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C-reactive protein-associated genetic variants and cancer risk: Findings from FINRISK 1992, FINRISK 1997 and Health 2000 studies

Katriina Heikkilä [a,*], *Kaisa Silander* [a], *Veikko Salomaa* [a], *Pekka Jousilahti* [a], *Seppo Koskinen* [a], *Eero Pukkala* [b], *Markus Perola* [a]

[a] *The National Institute for Health and Welfare, Helsinki, Finland*
[b] *Finnish Cancer Registry, Institute for Statistical and Epidemiological Cancer Research, Helsinki, Finland*

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ABSTRACT

Background: Evidence from prospective observational studies suggests that elevated circulating C-reactive protein (CRP) concentrations are associated with cancer risk, but it is unclear whether this association is causal. In order to examine this, we investigated whether genetic variants that are associated with circulating CRP concentrations are associated with cancer risk.

Methods: We pooled data from three population-based prospective Finnish studies: FINRISK 1992 ($n = 5289$), FINRISK 1997 ($n = 7160$) and Health 2000 ($n = 6299$). Cancer cases were identified from cancer registrations. Thirteen CRP-associated SNPs, identified from genome-wide association studies, were genotyped. We examined the associations of the SNPs and cancer risk using Cox, probit and instrumented probit regression models.

Results: Compared to common allele homozygotes, individuals carrying one or two variant T alleles at rs1892534 had 1.05-fold (95% confidence interval (CI): 0.90, 1.23) and 1.2-fold (95% CI: 1.01, 1.42) increased overall cancer risk, respectively. Individuals with one or two variant A alleles at rs1169300 or rs2464196 had approximately 1.5- and 2-fold increased risk of lung cancer, respectively (p trend for both: 0.007). CRP SNPs were not associated with colorectal, prostate or breast cancer risk nor was CRP-associated with the probability of developing cancer in the instrumented probit analyses.

Conclusions: We found some evidence for an association of a small number of CRP-associated SNPs with the overall cancer risk and lung cancer risk. Our instrumental variable analyses provided no clear evidence for a causal association of CRP and cancer. These findings suggest that circulating CRP concentrations are unlikely to have a causal role in cancer.

1. Introduction

Inflammation has an important role in cancer development. During an inflammatory response the interactions between immune system cells and molecules, carcinogens and pre-malignant and malignant cells drive on cancer initiation, promotion and progression. Some of these interactions are well known, others are unclear. Inflammatory cytokines, chemokines and growth and transcription factors promote cancer cell growth and proliferation and prevent the host

* *Corresponding author:* Address: The National Institute for Health and Welfare, P.O. Box 104, 00251 Helsinki, Finland. Tel.: +358 20 610 8727; fax: +358 (0) 20 610 8480.
E-mail address: katriina.heikkila@npeu.ox.ac.uk (K. Heikkilä).
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clearance of potentially malignant cells by inhibiting apoptosis.[1] Inflammatory processes also facilitate malignant cells' invasion of tissues by promoting cell motility, vascular permeability and angiogenesis.[1] Furthermore, oxidative damage, a by-product of inflammation, could initiate cancer development by causing inactivating mutations in tumour-suppressor genes or post-translational modifications in proteins involved in DNA repair or control of apoptosis.[1]

C-reactive protein (CRP) is an inflammatory effector and indicator protein. An individual's basal circulating CRP concentration is genetically determined[2,3] but many cancers[4] as well as biological and lifestyle factors, such as old age,[5] adiposity[6] and tobacco smoking[7,8], are associated with increased circulating CRP concentrations. Evidence from prospective observational epidemiology suggests that elevated circulating CRP concentrations are associated with an increased cancer risk.[9] However, cancers have a long latent period and even in studies in which CRP concentrations have been measured years or decades prior to cancer diagnosis, any observed associations of CRP with incident cancer may be due to reverse causality and a host response to the early neoplastic process. It is also possible that residual confounding from unmeasured biological or lifestyle factors has influenced the associations of CRP and cancer risk in observational studies.

One way to examine the relationship between CRP and cancer risk is to investigate whether genetic variants that determine or associate with circulating CRP concentrations are also associated with cancer risk, using such variants as instruments for circulating CRP.[10] As genetic variants are distributed independently of each other at population level, they are generally unrelated to potentially confounding socioeconomic or lifestyle factors and no disease process can influence an individual's genotype.[10] Hence, an association between a genetic variant that influences or is a marker of CRP concentration and any cancer is unlikely to be prone to reverse causality or residual confounding.

