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AMACR polymorphisms, dietary intake of red meat and dairy and prostate cancer risk

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Abstract

Background—Alpha-methylacyl CoA racemase (AMACR) is an enzyme involved in fatty acids metabolism. One of AMACRs primary substrates, phytanic acid, is principally obtained from dietary red meat/dairy which are associated with prostate cancer (PCa) risk. AMACR is also a tumor tissue biomarker over-expressed in PCa. In this study, we explored the potential relationship between *AMACR* polymorphisms, red meat/dairy intake and PCa risk.

Methods—Caucasian participants from two population-based PCa case-control studies were included. *AMACR* SNPs were selected to capture variation across the gene and regulatory regions. Red meat and dairy intake was determined from food frequency questionnaires. The odds ratio (OR) of PCa (overall and by disease aggressiveness) was estimated by logistic and polytomous regression. Potential interactions between genotypes and dietary exposures were evaluated.

Results—Data from 1,309 cases and 1,267 controls were analyzed. Carriers of the variant T allele (rs2287939) had an OR of 0.81 (95% CI 0.68-0.97) for less aggressive PCa, but no alteration in risk for more aggressive PCa. Red meat consumption was positively associated with PCa risk, and the association was stronger for more aggressive disease (lowest vs. highest tertile OR= 1.55, 95% CI 1.10-2.20). No effect modification of *AMACR* polymorphisms by either dietary red meat or dairy intake on PCa risk was observed.

Conclusions—Prostate cancer risk varied by level of red meat intake and by one *AMACR* SNP, but there was no evidence for gene-environment interaction. These findings suggest that the effects of *AMACR* polymorphisms and red meat and dairy on PCa risk are independent.

Introduction

Prostate cancer (PCa) development involves both genetic and environmental factors.(1-3) Twin studies of PCa have estimated that up to 42% of PCa risk is attributable to genetic factors, with the remaining risk due to environmental factors.(1,4) A western diet has been associated with a higher relative risk of PCa, and fat is a principal and distinguishing components of the western diet. Alpha-methylacyl CoA racemase (AMACR) metabolizes dietary fatty acids(5) and is a well established PCa tumor tissue biomarker.(6) AMACR has been implicated in PCa on multiple levels as both AMACR protein expression and mRNA

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transcripts are upregulated in PCa.(6,7) Further, in *in vitro* studies, blocking AMACR leads to a significantly impaired PCa proliferation.(8) Therefore, the over-expression of AMACR in PCa cells may lead to more efficient use of energy from these fatty acids.

Levels of phytanic acid, one of the primary substrates of AMACR, have been found to be elevated in PCa,(9) and there is biological rationale for its role in PCa. First, phytanic acid can increase AMACR expression in human prostate cells.(10) In addition, the metabolism of phytanic acid results in the production of reactive oxygen species that can lead to DNA damage.(11) Finally, phytanic acid (and its metabolites) binds to nuclear hormone receptors (retinoid acid receptors (RARs) and peroxisome proliferator-activated receptor (PPARα)), which have been linked to carcinogenesis.(12,13) Dietary sources high in phytanic acid include red meat and diary products, both of which have been associated with an elevation in PCa risk.(14,15)

Studies of single nucleotide polymorphisms (SNPs) in the *AMACR* gene have been performed and yielded conflicting results with regard to PCa risk. (16-20) Potentially, the level of red meat and dairy intake may modify the association between *AMACR* SNPs and PCa risk through altering levels of phytanic acid. To date, only one study has explored this potential gene-environment interaction.(19) In this study, we utilize data, DNA samples and food frequency questionnaires from two population-based case-control studies of PCa to explore the potential relationship between polymorphisms in *AMACR* and dietary red meat and dairy.

