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Serum adiponectin levels and tissue expression of adiponectin receptors are associated with risk, stage, and grade of colorectal cancer

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ABSTRACT

Adiponectin has been associated with colorectal cancer (CRC) risk. This study aims to investigate the association of both adiponectin and tissue expression of its receptors with CRC risk as well as clinicopathological characteristics, notably stage and grade. Determination of serum adiponectin and immunohistochemical expression of adiponectin receptors in adenocarcinoma/normal colorectal tissue was performed in samples from 104 newly diagnosed CRC patients and 208 age- and sex-matched controls. Multiple logistic regression odds ratios and 95% confidence intervals for CRC risk were derived, controlling for a series of covariates. Serum adiponectin was negatively associated with CRC risk (odds ratio, 0.72; confidence interval, 0.53-0.99) and also with tumor grade ($P = .05$). Expression of both adiponectin receptors was stronger in adenocarcinoma vs normal tissue ($P = .001$). AdipoR1 expression was negatively associated with nodal stage ($P = .03$); AdipoR2 expression was positively associated with tumor, node, metastasis stage ($P = .01$). Established positive associations with red meat consumption and diabetes, and negative associations with physical exercise and plant food consumption were confirmed along with a more than 60% higher risk associated with central obesity. Adiponectin levels and tissue expression of hormonal receptors seem to be associated not only with CRC risk but also with components of clinicopathological characteristics; given power limitations, these results should be interpreted with caution. The exact nature of the association and the underlying pathophysiological mechanisms need to be further examined in large prospective studies assessing adiponectin and its receptors as novel targets for exploring CRC growth.

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1. Introduction

Obesity, especially abdominal fat accumulation, leads to metabolic inequilibrium, adipose tissue dysfunction, and insulin resistance [1]. Body mass index (BMI), an index of total obesity, and waist to hip ratio (WHR), an index of central obesity, are positively associated with several malignancies, including colorectal cancer (CRC) [2-8]. It has been also recently shown that obesity is related with inflammation in the colorectal mucosa, whereas diet-induced weight loss reduces this inflammatory state, thereby potentially lowering CRC risk [9].

Adiponectin, a 30-kd complement C1q-related protein, is exclusively produced by adipocytes [10]; and its actions are mediated by binding and activating specific adiponectin receptors, AdipoR1 and AdipoR2 [11]. Insulin-sensitizing effects have been attributed to adiponectin [11]; in contrast to other adipokines, such as leptin, however, the circulating levels of this hormone are decreased in obese individuals [1,4]. Low adiponectin levels and insulin resistance have been linked to several obesity-related disease entities, such as type 2 diabetes mellitus, hypertension, and atherosclerosis [1,4,11,12]. Adiponectin has also been proposed as a biological link between obesity and several malignancies, including CRC [4,13-16]. Specifically, adiponectin levels are lower in CRC patients; and its receptors, expressed in both adenocarcinoma and normal colorectal tissue, may mediate its effects on cellular proliferation and apoptosis [17-20]. The above observations suggest that adiponectin may act directly on CRC cells and/or indirectly by regulating whole-body insulin sensitivity [13].

Adiponectin has been reported to be negatively associated with prostate [21,22], endometrial [23], renal cancer [14] grade and lung cancer stage [24], suggesting its possible involvement in cancer progression. There is paucity of data, however, regarding the relation of the hormone and its receptors with a common type of cancer, namely, CRC, and the clinicopathological implications. Specifically, a correlation of low adiponectin levels with increasing stage of nonmetastatic CRC has been reported in one study [25] and a negative correlation with CRC stage in another [26], but no statistically significant difference of adiponectin levels between early and metastatic stage of CRC was confirmed [27]. An inverse relation of AdipoR1 and AdipoR2 expression levels with unfavorable T stage and grade of CRC has been found [17]. Additionally, higher expression of both adiponectin receptors in adenocarcinoma compared with normal colorectal tissue has been noted [28], not confirmed, however, by Yoneda et al. [20]. No prior study has simultaneously evaluated circulating adiponectin and expression of its receptors in relation to CRC risk and clinicopathological characteristics, in both cases and controls, controlling for known confounding factors.

We have conducted a case-control study comprising 104 CRC patients and 208 age- and sex-matched controls, aiming to simultaneously explore the association of serum adiponectin levels and expression of adiponectin receptors with CRC risk and clinicopathological characteristics, notably tumor grade, stage, metastatic status, tumor location, and size.