The aim of our study was to investigate whether single nucleotide polymorphisms (SNPs) associated with circulating CRP concentrations are themselves, or as instruments for circulating CRP, associated with cancer risk. We are aware of only one previous study in which the investigators used an instrumental variable approach to examine the associations of CRP and cancer risk in a Danish population.[11] We have, for the first time, investigated these associations in a Finnish population.

2. Participants and methods

2.1. Participants

In order to avoid bias from population stratification, we conducted our study in a Finnish population, which is genetically relatively homogeneous.[12] We used prospective data from three population-based studies, FINRISK 1992, FINRISK 1997 and Health 2000. Details of the sampling and recruitment of the participants in the studies have been described previously.[13–16] Briefly, stratified clustered samples of men and women aged 30+ (in Health 2000) and 25–74 (in FINRISK 1992 and 1997) were identified from the Finnish population register and posted an invitation to participate. In all studies the participants completed a self-report questionnaire and attended a medical examination at baseline and were followed-up with the Finnish Cancer Registry for cancer registrations. Health 2000 was approved by the Ethics Committee of Epidemiology and Public Health of the Hospital District of Helsinki and Uusimaa, and the FINRISK studies by the Ethics Committee of the National Public Health Institute, Finland.

Our eligible study population consisted of individuals whose blood samples provided sufficient DNA yield for standard Sequenom analyses (>5 ng/μL), who had genotype data for at least 50% of the SNPs and who had concordant baseline and genotyped gender data. Of the 8028 Health 2000 participants 6334 had sufficient DNA and their blood samples were genotyped. 35 participants with >50% genotyping failure were excluded. Of the 5999 participants in FINRISK 1992, 5496 had sufficient DNA yield and their blood samples were genotyped. 131 individuals with >50% genotyping failure and 76 individuals with discrepant baseline and genotyped gender data were excluded. Of the 8141 FINRISK 1997 participants, 7239 had sufficient DNA and their blood samples were genotyped. We excluded individuals with discrepant baseline and genotyped gender data ($n = 27$) and incomplete phenotype data ($n = 52$). After all the exclusions we were left with 6299 Health 2000 participants, 5289 FINRISK 1992 participants and 7160 FINRISK 1997 participants. All eligible participants had data on cancer outcomes, age and sex and these summary characteristics are presented in Table 1.

2.2. Cancer outcomes

Individuals were defined as cancer cases according to the type of their first cancer, either a cancer registration during their lifetime or cancer on their death certificate. The date of the cancer was defined as the date of diagnosis or death, as appropriate. All cancers registered in the study population since the start of cancer registration in Finland in 1953 were included in the analyses.[17] As non-melanoma skin cancer is usually a relatively benign disease, individuals with non-melanoma skin cancer as their only cancer ($n = 200$) were treated as cancer free. We did, however, conduct sensitivity analyses with these individuals defined as cancer cases. Cancer events were coded using ICD-10 (International Classification of Diseases) and the cancer outcomes categorised into any cancer and colorectal (C18–C20), lung (C34), prostate (C61) and breast (C50) cancers.

2.3. Genotyping and biochemical analyses

We genotyped 13 SNPs in which previous genome-wide association studies have shown to be associated with circulating CRP concentrations.[18,19] Genotyping and quantification of circulating CRP concentrations are described in Supplementary Web Appendix 1. Briefly, CRP genotypes and circulating CRP concentrations were determined from 8 to 10 mL whole blood samples drawn after a minimum of four hour's fast at the baseline medical examination. Genotyping was done using Sequenom MassARRAY System and iPLEX Gold chemistry (Sequenom, San Diego, California), according to the manufacturer's instructions. Genotype clusters were reviewed using

Table 1 – Summary of eligible participants.

Participant characteristics	FINRISK 1992 (N = 5289)	FINRISK 1997 (N = 7160)	Health 2000 (N = 6299)	Total (N = 18 748)
Age (mean, SD)	44 (11.3)	48.5 (13.4)	54.3 (15.4)	49.3 (14.1)
Women (N, %)	2866 (54.2)	3664 (51.2)	3461 (54.9)	9991 (53.3)
Cancer cases (N, %)	409 (7.7)	571 (10.0)	579 (9.1)	1559 (8.3)
Colorectal cancer	33 (0.6)	53 (0.7)	67 (1.1)	153 (0.8)
Lung cancer	39 (0.7)	53 (0.7)	30 (0.5)	122 (0.7)
Breast cancer	94 (1.8)	92 (1.3)	142 (2.2)	328 (1.7)
Prostate cancer	60 (1.1)	113 (1.6)	90 (1.4)	263 (1.4)

SD: standard deviation.

the Typer 4.0 software (Sequenom) and genotype calls corrected where necessary.