Methods

Study Population

The study population consists of men in two prior population-based case-control studies of PCa.(21,22) Cases were residents of King County, Washington with histologically confirmed PCa who were identified from the Seattle-Puget Sound SEER cancer registry. In Study I, incident cases were diagnosed between January 1, 1993, and December 31, 1996. In Study II, incident cases were diagnosed between January 1, 2002, and December 31, 2005. Male residents of King County, Washington with no history of a physician's diagnosis of PCa were recruited as controls and were identified using random digit telephone dialing. Controls were frequency matched to cases by five-year age groups, and enrolled evenly throughout the study period. A total of 2,244 eligible cases were identified and 1,754 (78%) participated; 2,448 eligible controls were identified and 1,645 (67%) participated.

Genotyping

Blood was collected and genomic DNA isolated from those men who consented. Haplotype tagging SNPs (with a minor allele frequency > 5.0%) for *AMACR* were chosen to cover the transcript of interest (+ 5 kb upstream and downstream). Genotyping was performed with the Applied Biosystems (ABI) SNPlex™ Genotyping System. Proprietary GeneMapper® software was used for calling alleles (www.appliedbiosystems.com). Allele specific discrimination was determined with the ABI 3730*xl* DNA Analyzer. Blind duplicates were included (5%) as quality control and 98.5% or greater agreement was observed on genotyping calls. Laboratory personnel were blinded to participant case-control status and DNA batches contained similar numbers of case-control samples.

DNA was available from 83% and 82% of the interviewed cases and controls, respectively (1,309 Caucasian cases and 1,267 Caucasian controls). This study was limited to Caucasians as the allele frequencies differed significantly between Caucasians and African-Americans for 12 of 13 *AMACR* SNPs ($p < 0.05$) and the number of African Americans was too small for separate analyses of this subgroup.

Data Collection

In-person interviews were conducted by trained staff. Data on demographic and lifestyle factors, medical and family history, and PCa screening history (PSA and DRE) were collected. Body mass index (BMI) was determined from self-reported height and weight (one year prior to reference date: date of diagnosis for cases and a randomly assigned date for controls that approximated the distribution of cases' diagnosis dates). All subjects completed a self-administered food frequency questionnaire (FFQ) that inquired about usual dietary intake during the 3-5 years before the reference date. The FFQs used in both studies were similar. Specifically, the line items for meat and dairy were nearly identical across instruments so the dietary data were combined to define the dietary exposures of interest for this analysis.

The FFQ was divided into three sections: 1) adjustment questions; 2) food items; and, 3) summary questions. The adjustment items consisted of 13 questions on types of foods and preparation techniques, which were used to alter how analysis software calculates the nutrient content of specific food items. The main section consisted of 99 food items, with questions on usual frequency (from “never or less than once a month” to “2+ times/day” for foods and “6+ times per day” for beverages) and portion size (small, medium or large, compared to the stated medium portion size). Finally, the summary questions consisted of three questions on the usual intake of fruits and vegetables and use of fat in cooking. These questions are used to reduce the measurement bias from over-reported total food consumption when there are long lists within food groups (i.e., 22 vegetables). The nutrient database used to analyze the FFQ was derived from the Nutrition Data Systems for Research (NDS-R, University of Minnesota Nutrition Coordinating Center). The primary source for nutrient values in this database is the USDA Nutrient Database for Standard Reference and its periodic updates as well as information from food manufacturers. We excluded outliers (35 cases and 30 controls) for caloric intake (<800 kcal/day; > 5000 kcal/day) and for BMI (<18.5; > 45.0) (15 cases and 18 controls).

Clinical information on PCa cases was obtained from the SEER cancer registry, including Gleason score and tumor stage (SEER summary stage). For men who did not have a radical prostatectomy as primary treatment, staging was based on clinical information whereas pathological stage was used for those undergoing radical prostatectomy.

Statistical Analysis

SNPs were tested for Hardy-Weinberg equilibrium in controls using the Exact test. Assuming a dominant genetic model, the relative risk of PCa was estimated with logistic regression for each SNP genotype, adjusting for age (5-year groups). We evaluated family history of PCa and PSA screening history as potential confounders, but neither variable changed the risk estimates by > 10% and thus were not included in the final models. Polytomous regression was used to calculate risk estimates by disease aggressiveness (controls, less aggressive cases, more aggressive cases). Disease aggressiveness was based on a composite variable incorporating Gleason score, stage and PSA where more aggressive cases were defined by a Gleason score of 7(4+3) or greater, or non-localized stage, or PSA > 20 ng/mL at time of diagnosis.

