable proportion due to interaction (AP) was calculated together with its p-value and 95% CI.[24-27] The attributable proportion due to interaction between two interacting factors reflects the joint effect beyond the sum of the independent effects. When analysing interaction between alcohol consumption and HLA-DRB1 SE, we categorized alcohol consumption into the three categories (0, 0.1-4.9, 5 or more alcoholic drinks per week).

In order to estimate if alcohol consumption was associated with smoking, we calculated Pearson's correlation coefficient between smoking and alcohol. Trend test for a dose response relationship regarding alcohol consumption and risk of RA was performed by using a continuous variable for units of alcohol consumed in a logistic regression model as suggested by Armitage.[28] Since the strength of the association between alcohol consumption and RA risk was seen to differ significantly between the Swedish and the Danish study, we display the results in parallel.

We used the SAS software for windows, version 9.1 (SAS Institute, Cary, NC) to analyse the data.

RESULTS

Alcohol consumption and risk of RA overall and of ACPA-positive RA and ACPA-negative RA

The sex and age distribution of cases and controls, and their alcohol consumption is shown in table 1 for both the EIRA and CACORA materials. Compared with controls in EIRA, a considerably higher consumption of alcohol was reported by the controls in CACORA. The overall relationship between number of drinks per day and RA risk, as estimated by the logistic regression model when alcohol was entered as a continuous parameter, was −0.068 (95% CI −0.093 to −0.056) in EIRA and −0.021 (95% CI −0.035 to −0.008) in CACORA, corresponding to an average decrease in the odds of RA of about 5% per drink per week in EIRA and about 2% per drink per week in CACORA (both p-values for trend<0.001).

Table 2 displays the relationship between different categories of alcohol consumption and risk of RA overall as well as for the ACPA-positive and ACPA-negative subsets of RA for each of the two studies. A statistically significant dose-dependent reduction in the odds of RA overall was seen in individuals with higher alcohol consumption levels in both materials. In EIRA, the larger of the studies, the effect was present in males as well as in females (data not shown) and for ACPA-positive as well as for ACPA-negative RA (table 2). In CACORA, the inverse association between alcohol consumption and RA risk was significant only for the ACPA-positive subset.

Alcohol consumption, smoking and risk of ACPA-positive RA

We were interested in whether alcohol consumption might interact with smoking behaviour in influencing RA risk. Since smoking is known to influence only ACPA-positive disease, this interaction analysis was only carried out for ACPA-positive RA.

A statistically significant interaction between smoking and no alcohol consumption (table 3), was observed regarding ACPA-positive RA in both studies. Thus the risk reduction associated with alcohol consumption was in both studies more pronounced among ever smokers than never smokers (table 3).

Alcohol consumption, HLA-DRB1 SE alleles and risk of ACPA-positive RA

As risk to develop ACPA-positive RA is highly dependent on whether an individual carries none, one or two copies of the HLA-DRB1 SE alleles, we further wanted to investigate whether alcohol consumption influenced risk of RA differently according to HLA-DRB1 genotype. In both studies, the absolute risk reduction associated with alcohol consumption was more pronounced among carriers of HLA-DRB1 SE alleles than among non-carriers (supplementary table 1 and figure 1). When formally tested, this gene-environment interaction between HLA-DRB1 SE and alcohol consumption regarding risk of ACPA-positive RA was found to be statistically significant in both EIRA ($p<0.0001$) and CACORA ($p<0.0001$) when non-drinkers were contrasted to high consumers of alcohol (at least 5 drinks/week).

Alcohol consumption, smoking, HLA-DRB1 SE alleles and risk of ACPA-positive RA

Figure 1 displays the more complex picture when alcohol, smoking and HLA-DRB1 SE alleles are considered simultaneously (supplementary table 2). The overall pattern is seen to be fairly similar for the two studies. As seen, the risk reduction associated with alcohol consumption seems to be most pronounced among smokers carrying one or two HLA-DRB1 SE alleles (EIRA p-(trend)<0.0001); (CACORA p-(trend)<0.01).