2.4. Statistical analyses

CRP concentrations were natural log-transformed. CRP concentrations lower than 0.1 mg/L (in FINRISK studies) and 0.2 mg/L (in Health 2000) could not be detected by the assays used in these studies and the individuals whose CRP concentrations were measured as 0.1 or 0.2 mg/L were treated as having these values.

We checked Hardy–Weinberg equilibrium using chi-squared tests. Genotyping success rates for each SNP in each study were calculated as the proportion of successfully genotyped samples out of all the samples for which genotyping was attempted. Linkage disequilibrium was assessed using the r^2 statistic.

In order to maximise statistical power, we conducted separate complete-case analyses for each SNP and covariates. Thus, each model was based on data from participants who had complete data for the relevant SNP (or, in the instrumental variable analyses, two SNPs), cancer outcomes and covariates.

We used linear regression to examine the associations of the genotypes with circulating CRP concentrations and Cox proportional hazards regression to investigate the associations of the genotypes and cancer risk. The time at risk was defined as time from birth to the date of cancer diagnosis, date of death or the end of follow-up, whichever occurred first. The end of follow-up was 31st December 2006 for the FINRISK participants and 31st December 2007 for the Health 2000 participants. The median follow-up time since birth was 58 years and the median follow-up since the study enrolment was 10 years. We tested the proportional hazards assumption using the Schoenfeld-test and observed no major departures from proportionality. Models assuming dominant and additive effect of a SNP on cancer risk were estimated. Tests for trend in the additive models were conducted by modelling the association of cancer risk with the number of minor alleles of each SNP as an ordinal exposure variable. We adjusted our models for age (by the timescale in the model) and sex (in the models of colorectal and lung cancers). As there is evidence for fine-scale genetic population variation within Finland,[20] we conducted sensitivity analyses with additional adjustment for geographical area of origin (East versus West of Finland).

We used instrumental variable analysis to investigate the causal associations of CRP with cancer. This approach is based on the assumptions that if the CRP genotype is associated with the circulating CRP concentration but not with potential lifestyle and environmental confounders, and if the genotype exerts an effect on cancer risk only through its effect on circulating CRP, then the CRP genotype provides an objective proxy measure for circulating CRP. We used a combination of two independent SNPs (rs1169300 and rs2794520), which had strong evidence for an association with circulating CRP from genome-wide association studies,[18,19] as the instrument. Because the SNPs were independent ($r^2 = 0.001$), we did not estimate haplotypes. Instead, we summed up the number of variant alleles across the two SNPs, generating an ordinal instrumental variable with categories 0–4, circulating CRP concentrations being the lowest in category 0 and highest in category 4. Previous research and the analyses presented here suggest that the combined instrument as well as the constituent SNPs are associated with circulating CRP but not with lifestyle and environmental confounders. Furthermore, we are not aware of CRP-associated SNPs having an effect on cancer risk other than via circulating CRP. We examined the strength of the instrument using the *F*-statistic and modelled the associations of CRP with cancer outcomes using conventional probit and ivprobit regression. The probit model is an analytical technique transforming binary outcomes to scores on the cumulative standard normal probability distribution.[21] The coefficient in the model, the probit index, indicates how many standard deviations the score increases per unit increase in the exposure.

All statistical analyses were conducted using Stata SE 10.1 (Stata Corporation, Texas, United States).

3. Results

3.1. CRP-associated SNPs and cancer risk

The 13 SNPs included in our analyses are listed in Table 2. With one exception (rs7730843), all the SNPs analysed here were associated with circulating CRP concentrations in our pooled dataset (Supplementary Web Table W1). We observed no clear associations of the SNPs and the overall cancer risk (Table 2), although compared with the CC genotype, carrying one (hazard ratio (HR): 1.05, 95% confidence interval (CI): 0.90, 1.23) or two (HR: 1.20, 95% CI: 1.01, 1.42) variant T alleles at rs1892534 was associated with a slightly elevated risk of

Table 2 – Associations of CRP-associated SNPs and any incident cancer in FINRISK 1992, FINRISK 1997 and Health 2000 studies.