Dietary red meat and dairy intake were determined from the FFQs. Tertiles were calculated for average servings per day of red meat, total dairy, high-fat dairy and low-fat dairy intake from distributions in the control population. High-fat dairy consisted of full fat milk, cheese and yogurt. Low-fat dairy consisted of reduced fat milk, non-fat milk, low fat or non-fat frozen desserts, low fat cheese and yogurt. The relative risk of PCa was then determined with logistic regression adjusting for age. We evaluated family history of PCa, PSA

screening history, BMI and total caloric intake as potential confounders. PSA screening history, BMI and total caloric intake all resulted in a change in the risk estimate of > 10% and were included in the final models. Polytomous regression was used to calculate the relative risks by disease aggressiveness.

To evaluate whether red meat or dairy intake modified the effect of *AMACR* variant alleles on PCa risk, models with an interaction term were evaluated. This model also included age, PSA screening history, BMI and total caloric intake. The reduced model, without the interaction term, was then compared with the full model, containing the interaction term, using a likelihood ratio test. All statistical analyses were conducted using STATA software, Version 11 (Stata, Inc., College Station, TX).

Results

Two SNPs (rs15612 and rs16892096) were not in HWE (p-value < 0.05) in controls and were excluded from further analyses. Also, two SNPs were in perfect linkage disequilibrium (rs2287939 and rs3776543), so only one (rs2287939) was included in the analysis. Table 1 gives the characteristics of cases and controls. Cases were more likely to have a family history of PCa (22% vs. 11%, $p < 0.001$) and report more PSA tests within the preceding five years ($p < 0.001$) before reference date. Of the cases, 435 (33%) had a comparatively more aggressive phenotype with Gleason Score ≥ 4+3 or non-localized stage disease or PSA > 20 ng/mL at diagnosis. Table 2 shows the genotype distributions for cases and controls, along with the age-adjusted ORs for PCa based on a dominant genetic model. None of these results reached statistical significance. However, when evaluating the risk by PCa aggressiveness (Table 3), carriers of the variant T allele in rs2297939 had a 19% reduction (OR= 0.81, 95% CI 0.68 – 0.97) in the relative risk for less aggressive PCa but no alteration in the risk of more aggressive PCa (OR = 1.06, 95% CI 0.84 – 1.32). Based on analysis using a likelihood ratio test, carriers of the T allele demonstrated a significantly lower risk of less compared to more aggressive PCa ($p = 0.02$) than did carriers of the C allele. A second SNP (rs2278008) also demonstrated a statistically significant ($p = 0.04$) difference in risk estimates for carriers of the variant C allele in the less vs. more aggressive PCa group, however the ORs for less vs. more aggressive disease were not significantly different.

Table 4 gives the multivariate risk estimates of PCa based on dietary intake of red meat and dairy. Higher red meat intake was associated with a 43% elevation in the relative risk for PCa (95% CI 1.11 – 1.84). When examining whether red meat intake was associated with less vs. more aggressive PCa in the polytomous model, red meat consumption was associated with an increased risk in both groups. Compared to the lowest tertile of red meat consumption, there was a 38% and 55% increased relative risk of less aggressive (95% CI 1.05 – 1.82) and more aggressive PCa (95% CI 1.10 – 2.20), respectively. No associations were seen for different categories of dairy intake: total dairy, high fat dairy or low fat dairy. There was no evidence for any interactions (all p-values > 0.1) between levels of meat intake and *AMACR* genotype on overall risk of PCa (Table 5) or PCa aggressiveness (data not shown). There was also no evidence for any interaction between genotypes and dairy intake on PCa risk.