2. Methods

During a 5-year period (2002-2007), 104 histologically confirmed, incident cases of CRC, diagnosed according to C18, C19, and C20 codes of the *International Classification of Diseases, 10th Revision* [29], in 6 hospitals in Athens, Greece, were recruited in the study. Study subjects had received no prior cancer treatment and had never been diagnosed with any other type of cancer, familial adenomatous polyposis, or inflammatory bowel disease.

Surgically resected cancer specimens were postoperatively examined by an expert pathologist at each collaborating hospital, blinded as to study design and focus. Colorectal cancer stage was assessed according to the sixth edition of American Joint Committee on Cancer (tumor, node, metastasis [TNM]) stage classification system [30]. Differentiation and lymphovascular invasion status were assessed; and grade was classified as well, moderately, and poorly differentiated adenocarcinoma. Nodal stage was assessed through pathological examination of surgically resected regional lymph nodes and classified according to detection of cancer cells in none (N0), 1 to 3 (N1), and at least 4 (N2) regional lymph nodes, respectively. Finally, the maximum diameter was used as measure of tumor size of the primary tumor; and *splenic flexure* was defined as the anatomical point for the distinction between proximal and distal location of the tumor.

Eligible controls were subjects presenting to the same hospitals for routine health screening and who had a negative self-reported medical history for cancer, hepatic disease, major hormonal or hematological disorders, asthma, autoimmune disease, HIV infection, advanced heart failure, recent myocardial infarction, stroke, chronic kidney failure, nephrotic syndrome, acute pancreatitis, bone fracture, benign neoplasms, or other disorders of the gastrointestinal tract, as previously described [14,24]. Age- (±5 years) and sex-matched controls were enrolled with a 1:2 case-control ratio. Four potential controls refused or were unable to collaborate and were properly substituted for a total of 208 control subjects.

During an approximately 30-minute in-person interview, conducted by trained interviewers (SG, IM) with the use of a precoded questionnaire, each subject provided information pertaining to demographic, anthropometric, lifestyle, and medical history variables. Anthropometrics were measured with standard techniques, as previously described [14,15,24]. Self-reported weight 2 months before diagnosis was also recorded.

Overnight fast venous blood samples were taken from all cases and controls (no later than 9:00 AM) for hormonal measurements and determination of glucose levels. All coded samples were centrifuged; and sera were stored in deep freezers at –70°C before being air shipped with dry ice — blinded as to case-control status — to the Beth Israel Deaconess Medical Center in Boston, MA. Adiponectin levels were measured in 1 batch by trained technicians using a radioimmunoassay procedure. The sensitivity of the assay was 2 ng/mL, and the intraassay coefficient of variation was less than 10 [14,15,24]. Average preservation time was similar for cases and controls, although adiponectin levels do not systematically change with storage time.

Surgically derived colorectal tissue paraffin blocks were provided for 75 of 104 patients. Sections out of these blocks generated 71 formalin-fixed paraffin-embedded colorectal adenocarcinoma specimens and 51 nontumor colorectal specimens. Forty-seven of the tumor specimens were matched to nontumor ones from the same patient. Immunohistochemical methods for the determination of adiponectin receptors AdipoR1 and AdipoR2 have been previously described [28]. The primary antibodies used were the rabbit anti-human AdipoR1 (raised against amino acid residues 357-375) antiserum and the rabbit anti-human AdipoR2 (raised against amino acid residues 374-386) antiserum (both from Phoenix Pharmaceuticals, Belmont, CA) used at 1:500 and 1:200 dilution, respectively. The secondary antibody was a biotinylated anti-rabbit antibody (1:400 dilution) and was applied for 30 minutes at room temperature, followed by the Vectastain Elite ABC Reagent (Vector Laboratories, Burlingame, CA) for 30 minutes. The peroxidase reaction was visualized with the chromogen diaminobenzidine. Slides from normal striated muscle for AdipoR1 and from liver for AdipoR2 served as positive controls. Slides treated with the primary antibody, being replaced by buffer solution without any antibody admixture, served as negative controls. Membranous as well as cytoplasmic stain intensity and extension were evaluated semiquantitatively by an expert pathologist on a scale of 0 (any stain intensity in <5% of tissue), + (weak staining in 5%-100% or strong staining in 5%-50% of tissue), and ++ (strong staining in >50% of tissue).

No part of this article has been previously reported. The study protocol was approved by the University of Athens Medical School Ethics Committee, study design and performance were in accordance with the Helsinki Declaration of 1975, and participants provided written informed consent.