Discussion

The major finding in the present report is that alcohol consumption exhibits a dose-dependent inverse association with RA. Furthermore, alcohol consumption is associated with attenuation of the effect of the best established risk factors for RA, smoking and HLA-DRB1 SE with regard to ACPA-positive RA.

It is methodologically demanding to investigate the relationship between two life style factors (alcohol, smoking) and one genetic factor (HLA-DRB1 SE) in two subsets of RA, since these factors may be interrelated to varying degrees in RA patients, and as completeness of information and non-biased recruitment of cases and controls is crucial for the reliability of results. The present two studies both utilised unique features of the Scandinavian health care systems in capturing representative and population-based collections of patients and well matched controls. The alcohol consumption differed between the studies. In the Danish CACORA study, the average alcohol consumption was higher than in the Swedish EIRA study, a well-known country difference which is in accordance with WHO information on alcohol consumption in Denmark and Sweden.[29] The high response frequencies of cases and controls should minimise the risk for selection bias. As always in case-control studies with retrospective exposure information, recall bias is a concern. Most often recall bias relates to the tendency to over-report previous exposure among cases relative to the controls. In our study, the situation is the opposite, i.e. if our results should be explained by recall bias, cases should have systematically understated their previous alcohol consumption in relation to the statements among controls. In order to highlight if RA cases tend to change their alcohol consumption by time, we studied the association between disease duration and alcohol consumption in EIRA. Reported alcohol consumption in RA cases with disease duration less than 6 months did not differ from that of patients with longer disease duration. On the assumption that recall bias is influenced by disease duration, the lack of such association is reassuring. Another potential bias may stem from RA patients treated with methotrexate or non-steroidal anti inflammatory drugs (NSAID). These patients may have been advised to abstain from alcohol by their physician. However, in EIRA, we did not find any differences regarding reported alcohol consumption between cases taking methotrexate or NSAID and cases not taking these medications. In CACORA, questions were asked specifically for the alcohol consumption 10 years back in time which should minimise the risk of differential recall between cases and controls. Although our findings need replication we consider our ability to use two parallel but

completely independent studies performed in two countries with similar socioeconomic and cultural conditions to be a strength of this study. Recently, similar findings as ours with regard to alcohol consumption and risk of RA, was presented from a prospective cohort study conducted in south of Sweden. [15]

A further concern is that information on interacting exposures such as smoking must be accurate and detailed, and that replication is needed also in these respects. Also here, we are confident that the Swedish and Danish studies on the interaction between smoking and RA have been so consistent, and that these results have been confirmed also in a Dutch study.[2-3,6] Why these results were not fully replicated in a recent study from North America is not yet clear, but may depend on regional differences in the complex interplay between a variety of genetic and environmental factors besides smoking.[30] Also from this perspective, our combination of independent studies from culturally and genetically similar environments in Sweden and Denmark is advantageous.

Somewhat unexpectedly, we observed an interaction between lack of alcohol and smoking and between lack of alcohol and presence of HLA-DRB1 SE alleles, respectively, with regard to risk of ACPA positive RA. The method used to calculate this interaction is based on a calculation of deviation from additivity and the existence of such interactions is an indication of at least one pathway towards disease in which both risk factors are required.[24-26] Further research into mechanisms behind the interactions between smoking and alcohol may thus be of value in helping to understand molecular aspects of the RA pathology, similarly to what appears to be the case from our recent studies of interactions between smoking and the HLA-DRB1 alleles.[2]