Discussion

In this population-based case-control study, we explored the relationships between red meat and dairy intake, *AMACR* polymorphisms and the relative risk of PCa. We found that *AMACR* polymorphisms and red meat intake were both associated with relative risk of PCa, yet we found no evidence for any gene-environment interactions. These findings suggest that the effects of *AMACR* polymorphisms and red meat on PCa risk are independent.

AMACR is involved in the β-oxidative metabolism of certain fatty acids and has been found to be upregulated in a variety of cancers, including PCa.(6) The mechanism by which AMACR affects PCa risk is not fully understood. One possibility is that the reactive oxygen species created from the enzymatic activity of AMACR leads to DNA damage.(11) Alternatively, AMACR may impact carcinogenesis by affecting levels of the androgen receptor and IGF-1.(23) Phytanic acid is a major substrate for AMACR and the primary dietary source of phytanic acid is red meat and dairy, both of which have been positively associated with PCa risk.(15,24) Athough the data on the relationship between phytanic acid levels and PCa risk has been mixed(9,25) phytanic acid can upregulate the expression of *AMACR* and enhance its enzymatic activity.(10) Further, phytanic acid may be involved in PCa development as it acts as a substrate for ligands in signal transduction pathways that may be involved with carcinogenesis. (26) Accordingly, the potential interaction between *AMACR* gene variants and red meat/dairy intake is of interest.

We found a reduction in the relative risk of less aggressive PCa (localized stage, Gleason 2-7 (3+4), PSA < 20ng/mL at diagnosis) with one *AMACR* polymorphism (rs2287939). This is a coding non-synonymous SNP that results in an amino acid change at position 201 (leucine to serine). The literature on *AMACR* SNPs and PCa risk presents conflicting results. In two studies,(16,17) rs2287939 was associated with a risk reduction in familial, but not sporadic, PCa. The ORs for sporadic disease were similar, although non-significant in both studies. An Australian study that did not evaluate rs2287939 found two *AMACR* SNPs (rs3195676 and rs10941112) were associated with reduced risks of sporadic PCa(27) and these SNPs are in modest linkage disequilibrium with rs2287939. Two other studies found no significant associations between PCa and *AMACR* SNPs. (19,20) The most extensively previously studied SNP (rs3195676) was associated with a borderline risk reduction in our study (OR 0.86, 95% CI 0.72 – 1.04).

Although most, but not all studies, have found an increased risk of PCa in those consuming higher levels of red meat (15), few studies have explored red meat intake in relation to more aggressive PCa. Two analyses from the Health Professionals Follow-Up Study reported higher relative risks (RR) of advanced (RR= 2.6, 95% CI 1.2 – 5.8) (28) and metastatic (RR= 1.6, 95% CI 1.0 – 2.5) (29) PCa in those consuming higher quantities of red meat. In our study, we found that those in the highest tertile of average servings/day of red meat had a 55% increase in the relative risk of more aggressive PCa (95% CI 1.10 – 2.20).

Considering the suggested associations of both red meat intake and *AMACR* polymorphisms with PCa risk and their potential biological relationship, we explored the possible interactions of *AMACR* genotypes with both red meat (via phytanic acid) and dairy on PCa risk. We did not find any evidence for effect modification. One prior study investigated this issue and found that those consuming > 25th percentile of red meat who were homozygote carriers of the variant allele for rs3195676 had a reduced risk of PCa.(19) In that study, however, none of the individual *AMACR* SNPs or quantity of red meat intake was independently associated with PCa risk.

There are limitations to our study. Although participants answered the FFQ with consideration to consumption over the prior 3-5 years, we cannot exclude recall bias, although it is unlikely that dietary recall would be less reliable in cases than controls. Further, although red meat and dairy are primary sources of pytanic acid, there are other dietary sources we did not consider. In addition, our finding of an association between an *AMACR* polymorphism (rs2287939) and PCa risk is modest, and failed to retain statistical significance following adjustment for multiple testing. As a result, this finding should be interpreted cautiously. Despite these limitations, our data are consistent with prior findings of an increased risk of aggressive PCa in men with higher dietary red meat intake from the

Health Professionals Follow-Up Study, (28,29) while suggesting a reduction in the relative risk of less aggressive PCa with one *AMACR* SNP (rs2287939). Although we did not find any evidence for gene-environment interactions between red meat and dairy consumption and *AMACR* polymorphisms, future study of candidate genes such as *AMACR* and environmental exposures may help to advance understanding of the multifactorial nature of PCa risk.