2.1. *Statistical analysis*

Descriptive values (mean, standard deviation [SD], percentiles) of demographic, anthropometric, and lifestyle variables; diabetes mellitus status; and hormonal levels were calculated among cases and controls; and differences were observed through χ^2 test or *t* test. To study the association of adiponectin levels with CRC risk, we modeled the data through multiple conditional logistic regression using case-control status as the outcome variable and adiponectin (in increments of 1 SD of the hormone among controls) as the main predictor variable of interest. Covariates were BMI at the time of diagnosis (in 2 kg/m^2 increments), WHR (in quintiles), weight change in the 2 months before diagnosis, education, alcohol consumption (glasses per month), smoking (cigarettes smoked daily × years of smoking), physical exercise (minutes per day), plant food and red meat consumption (portions per month in quintiles), as well as diabetes mellitus status (self-reported history of diabetes mellitus or fasting glucose level ≥126 mg/dL). Subsequently, mean adiponectin levels by disease stage and tumor characteristics were calculated; and possible associations were investigated through multiple logistic regression analysis, adjusting for age, sex, BMI at diagnosis, WHR, and weight change during the last 2 months. Immunohistochemical expression of adiponectin receptors among patients providing tumor and nontumor colorectal tissue specimens was analyzed by tissue status using Fisher exact test. Finally, multiple logistic regression analysis was used to test the association of immunohistochemical expression of AdipoR1 and AdipoR2 by disease stage and tumor characteristics, controlling for age, sex, BMI at diagnosis, WHR, weight change during the last 2 months, and adiponectin. The SAS statistical package

Table 1 – Distribution of 104 CRC cases and 208 age- and sex-matched controls by demographic, anthropometric, and lifestyle variables; diabetes mellitus status; and serum adiponectin levels

Variable	Cases		Controls		*P* value
	N	%	N	%	
Education (y)					.67[a]
<6	20	19.2	37	17.8	
6-11	59	56.7	111	53.4	
≥12	25	24.1	60	28.8	
BMI at diagnosis (kg/m^2)					<.0001[a]
<25.0	38	36.6	40	19.2	
25.0-26.9	25	24.0	48	23.1	
27.0-28.9	26	25.0	43	20.7	
29+	15	14.4	77	37.0	
Weight change during the last 2 mo (kg)					<.0001[a]
5+ Weight gain	0	0.0	7	3.4	
1-5 Weight gain	3	2.9	42	20.2	
0	47	45.2	114	54.8	
1-5 Weight loss	36	34.6	44	21.1	
5+ Weight loss	18	17.3	1	0.5	
Alcohol consumption (glasses/mo)					.66[a]
0	35	33.7	46	22.1	
1-30	38	36.5	117	56.3	
31-60	13	12.5	26	12.5	
61+	18	17.3	19	9.1	
Smoking (cigarettes smoked daily × years smoking)					.79[a]
0	53	50.9	100	48.1	
1-649	19	18.3	53	25.5	
650-1399	18	17.3	31	14.9	
≥1400	14	13.5	24	11.5	
Physical exercise (min/d)					.02[a]
0	29	27.9	37	17.8	
1-29	33	31.7	53	25.5	
30-59	12	11.5	44	21.2	
60+	30	28.9	74	35.5	
Diabetes mellitus					.03[b]
No	75	72.1	172	82.7	
Yes	29	27.9	36	17.3	
	Mean	SD	Mean	SD	
WHR	101.44	14.74	94.50	9.17	<.0001[c]
Plant food consumption (portions/mo)	68.80	52.87	74.00	44.11	.39[c]
Red meat consumption (portions/mo)	8.54	7.56	5.30	4.17	<.0001[c]
Adiponectin (μg/mL)	9.45	6.14	10.38	3.21	.15[c]

[a] *P* value derived from χ^2 for trend.
[b] *P* value derived from χ^2 for contrast.
[c] *P* value derived from *t* test.

(Version 9.1; SAS Institute, Cary, NC) was used in all analyses, and the level of statistical significance was set at .05.

3. Results

Data concerning demographic, anthropometric, and lifestyle variables; diabetes mellitus status; and adiponectin levels for the 104 CRC patients and 208 age- and sex-matched controls are shown in Table 1. Men comprised 62.5% of the matched data set; and the mean age was 69.8 and 69.1 years among cases and controls, respectively. These data serve mostly descriptive purposes and are not directly interpretable because of mutual confounding. They confirm, however, the established positive association of CRC with WHR, red meat consumption, and diabetes mellitus. Besides, patients with CRC used to exercise less and, as expected in case-control studies, had lower BMI when compared with their respective controls, which was in accordance with a more profound weight loss during the 2 months before presentation.