Evidence that alcohol intake may protect against development of arthritis has recently been obtained also from studies on experimental arthritis, where administration of alcohol both reduced the incidence and severity of collagen induced arthritis in mice.[12] Further studies on the mechanisms of this protective effect indicated an effect of alcohol on the NFKB-dependent pro-inflammatory signalling pathways, but limited effects on the anti-collagen immune response, i.e. on adaptive immunity.[8,11-12] These data lend some support to the notion that alcohol intake may be causatively associated with a reduced risk for RA also in humans. The evidence from the larger of the two studies, the EIRA, that alcohol intake is inversely related to both ACPA+ and ACPA− RA is compatible with an effect on general pro-inflammatory mechanisms also in human disease. We do not, however, have any good biological hypothesis to explain neither the interaction between alcohol intake and HLA-DRB1 SE, nor the observed interaction between smoking and alcohol intake. Together this calls for extended studies on biological effects of alcohol and related compounds in relation to arthritis.

Seen in a wider perspective of inflammatory diseases, the now observed association of alcohol on the risk to develop RA shows similarities to what has for long been known for cardiovascular disease. Here, moderate alcohol consumption is dose-dependently associated with a decreased risk to develop cardiovascular disease.[31-32] Despite many years of research, no clear-cut mechanism has, however, been identified that can explain these relationships. Our current demonstration of a similar effect in a classic inflammatory disease, RA, may shed some light also on the relationship between alcohol consumption and other inflammatory diseases; these relationships should be subject to more combined studies in the future.

In summary, this study has several potential medical implications. From a public health perspective, we consider the findings of interest as they provide new information on how modifiable life style factor may influence the risk of developing RA. In this context, the main message remains that cessation of smoking is the most effective way to diminish risk for RA, irrespective of genetic constitution, but that this recommendation should not necessarily be

combined with a recommendation to stop moderate alcohol consumption. Equally interesting are the potential biological implications. The fact that data from both animal and human studies suggest that arthritis risk can be reduced by alcohol – or with other agents having similar effects – should encourage further studies on how such prevention can be achieved.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

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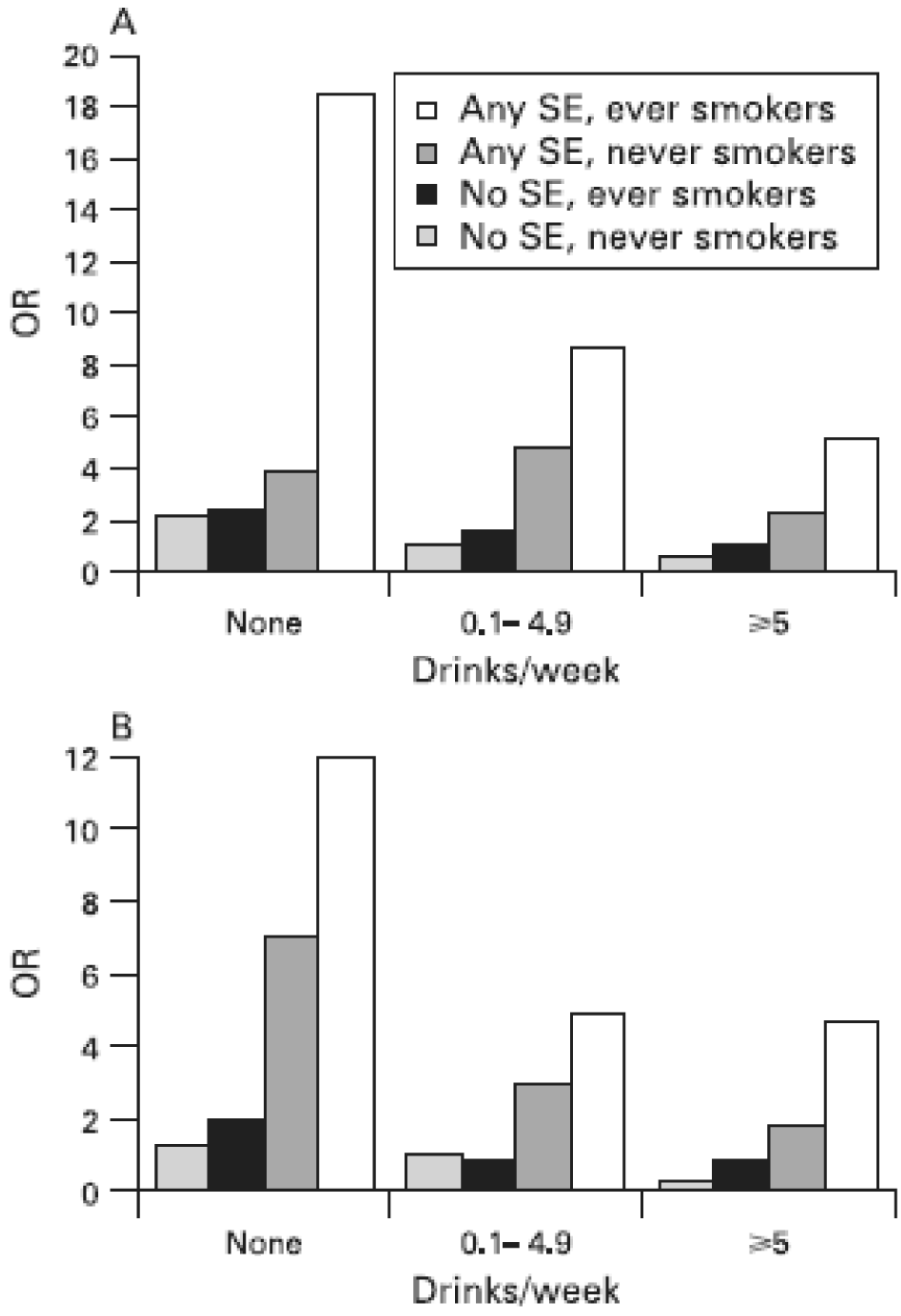