Conclusion

Red meat consumption was associated with an increased relative risk PCa, and the association is stronger for men with more aggressive disease features. Carriers of the variant T allele in *AMACR* SNP (rs2287939) had a lower relative risk of less aggressive PCa, but no alteration in risk was observed for more aggressive PCa. We found no evidence for an interaction between *AMACR* polymorphisms and dietary red meat or dairy intake on PCa risk.

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Table 1
Characteristics of Caucasian Cases and Controls with DNA Available

	Cases N (%)	Controls N (%)	P-value
Characteristics	1,309 (100)	1,267 (100)	
Age			
35-49	102 (7.8)	102 (7.8)	0.35
50-54	189 (14.4)	189 (14.4)	
55-59	325 (24.8)	325 (24.8)	
60-64	395 (30.2)	395 (30.2)	
65-69	153 (11.7)	153 (11.7)	
70-74	144 (11.4)	145 (11.1)	
Family history of PCa*			
Negative	1026 (78.4)	1125 (88.8)	< 0.001
Positive	283 (21.6)	142 (11.2)	
PSA test within 5 years prior to referent date+			
None	288 (22.1)	452 (35.9)	< 0.001
1 – 2	320 (24.5)	239 (19.0)	
≥ 3	638 (48.9)	379 (30.1)	
Unknown	58 (4.5)	188 (14.9)	
BMI**			
< 25.0	429 (32.8)	389 (30.7)	0.34
25.0 – 29.9	619 (48.9)	638 (48.7)	
≥ 30.0	242 (18.5)	259 (20.4)	
PSA level (ng/mL)+			
0.0 – 3.9	178 (13.6)	1176 (92.8)	< 0.001
4.0 – 9.9	722 (55.2)	74 (5.8)	
≥ 10	307 (23.4)	17 (1.4)	
Missing	102 (7.8)	0 (0.0)	
Gleason score			
2 – 6	747 (57.2)		
3+4	356 (27.3)		
4+3	76 (5.8)		
8 – 10	126 (9.7)		
Tumor stage			
Local	1023 (78.2)		
Regional	254 (19.4)		
Distant	32 (2.4)		
Treatment			
Radical prostatectomy	770 (58.8)		
Radiation	359 (27.4)		
Androgen deprivation	61 (4.7)		
Other	4 (0.3)		

	Cases N (%)	Controls N (%)	P-value
Active surveillance	115 (8.8)		

*First-degree family history of prostate cancer

**BMI = body mass index (weight/height2)

+PSA measured at diagnosis (cases) or interview (controls)

Table 2

Genotype Distributions and Age-adjusted Relative Risks (RR) of Prostate Cancer Associated with *AMACR* SNPs in Caucasian Men