SNP	Gene	Chromosome	N cancer-free	N cancer	HR (95% CI)
rs8192284	*IL6R*	1	17,117	1548	
aa			8677	759	1.0
ac			6930	641	1.03 (0.93, 1.14)
cc			1510	148	1.16 (0.97, 1.38)
p trend					0.2
c-carrier			8440	789	1.05 (0.95, 1.16)
rs2592887	*CRP*	1	17,053	1546	
cc			6141	551	1.0
ct			8072	737	1.00 (0.90, 1.12)
tt			2840	258	0.96 (0.83, 1.12)
p trend					0.7
t-carrier			10,912	995	0.99 (0.89, 1.10)
rs2794520	*CRP*	1	17,170	1557	
gg			6807	614	1.0
ga			7948	731	1.02 (0.92, 1.14)
aa			2415	212	0.94 (0.80, 1.10)
p trend					0.6
a-carrier			10,363	943	1.00 (0.90, 1.11)
rs2650000	*HNF1A*	12	17,168	1558	
cc			5658	518	1.0
ca			8357	744	1.01 (0.90, 1.13)
aa			3153	296	1.10 (0.95, 1.27)
p trend					0.2
a-carrier			11,510	1040	1.03 (0.93, 1.15)
rs7953249	*HNF1A*	12	17,166	1559	
aa			4454	399	1.0
ag			8546	772	1.03 (0.92, 1.17)
gg			4166	388	1.10 (0.96, 1.26)
p trend					0.2
g-carrier			12,712	1160	1.06 (0.94, 1.18)
rs7310409	*HNF1A*	12	17,179	1557	
gg			5562	507	1.0
ga			8487	764	1.04 (0.93, 1.17)
aa			3130	286	1.06 (0.92, 1.23)
p trend					0.4
a-carrier			11,617	1050	1.05 (0.94, 1.17)
rs1169300	*HNF1A*	12	17,122	1556	
gg			8264	758	1.0
ga			7232	648	1.02 (0.92, 1.14)
aa			1626	150	1.02 (0.85, 1.21)
p trend					0.7
a-carrier			8858	798	1.02 (0.93, 1.13)
rs2464196	*HNF1A*	12	17,165	1553	
gg			8273	757	1.0
ga			7267	645	1.02 (0.92, 1.13)
aa			1625	151	1.03 (0.86, 1.22)
p trend					0.7
a-carrier			8892	796	1.02 (0.92, 1.13)
rs2075650	*APOE*	19	17,162	1532	
aa			11,751	1045	1.0
ag			4890	444	1.05 (0.94, 1.17)
gg			521	43	0.89 (0.66, 1.21)
p trend					0.8
g-carrier			5411	487	1.04 (0.93, 1.15)

(*continued on next page*)

Table 2 (continued)

SNP	Gene	Chromosome	N cancer-free	N cancer	HR (95% CI)
rs1892534[a]	*LEPR*	1	10,558	987	
cc			2816	244	1.0
ct			5243	478	1.05 (0.90, 1.23)
tt			2499	265	1.20 (1.01, 1.42)
p trend					0.043
t-carrier			7742	743	1.10 (0.95, 1.27)
rs7730843[b]	*SLC1A3*	9	5505	554	
gg			3513	380	1.0
ga			1749	150	0.84 (0.70, 1.02)
aa			243	24	0.97 (0.64, 1.46)
p trend					0.2
a-carrier			1992	174	0.86 (0.72, 1.03)
rs10778213[c]	Unknown	10	12,265	1147	
cc			4261	375	1.0
ct			5895	553	0.98 (0.87, 1.13)
tt			2109	219	1.15 (0.97, 1.36)
p trend					0.2
t-carrier			8004	772	1.03 (0.91, 1.16)
rs769449[d]	*APOE*	19	6566	566	
gg			4582	394	1.0
ga			1814	152	0.96 (0.80, 1.16)
aa			170	20	1.36 (0.87, 2.14)
p trend					0.7
a-carrier			1984	172	1.00 (0.83, 1.19)

Abbreviations: IL6R: interleukin-6 receptor; HNF1A: hepatocyte nuclear factor 1A; APOE: apolipoprotein E; LEPR: leptin receptor; SLC1A3: solute carrier family 1, member 3.

[a] FINRISK 1992 and Health 2000 only.

[b] Health 2000 only.

[c] FINRISK 1997 and Health 2000 only.

[d] FINRISK 1997 only.

cancer. However, as rs1892534 was successfully genotyped only in FINRISK 1992 and Health 2000 studies, the number of cancer cases with this SNP measurement was modest ($n = 987$) and the estimates thus imprecise.

There were no clear associations of any of the CRP SNPs with the risk of colorectal, prostate or breast cancers (Table 3). We observed weak evidence for associations of rs1892534 with an increased prostate cancer risk (p trend: 0.06) and rs2794520 with an increased breast cancer risk (p trend: 0.07) but the numbers of cancer cases (prostate cancer $n = 150$, breast cancer $n = 328$) were small and the effect estimates thus imprecise.