Table 2 presents the odds ratios (ORs) for CRC by adiponectin increment, derived from multiple logistic regression models in which covariates studied were successively introduced. Thus, the crude OR for adiponectin was 0.83 (P = .06) and changed to 0.72 (P = .04) after adjustment for potential confounders. As expected, WHR and recent weight loss were positively related to CRC risk, whereas physical exercise had an inverse association with CRC and so did BMI at diagnosis. Regarding food items, plant food consumption showed a protective effect, whereas red meat consumption was associated with increased risk. No profound association was found for education, alcohol consumption, smoking, and diabetes mellitus in this data set. Of note, among controls, there was a negative correlation of adiponectin with both BMI and WHR (data not shown).

In Table 3, an inverse association of adiponectin levels with less well-differentiated disease grade was noted (P = .05), whereas the associations with other disease characteristics, namely, TNM stage, tumor location (proximal or distal colon), or tumor size quartiles, were far from reaching statistical significance.

Immunohistochemical expression of adiponectin receptors (AdipoR1 and AdipoR2) among 71 tumor and 51 nontumor colorectal tissue specimens is shown in Table 4. The proportions of both adiponectin receptors in tumor vs nontumor specimens were statistically significantly different (P = .001). Specifically (Fig. 1), expression of adiponectin receptors was

Table 2 – Conditional logistic regression-derived ORs and 95% confidence intervals (95% CIs) for CRC by serum adiponectin levels, controlling for anthropometric, demographic, and lifestyle variables as well as diabetes mellitus

Variable	Category or increment	ORs [a] (95% CIs) P value	ORs [b] (95% CIs) P value	ORs [c] (95% CIs) P value	ORs [d] (95% CIs) P value	ORs [e] (95% CIs) P value	ORs [f] (95% CIs) P value
Adiponectin	1 SD among controls	0.83 (0.68-1.01) .06	0.72 (0.58-0.90) .004	0.72 (0.57-0.89) .003	0.71 (0.56-0.91) .007	0.69 (0.51-0.93) .01	0.72 (0.53-0.99) .04
BMI at diagnosis	2 kg/m^2		0.56 (0.44-0.72) <.0001	0.46 (0.34-0.62) <.0001	0.47 (0.33-0.66) <.0001	0.44 (0.30-0.65) <.0001	0.44 (0.29-0.65) <.0001
WHR	1 Quintile more			1.73 (1.38-2.18) <.0001	1.73 (1.35-2.23) <.0001	1.67 (1.26-2.21) .0004	1.64 (1.23-2.18) .0007
Weight change	1 Level more				4.00 (2.37-6.77) <.0001	5.27 (2.79-9.98) <.0001	5.21 (2.75-9.85) <.0001
Education	1 Level more					1.03 (0.77-1.38) .87	1.06 (0.79-1.43) .70
Alcohol consumption	1 Level more					1.04 (0.70-1.55) .83	1.08 (0.73-1.61) .70
Smoking	1 Level more					0.94 (0.64-1.37) .73	0.87 (0.58-1.31) .51
Physical exercise	30 min/d more					0.65 (0.46-0.90) .01	0.61 (0.43-0.87) .007
Plant food consumption (portions/mo)	1 Quintile more					0.77 (0.60-1.00) .05	0.77 (0.60-1.00) .05
Red meat consumption (portions/mo)	1 Quintile more					1.34 (1.03-1.74) .03	1.36 (1.04-1.78) .03
Diabetes mellitus	Yes vs no						1.76 (0.72-4.31) .22

[a] Model 1: unadjusted OR for adiponectin.
[b] Model 2: odds ratio for mutually adjusted adiponectin and BMI at diagnosis.
[c] Model 3: odds ratio for mutually adjusted adiponectin, BMI at diagnosis, and WHR.
[d] Model 4: odds ratio for mutually adjusted adiponectin, BMI at diagnosis, WHR, and weight change during the last 2 months.
[e] Model 5: odds ratio for mutually adjusted adiponectin, BMI at diagnosis, WHR, weight change during the last 2 months, education, alcohol consumption, smoking, physical exercise, plant food consumption, and red meat consumption.
[f] Model 6: odds ratio for mutually adjusted adiponectin, BMI at diagnosis, WHR, weight change during the last 2 months, education, alcohol consumption, smoking, physical exercise, plant food consumption, red meat consumption, and diabetes mellitus.