SNP	Genotype	Cases	Controls	Age-Adjusted RR (95% CI)
rs34680				
	CC	908 (75.1)	922 (76.1)	1.00
	CT + TT	301 (24.9)	290 (23.9)	0.98 (0.81 – 1.20)
rs34688				
	CC	875 (69.4)	877 (70.4)	1.00
	CT + TT	386 (30.6)	368 (29.6)	1.00 (0.83 – 1.20)
rs250412				
	GG	471 (37.5)	466 (37.4)	1.00
	GA + AA	786 (62.5)	780 (62.6)	0.99 (0.84 – 1.18)
rs2278008				
	TT	696 (56.1)	670 (54.2)	1.00
	TC + CC	544 (43.9)	566 (45.8)	0.93 (0.78 – 1.10)
rs2287939				
	CC	636 (50.8)	598 (47.9)	1.00
	CT + TT	617 (49.2)	651 (52.1)	0.93 (0.78 – 1.10)
rs2652130				
	AA	987 (78.2)	973 (78.0)	1.00
	AC + CC	275 (21.8)	274 (22.0)	0.95 (0.78 – 1.16)
rs3195676				
	AA	371 (30.0)	340 (27.5)	1.00
	AG + GG	864 (70.0)	897 (72.5)	0.86 (0.72 – 1.04)
rs6863657				
	GG	1021 (80.9)	1020 (81.7)	1.00
	GA + AA	241 (19.1)	228 (18.3)	1.05 (0.85 – 1.30)
rs7721230				
	AA	477 (39.2)	457 (37.4)	1.00
	AG + GG	740 (60.8)	765 (62.6)	0.92 (0.77 – 1.09)
rs10941112				
	AA	370 (29.8)	343 (27.7)	1.00
	AG + GG	870 (70.2)	895 (72.3)	0.89 (0.74 – 1.07)

*Variable number of cases/controls due to failed genotyping

Table 3

Age-adjusted Relative Risks of Prostate Cancer by *AMACR* Genotypes and Disease Aggressiveness* in Caucasian Men

SNP	Genotype	Less Aggressive	More Aggressive	p-value ^
rs34680				
	CC	1.00 (referent)	1.00 (referent)	
	CT + TT	1.12 (0.91 – 1.38)	0.91 (0.70 – 1.19)	0.15
rs34688				
	CC	1.00 (referent)	1.00 (referent)	
	CT + TT	1.09 (0.90 – 1.32)	0.96 (0.75 – 1.22)	0.32
rs250412				
	GG	1.00 (referent)	1.00 (referent)	
	GA + AA	1.00 (0.83 – 1.20)	0.99 (0.79 – 1.24)	0.92
rs2278008				
	TT	1.00 (referent)	1.00 (referent)	
	TC + CC	0.85 (0.71 – 1.02)	1.08 (0.87 – 1.36)	0.04
rs2287939				
	CC	1.00 (referent)	1.00 (referent)	
	CT + TT	0.81 (0.68 – 0.97)	1.06 (0.84 – 1.32)	0.02
rs2652130				
	AA	1.00 (referent)	1.00 (referent)	
	AC + CC	1.07 (0.87 – 1.32)	0.84 (0.63 – 1.10)	0.10
rs3195676				
	AA	1.00 (referent)	1.00 (referent)	
	AG + GG	0.90 (0.74 – 1.09)	0.84 (0.66 – 1.07)	0.58
rs6863657				
	GG	1.00 (referent)	1.00 (referent)	
	GA + AA	1.03 (0.82 – 1.29)	1.12 (0.84 – 1.48)	0.60
rs7721230				
	AA	1.00 (referent)	1.00 (referent)	
	AG + GG	0.94 (0.78 – 1.13)	0.89 (0.71 – 1.12)	0.69
rs10941112				
	AA	1.00 (referent)	1.00 (referent)	
	AG + GG	0.92 (0.76 – 1.12)	0.85 (0.67 – 1.08)	0.53

* More aggressive cases were defined by a Gleason score of 7(4+3) or greater, or non-localized stage, or PSA > 20 ng/mL at time of diagnosis

^ P-value from a likelihood ratio test for the difference in risk estimates for less versus more aggressive PCa.