Two SNPs were associated with an increase in lung cancer risk (Table 3). Individuals carrying one or two variant A alleles at rs1169300 or rs2464196 (both on chromosome 12q24.31) had approximately 1.5- and 2-fold increased risk of lung cancer, respectively (p trend for both: 0.007). These SNPs are located on the *HNF1A* (hepatocyte nuclear factor 1A) gene within four kilobases of each other and were in linkage disequilibrium ($r^2 = 0.99$). Carrying the variant allele at four other SNPs (G at rs7953249, A at rs7310409, A at rs265000 or T at rs10778213) was associated with a slightly increased lung cancer risk. However, the effect estimates for these SNPs were imprecise and are likely to be due to linkage disequilibrium ($r^2 = 0.7$–0.8 for all pair-wise comparisons).

The findings of the sensitivity analyses (in the individuals with non-melanoma skin cancer as their only cancer included in the overall number of cancer cases and with additional adjustment for the geographical area of origin) did not markedly differ in direction or magnitude from the main findings and are not presented.

3.2. *Instrumental variable analyses*

We considered the combined total number of variant alleles at rs1169300 and rs2794520 to be a valid instrument for circulating CRP because it was a relatively strong instrument ($F = 39.0$) and not associated with age, sex or body mass index. Furthermore, we have no reason to believe that the combination of alleles at these SNPs would be associated with cancer risk through any other mechanism than CRP.

In the conventional probit analyses elevated circulating CRP was associated with small increases in the probability of having cancer overall or colorectal, lung and prostate cancers specifically, though not with the probability of having breast cancer (Table 4). In the instrumented probit analyses the point estimates indicate that CRP may be associated with a slightly increased probability of having colorectal, prostate or breast cancer or cancer overall and a slightly decreased probability of having lung cancer. However, the 95% CIs cross the null-value making the findings compatible with no association or associations in the opposite directions. The estimates were little affected by adjustment for baseline characteristics.

Table 3 – Associations of CRP-associated SNPs and main types of incident cancer in FINRISK 1992, FINRISK 1997 and Health 2000.