Figure 1.
Histograms showing OR of antibodies to citrullinated peptide antigen-positive rheumatoid arthritis for different combinations of alcohol consumption, smoking (never/ever) and presence of absence of HLA-DRB1 shared epitope (SE) alleles compared with never smokers with low alcohol consumption (0.1–4.9 drinks/week) and without SE alleles, by study (A, Epidemiological Investigation of Rheumatoid Arthritis (EIRA); B, Case–Control Study on Rheumatoid Arthritis (CACORA)). Further details are given in supplementary table 2.

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Table 1

Characteristics of cases and controls in the EIRA and CACORA studies

	EIRA			CACORA		
Characteristics	**Cases (n = 1204)**	**Controls (n = 871)**	**p-value**	**Cases (n = 444)**	**Controls (n = 533)**	**p-value**
Women (%)	73 (n=879)	74 (n=645)	0.59	70.3 (n=312)	61.4 (n=327)	0.004
Mean age (SD*)	51.1 (12.5)	52.1 (11.7)	0.07	49.1 (11.1)	49.8 (10.5)	0.31
RA disease duration	0.8 years†	……..		2.3 years‡	… … … …	
ACPA-positive (%)	61.1 (n=735)	1.9 (n=16)	<0.0001	69.4 (n=308)	-§	… …
Ever smokers (%)	66.1 (n=796)	60.3 (n=525)	0.006	69.8 (n=310)	60.6 (n=323)	0.003
HLA-DRB1 SE alleles						
Heterozygous carriers (%)	49.3 (n=593)	42.8 (n=373)	<0.0001	45.1 (n=200)	44.5 (n=237)	0.77
Homozygous carriers (%)	24.5 (n=295)	9.8 (n=85)	<0.0001	30.0 (n=133)	8.4 (n=45)	<0.0001
Alcohol consumption						
None-drinkers (%)	15.1 (n=182)	12.5 (n=109)	0.046	18.0 (n=80)	10.1 (n=54)	0.0002
Average alcohol (drinks) intake per week (SD*)	2.9 (4.2)	4.1 (5.6)	<0.0001	6.6 (8.9)	9.0 (8.7)	0.003
Median, units/week	1.9	2.9		4	5	
75th percentile, drinks/week	3.8	4.9		8.5	12	
Correlation	0.11	0.17		0.22	0.17	
Smoking and alcohol consumption	(p<0.0001)	(p<0.0001)		(p<0.0001)	(p=0.0005)	

*Standard deviation.