Table 4

Relative Risks (RR)* of Prostate Cancer Associated with Red Meat and Dairy Intake in Caucasian Men. By Disease Aggressiveness

	All Cases			Less Aggressive			More Aggressive		
Dietary Factor **	**RR**	**95% CI**	**P (trend)**	**RR**	**95% CI**	**P (trend)**	**RR**	**95% CI**	**P (trend)**
Red meat									
≤ 0.58 serv/day	1.00	Referent	< 0.01	1.00	referent	0.02	1.00	referent	0.01
0.59 – 1.09	1.21	0.97 – 1.51		1.11	0.87 – 1.42		1.43	1.06 – 1.96	
> 1.09	1.43	1.11 – 1.84		1.38	1.05 – 1.82		1.55	1.10 – 2.20	
Total dairy									
≤ 1.33 serv/day	1.00	Referent	0.90	1.00	referent	0.84	1.00	referent	0.57
1.34 – 2.55	1.06	0.86 – 1.29		1.11	0.88 – 1.39		0.95	0.71 – 1.27	
> 2.55	1.01	0.83 – 1.23		0.98	0.78 – 1.23		1.08	0.82 – 1.43	
High-fat dairy ^									
≤ 0.38 serv/day	1.00	Referent	0.44	1.00	Referent	0.62	1.00	Referent	0.40
0.39 – 0.83	0.98	0.80 – 1.20		0.94	0.74 – 1.18		1.08	0.81 – 1.44	
> 0.83	1.08	0.88 – 1.32		1.06	0.85 – 1.33		1.13	0.85 – 1.51	
Low-fat dairy+									
≤ 0.53 serv/day	1.00	Referent	0.98	1.00	Referent	0.69	1.00	Referent	0.57
0.54 – 1.58	1.04	0.85 – 1.27		1.10	0.88 – 1.38		0.92	0.69 – 1.23	
> 1.58	1.00	0.81 – 1.22		0.95	0.76 – 1.20		1.08	0.82 – 1.43	

*Adjusted for age, PSA screening history, BMI and total caloric intake

**Dietary factors were grouped into tertiles based on the distribution of intake in controls

^High-fat dairy consists of full fat milk, cheese and yogurt

+Low-fat dairy consists of reduced fat milk, non-fat milk, low fat or non-fat frozen desserts, low fat cheese and yogurt

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Table 5

Relative Risks (RR)* of Prostate Cancer Associated with *AMACR* Genotypes Stratified by Red Meat Consumption in Caucasian Men ^

		Tertile of Red Meat Intake		
SNP	**Genotype**	**1st RR (95% CI)**	**2nd RR (95% CI)**	**3rd RR (95% CI)**
rs34680				
	CC	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	CT + TT	0.86 (0.59 – 1.24)	1.35 (0.94 – 1.93)	0.87 (061 – 1.23)
rs34688				
	CC	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	CT + TT	0.80 (0.56 – 1.12)	1.25 (0.90 – 1.74)	0.97 (0.70 – 1.34)
rs250412				
	GG	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	GA + AA	0.99 (0.72 – 1.37)	0.94 (0.68 – 1.29)	0.94 (0.69 – 1.28)
rs2278008				
	TT	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	TC + CC	0.97 (0.70 – 1.33)	0.78 (0.57 – 1.07)	0.99 (0.73 – 1.34)
rs2287939				
	CC	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	CT + TT	0.99 (0.73 – 1.36)	0.81 (0.60 – 1.10)	0.91 (0.67 – 1.23)
rs2652130				
	AA	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	AC + CC	0.75 (0.51 – 1.12)	1.22 (0.85 – 1.75)	0.83 (0.57 – 1.18)
rs3195676				
	AA	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	AG + GG	0.83 (0.59 – 1.17)	0.86 (0.61 – 1.19)	0.78 (0.56 – 1.10)
rs6863657				
	GG	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	GA + AA	1.01 (0.67 – 1.51)	1.02 (0.69 – 1.50)	1.14 (0.77 – 1.68)
rs7721230				
	AA	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	AG + GG	0.89 (0.65 – 1.23)	0.91 (0.66 – 1.25)	0.80 (0.59 – 1.10)
rs10941112				
	AA	1.00 (Referent)	1.00 (Referent)	1.00 (Referent)
	AG + GG	0.88 (0.62 – 1.24)	0.88 (0.63 – 1.23)	0.80 (0.57 – 1.13)

*Adjusted for age, family history of prostate cancer, PSA screening, BMI and total caloric intake

^There was no evidence for interaction by LR test (all $p > 0.1$)