SNP	N cancer-free	Colorectal cancer		Lung cancer		Prostate cancer		Breast cancer	
		N cancer	HR (95% CI)	N cancer	HR (95% CI)	N cancer	HR (95% CI)	N cancer	HR (95% CI)
rs8192284	17,117	152		120		263		324	
aa	8677	69	1.0	50	1.0	125	1.0	165	1.0
ac	6930	72	1.25 (0.90, 1.74)	59	1.40 (0.96, 2.04)	108	1.05 (0.81, 1.36)	130	0.97 (0.77, 1.22)
cc	1510	11	0.95 (0.50, 1.79)	11	1.25 (0.65, 2.41)	30	1.44 (0.97, 2.15)	29	1.03 (0.70, 1.53)
p trend			0.5		0.2		0.1		0.9
c-carrier	8440	83	1.20 (0.87, 1.65)	70	1.37 (0.95, 1.97)	138	1.12 (0.88, 1.42)	159	0.98 (0.79, 1.22)
rs2592887	17,053	151		118		261		327	
cc	6141	57	1.0	41	1.0	95	1.0	110	1.0
ct	8072	74	0.96 (0.68, 1.36)	60	1.13 (0.76, 1.69)	126	0.99 (0.76, 1.29)	151	1.03 (0.81, 1.32)
tt	2840	20	0.70 (0.42, 1.17)	17	0.87 (0.49, 1.54)	40	0.86 (0.60, 1.25)	66	1.25 (0.92, 1.70)
p trend			0.2		0.8		0.5		0.2
t-carrier	10,912	94	0.89 (0.64, 1.24)	77	1.06 (0.73, 1.55)	166	0.95 (0.74, 1.23)	217	1.09 (0.87, 1.37)
rs2794520	17,170	152		121		263		327	
gg	6807	62	1.0	54	1.0	102	1.0	120	1.0
ga	7948	75	1.02 (0.73, 1.43)	58	0.95 (0.66, 1.38)	124	1.03 (0.80, 1.34)	147	1.05 (0.82, 1.33)
aa	2415	15	0.64 (0.36, 1.13)	9	0.46 (0.23, 0.94)	37	0.99 (0.68, 1.44)	60	1.38 (1.01, 1.88)
p trend			0.2		0.07		0.9		0.07
a-carrier	10,363	90	0.92 (0.67, 1.28)	67	0.83 (0.58, 1.19)	161	1.02 (0.80, 1.31)	207	1.13 (0.90, 1.41)
rs2650000	17,168	153		122		263		328	
cc	5658	44	1.0	32	1.0	95	1.0	122	1.0
ca	8357	79	1.27 (0.88, 1.84)	55	1.19 (0.77, 1.84)	119	0.89 (0.68, 1.16)	150	0.86 (0.68, 1.09)
aa	3153	30	1.36 (0.85, 2.16)	35	2.07 (1.28, 3.35)	49	1.01 (0.72, 1.44)	56	0.87 (0.63, 1.19)
p trend			0.2		0.004		0.9		0.3
a-carrier	11,510	109	1.29 (0.91, 1.84)	90	1.43 (0.95, 2.14)	168	0.92 (0.71, 1.18)	206	0.86 (0.69, 1.08)
rs7953249	17,166	153		122		263		328	
aa	4454	35	1.0	27	1.0	72	1.0	92	1.0
ag	8546	76	1.17 (0.78, 1.74)	54	1.04 (0.65, 1.64)	123	0.91 (0.68, 1.22)	159	0.92 (0.71, 1.18)
gg	4166	42	1.39 (0.89, 2.18)	41	1.65 (1.01, 2.68)	68	1.09 (0.78, 1.52)	77	0.93 (0.69, 1.26)
p trend			0.1		0.034		0.6		0.6
g-carrier	12,712	118	1.24 (0.85, 1.81)	95	1.23 (0.80, 1.89)	191	0.97 (0.74, 1.27)	236	0.92 (0.72, 1.17)
rs7310409	17,179	153		122		262		328	
gg	5562	51	1.0	31	1.0	86	1.0	113	1.0
ga	8487	71	0.98 (0.68, 1.41)	62	1.37 (0.89, 2.12)	121	0.99 (0.75, 1.30)	164	0.99 (0.78, 1.26)
aa	3130	31	1.18 (0.76, 1.85)	29	1.70 (1.03, 2.83)	55	1.24 (0.88, 1.74)	51	0.84 (0.60, 1.17)
p trend			0.5		0.036		0.3		0.4
a-carrier	11,617	102	1.03 (0.74, 1.45)	91	1.47 (0.97, 2.20)	176	1.06 (0.82, 1.37)	215	0.95 (0.76, 1.19)
rs1169300	17,122	152		122		261		328	
gg	8264	74	1.0	44	1.0	129	1.0	172	1.0
ga	7232	65	1.07 (0.76, 1.49)	58	1.49 (1.01, 2.20)	104	0.97 (0.75, 1.26)	132	0.91 (0.73, 1.15)
aa	1626	13	0.93 (0.51, 1.67)	18	1.97 (1.14, 3.40)	28	1.12 (0.75, 1.69)	24	0.71 (0.46, 1.09)
p trend			0.9		0.007		0.7		0.1
a-carrier	8858	78	1.04 (0.76, 1.43)	76	1.58 (1.10, 2.28)	132	1.00 (0.79, 1.28)	156	0.87 (0.70, 1.09)
rs2464196	17,165	153		122		263		327	
gg	8273	75	1.0	46	1.0	129	1.0	172	1.0
ga	7267	65	1.05 (0.75, 1.47)	57	1.47 (1.00, 2.16)	105	0.98 (0.76, 1.27)	131	0.90 (0.72, 1.14)
aa	1625	13	0.91 (0.51, 1.64)	18	1.97 (1.14, 3.40)	29	1.17 (0.78, 1.74)	24	0.71 (0.46, 1.09)
p trend			0.9		0.007		0.6		0.1
a-carrier	8892	78	1.03 (0.75, 1.41)	75	1.56 (1.08, 2.26)	134	1.02 (0.80, 1.29)	155	0.87 (0.70, 1.07)
rs2075650	17,162	152		119		261		328	
aa	11,751	109	1.0	78	1.0	180	1.0	223	1.0
ag	4890	40	0.92 (0.64, 1.32)	38	1.21 (0.82, 1.78)	71	0.97 (0.74, 1.28)	100	1.10 (0.87, 1.39)
gg	521	3	0.61 (0.19, 1.91)	3	0.81 (0.25, 2.56)	10	1.21 (0.64, 2.29)	5	0.49 (0.20, 1.20)
p trend			0.4		0.6		0.8		0.8
g-carrier	5411	43	0.88 (0.62, 1.26)	41	1.17 (0.80, 1.71)	81	1.00 (0.77, 1.30)	105	1.04 (0.82, 1.31)
rs1892534[a]	10,558	100		69		150		235	
cc	2816	24	1.0	22	1.0	34	1.0	64	1.0
ct	5243	49	1.10 (0.68, 1.79)	30	0.72 (0.42, 1.25)	69	1.10 (0.73, 1.65)	111	0.92 (0.68, 1.26)
tt	2499	3	1.20 (0.69, 2.09)	17	0.81 (0.43, 1.53)	47	1.50 (0.97, 2.34)	60	1.02 (0.72, 1.45)
p trend			0.5		0.5		0.06		0.9
t-carrier	7742	76	1.14 (0.72, 1.80)	47	0.75 (0.45, 1.25)	116	1.23 (0.84, 1.80)	171	0.96 (0.72, 1.27)
rs7730843[b]	5505	67		28		87		135	
gg	3513	46	1.0	17	1.0	64	1.00	88	1.0
ga	1749	19	0.89 (0.52, 1.52)	10	1.19 (0.54, 2.59)	22	0.75 (0.46, 1.21)	37	0.90 (0.62, 1.33)
aa	243	2	0.68 (0.17, 2.81)	1	0.92 (0.12, 6.93)	1	0.24 (0.03, 1.75)	10	1.77 (0.92, 3.40)
p trend			0.5		0.8		0.07		0.5
a-carrier	1992	21	0.87 (0.52, 1.45)	11	1.16 (0.54, 2.47)	23	0.68 (0.42, 1.10)	47	1.01 (0.71, 1.44)
rs10778213[c]	12,265	120		81		203		234	
cc	4261	41	1.0	18	1.0	69	1.0	76	1.0
ct	5895	59	0.96 (0.65, 1.43)	45	1.65 (0.95, 2.85)	89	0.85 (0.62, 1.17)	118	1.06 (0.79, 1.41)