†Newly diagnosed cases; mean duration between onset of first symptoms and inclusion in the study.

‡Prevalent cases; mean interval between RA diagnosis and inclusion in the study.

§ ACPA not measured among the controls in CACORA.

Table 2

Odds ratio (OR) with 95% confidence interval (95% CI) of rheumatoid arthritis for subjects in different alcohol consumption categories, by ACPA status and study (EIRA and CACORA)

		EIRA			CACORA		
	Alcohol Consumption	**exp ca/co***	**OR†**	**95 % CI**	**exp ca/co***	**OR†**	**95 % CI**
RA overall							
	Non-drinkers	182/109	1.1	0.8 - 1.4	80/54	1.7	1.1 - 2.5
	Low	674/396	1.0 §	Ref.	186/215	1.0 §	Ref.
	Moderate	171/175	0.6	0.4 - 0.7	111/134	1.0	0.7 - 1.3
	High	177/190	0.5	0.4 - 0.6	67/130	0.6	0.4 - 0.9
Trend			p<0.0001			p=0.0003	
ACPA-positive RA							
	Non-drinkers	120/109	1.3	1.0 - 1.8	62/54	2.0	1.3 - 3.1
	Low	410/396	1.0 §	Ref.	125/215	1.0 §	Ref.
	Moderate	97/175	0.5	0.4 - 0.7	77/134	1.0	0.7 - 1.4
	High	108/190	0.5	0.3 - 0.6	44/130	0.6	0.4 - 0.9
Trend			p<0.0001			p<0.0001	
ACPA-negative RA							
	Non-drinkers	62/109	0.9	0.6 - 1.3	18/54	1.1	0.6 - 2.1
	Low	264/396	1.0 §	Ref.	61/215	1.0 §	Ref.
	Moderate	74/175	0.7	0.5 - 0.9	34/134	0.9	0.6 - 1.5
	High	69/190	0.5	0.4 - 0.7	23/130	0.9	0.5 - 1.5
Trend			p=0.0005			p=0.43	

*Number of exposed (exp) cases (ca) and controls (co),

†Odds ratio adjusted for sex, age, smoking (never/ever) and residential area (EIRA only),

§Reference category.

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Table 3

Odds ratio (OR) with 95% confidence interval (95% CI) of ACPA-positive rheumatoid arthritis for subjects exposed to different combinations of alcohol and smoking.

EIRA (ACPA-positive RA)						
Alcohol consumption	**Never smoking**			**Ever smoking**		
	exp ca/co*	**OR†**	**95% CI**	**exp ca/co***	**OR†**	**95% CI**
Drinkers	159/269	1.0‡	Ref.	456/492	1.7	1.3 - 2.1
Non-drinkers	47/76	1.1	0.7 - 1.7	73/33	4.2	2.6 - 6.6
p-interaction §				p< 0.0001		

CACORA (ACPA-positive RA)						
	Never smoking			**Ever smoking**		
	exp ca/co*	**OR†**	**95% CI**	**exp ca/co***	**OR†**	**95% CI**
Drinkers	63/181	1.0‡	Ref	183/298	1.9	1.3 - 2.7
Non-drinkers	20/29	1.9	1.0 - 3.6	42/25	4.8	2.7 - 8.5
p-interaction §				p< 0.0001		

*Number of exposed (exp) cases (ca) and controls (co),

†Odds ratio adjusted for sex, age and residential area (EIRA only).

‡Reference category.

§The attributable proportion due to interaction (AP) between smoking status and lack alcohol consumption (95 % confidence interval) were: for EIRA, AP = 0.6 (0.5 - 0.7) and for CACORA, AP = 0.4 (0.3 - 0.5)