(*continued on next page*)

Table 3 (continued)

SNP	N cancer-free	Colorectal cancer		Lung cancer		Prostate cancer		Breast cancer	
		N cancer	HR (95% CI)	N cancer	HR (95% CI)	N cancer	HR (95% CI)	N cancer	HR (95% CI)
tt	2109	20	0.97 (0.57, 1.66)	18	1.92 (1.00, 3.69)	45	1.28 (0.88, 1.87)	40	1.03 (0.70, 1.51)
p trend			0.9		0.041		0.4		0.8
t-carrier	8004	79	0.97 (0.66, 1.41)	63	1.72 (1.02, 2.90)	134	0.96 (0.72, 1.29)	158	1.05 (0.80, 1.38)
rs769449[d]	6566	53		51		112		92	
gg	4582	41	–[e]	34	1.0	82	1.0	60	1.0
ga	1814	12	–	15	1.08 (0.59, 1.98)	26	0.79 (0.51, 1.23)	28	1.17 (0.75, 1.84)
aa	170	–	–	2	1.54 (0.37, 6.41)	4	1.32 (0.49, 3.61)	4	1.82 (0.66, 5.01)
p trend			–		0.6		0.6		0.3
a-carrier	1984	12	0.67 (0.35, 1.28)	17	1.12 (0.62, 2.00)	30	0.83 (0.55, 1.27)	32	1.23 (0.80, 1.88)

[a] FINRISK 1992 and Health 2000 only.
[b] Health 2000 only.
[c] FINRISK 1997 and Health 2000 only.
[d] FINRISK 1997 only.
[e] Not estimated due to insufficient numbers of participants with available data.

4. Discussion

4.1. CRP-associated SNPs and cancer risk

We found no clear associations of CRP-associated SNPs and the overall cancer risk or the risk of colorectal, prostate or breast cancers. Overall, our findings agree with those of previous studies suggesting that genetic determinants or markers of circulating CRP are not associated with cancer risk. No associations have been reported between variants on the CRP gene and the overall cancer risk or the risk of prostate, breast or endometrial cancers.[11,22,23] However, the evidence for an association of genetic determinants of CRP and colorectal cancer risk is contradicting: association of CRP SNPs with colorectal cancer[24] and benign colorectal adenoma[25] have been reported in some studies, whilst in others no associations were found.[22,26] One explanation for these inconsistent findings could be that cancer is a heterogeneous group of diseases and analysing different cancer types or sub-types (e.g. colon and rectal cancers) as combined outcomes may conceal any associations of CRP SNPs with some malignancies. Another explanation could be that the elevated circulating CRP concentrations in individuals with cancer, which have been reported in previous observational studies[4] and are often observed in clinical settings, are markers of a host response to the malignant process or disease progression or severity, and unrelated to genetically elevated CRP.

In a meta-analysis of observational studies of circulating CRP and cancer, the evidence for an association was most convincing for lung cancer (random effects overall odds ratio per doubling of CRP: 1.32, 95% CI: 1.08, 1.61).[9] A small number of SNPs in our study were associated with an increased lung cancer risk and one previous study, in which variant allele T at rs1205 on the CRP gene[22] was associated with a slightly increased lung cancer risk, supports the hypothesis that some CRP-associated SNPs may be associated with lung cancer risk. However, these findings were not corroborated by a genome-wide association study of lung cancer.[27]

4.2. Instrumental variable analyses

Our instrumental variable analyses provided no clear evidence for a causal association of CRP and the cancer risk. Our findings were similar to those of the only previous study using CRP-associated SNPs as instruments for circulating CRP

Table 4 – Associations of CRP with cancer outcomes, based on standard and instrumental probit model analyses (a combination of rs1169300 and rs2794520 as an instrument).

Model[a]	Probit index (95% CI)	
p trend	Minimum adjusted[b]	Fully adjusted[c]
Any cancer (n = 1156)		
Probit	0.07 (0.04, 0.09) $p < 0.001$	0.08 (0.05, 0.11) $p < 0.001$
Instrumented probit	0.10 (−0.14, 0.34) $p = 0.4$	0.09 (−0.13, 0.32) $p = 0.4$
Colorectal cancer (n = 116)		
Probit	0.08 (0.02, 0.13) $p = 0.009$	0.08 (0.02, 0.13) $p = 0.01$
Instrumented probit	0.07 (−0.46, 0.61) $p = 0.8$	0.07 (−0.44, 0.58) $p = 0.8$
Lung cancer (n = 99)		
Probit	0.14 (0.087, 0.20) $p < 0.001$	0.16 (0.10, 0.23) $p < 0.001$
Instrumented probit	−0.12 (−0.64, 0.39) $p = 0.7$	−0.16 (−0.64, 0.33) $p = 0.5$
Prostate cancer (n = 196)		
Probit	0.04 (−0.005, 0.09) $p = 0.08$	0.05 (0.003, 0.10) $p = 0.037$
Instrumented probit	0.03 (−0.41, 0.47) $p = 0.9$	0.03 (−0.38, 0.45) $p = 0.9$
Breast cancer (n = 229)		
Probit	−0.02 (−0.06, 0.03) $p = 0.5$	−0.01 (−0.06, 0.04) $p = 0.7$
Instrumented probit	0.14 (−0.24, 0.52) $p = 0.3$	0.12 (−0.25, 0.49) $p = 0.5$

[a] Based on individuals with available data on the instrumental SNPs, circulating CRP, age, sex and body mass index (BMI). Of these 13,044 individuals 1156 had some form of cancer and 11,888 were cancer-free.
[b] Adjusted for age only.
[c] Adjusted for age, sex (in models of colorectal and lung cancer) and body mass index.

concentrations, in which no evidence for a causal association of CRP with cancer was reported.[11] However, similarly to the previous study, our analyses of the main cancer types were based on modest numbers of cancer cases and the instrumented effect estimates thus imprecise.

Taken together, our findings and those of previous studies suggest that the associations of elevated circulating CRP concentrations and increased cancer risk reported in observational studies, even in those with a prospective design and adjustment for a large number of potential confounders may be due to residual confounding or reverse causality. Thus, the elevated CRP concentrations in individuals with cancer are more likely to be indicators of the severity or extent of the malignant process than causal agents in cancer development. Similarly, the genetic determinants or markers of CRP concentrations may be indicators and predictors of cancer progression, rather than causes of it. Indeed, some emerging evidence suggests that variants of the CRP gene are related to progression and metastasis in prostate[28] and thoracic oesophageal[29] cancers.

4.3. *Strengths and weaknesses of our study*

An important strength of our study is that we investigated a larger number of SNPs than previous studies focusing on CRP-related genetic variants and cancer. We used data from three population-based studies and ascertained cancer cases from cancer registrations, which capture over 95% of all cancer events in Finland.[17] It is unlikely that population stratification has biased our findings because CRP concentrations are not markedly ethnically patterned[10] and the population from which our participants came from is genetically homogeneous.[12] Missing SNP or circulating CRP data may have reduced the power of our analyses but it is unlikely that these data have biased our findings as they were missing due to insufficient DNA or blood, or failure in laboratory analyses, and thus likely to be missing completely at random. However, it is challenging to recruit a study population that will develop sufficient numbers of incident cancers, let alone cases of histological or other sub-types of cancer, to ensure statistical power for analyses of the risk of specific cancer types, and our analyses were based on modest numbers of cancer cases. Also, we conducted a large number of statistical tests, which increases the probability that some of the associations we observed may be due to chance.

5. Conclusions

We found no clear evidence for a relationship between CRP-associated SNPs and the overall cancer risk or the risk of colorectal, lung, prostate or breast cancers. Our instrumental variable analyses provided no evidence for a causal association between CRP and cancer. Taken together, these findings support the hypothesis that elevated circulating CRP concentrations are more likely to be indicators of a host response to the malignant process and cancer progression or severity, rather than have a causal role in cancer development.

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Conflict of interest statement

None declared.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.ejca.2010.07.032.

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