Table 3 – Mean values and SDs of adiponectin levels by disease stage and tumor characteristics of 104 CRC patients along with P values derived from multiple logistic regression analysis, controlling for age, sex, BMI at diagnosis, WHR, and weight change during the last 2 months

Variable	n	%	Adiponectin		
			Mean	SD	P value
TNM stage					.65
O, I	19	18.3	8.8	7.39	
II	31	29.8	9.2	5.46	
III	26	25.0	9.4	6.18	
IV	28	26.9	10.3	6.14	
Grade					.05
Well differentiated	8	7.7	11.8	11.06	
Moderately differentiated	80	76.9	9.7	5.88	
Poorly differentiated	16	15.4	7.2	3.36	
Tumor location					.48
Proximal colon	22	21.2	11.0	7.93	
Distal colon	82	78.8	9.0	5.55	
Tumor size, quartiles (cm)[a]					.22
1st	19	21.8	11.2	8.41	
2nd	22	25.4	7.8	4.25	
3rd	21	24.1	9.6	6.03	
4th	25	28.7	8.8	5.70	

[a] Tumor size was available for 87 of 104 cases.

higher in cancerous than in normal tissue (27% vs 6% showed a strong positive staining for AdipoR1 and 37% vs 4% for AdipoR2). Membranous as well as cytoplasmic localization of adiponectin receptors was noted in both tumor and nontumor specimens. Matched analysis of the restricted data set of 47 patients with both tumor and nontumor tissue available practically yielded the same results (data not shown).

Distribution of the immunohistochemical expression of adiponectin receptors among 71 adenocarcinoma specimens by disease stage and tumor characteristics is presented in Table 5. Multivariate analysis–derived results showed overall that there was a statistically significantly higher expression of AdipoR2 in advanced TNM stage (P = .01). When the 3 components of TNM staging were examined separately, however, a negative association of AdipoR1 expression with N stage (P = .03) and a positive association of AdipoR2 immunoreactivity with T stage (P = .004) emerged, whereas expression of both adiponectin receptors was not related to metastatic status, grade, tumor location, or size (T and N stages were available for 70 of 71 tumor specimens; data not shown). The numbers were, however, small to allow firm conclusions.

4. Discussion

The present case-control study confirms the inverse association of serum adiponectin levels with CRC risk and, for the first time, shows an additional inverse association with advanced tumor grade. Immunohistochemically, the stronger expression of both AdipoR1 and AdipoR2 receptors was also confirmed in adenocarcinoma compared with normal colorectal tissue adjacent to the tumor of the same individuals. Moreover, an inverse association of AdipoR1 expression with regional lymph node metastasis (N stage) and an enhancement of AdipoR2 expression with advanced TNM stage were noted for the first time, despite the limited power of the study, whereas AdipoR2 immunoreactivity was also positively related with T stage.

A protective role of adiponectin has been proposed for several malignancies, such as endometrial [23], lung [24], breast [16], and renal cancer [14]. In line with the majority of prospective and case-control studies [26,31-35], we have found a protective role of lower plasma adiponectin levels concerning CRC, which is approximately of the similar size of effect. Two investigations [36,37] without adjustment for major confounding factors reported no relation between adiponectin and CRC risk, whereas another study noted a positive association for leptin-adiponectin ratio, but not for adiponectin [38].

It has been proposed that adiponectin exerts its protective role either directly on cancer cells or indirectly by regulating whole-body insulin sensitivity. Thus, obesity, a condition associated with reduced levels of this adipokine, is considered to lead to chronic hyperinsulinemia and increased levels of bioavailable insulin-like growth factor–I and insulin-like growth factor–II, both promoting cellular proliferation and inhibiting apoptosis [4,13,39,40]. The hypothesis of a direct antiproliferative and proapoptotic role of adiponectin is supported by the expression of its receptors in several cancer cell types [13]. Adiponectin has been shown to inhibit CRC cell growth via activation of adenosine monophosphate–activated protein

Table 4 – Distribution of 71 tumor and 51 nontumor colorectal tissue specimens by AdipoR1 and AdipoR2 immunohistochemical expression

Variable	Tumor tissue		Nontumor tissue		P value[a]
	n	%	n	%	
AdipoR1 expression					.001
None (0)	22	31.0	10	19.6	
Positive (+)	30	42.2	38	74.5	
Strong positive (++)	19	26.8	3	5.9	
AdipoR2 expression					.001
None (0)	15	21.1	22	43.1	
Positive (+)	30	42.3	27	53.0	
Strong positive (++)	26	36.6	2	3.9	

[a] P values derived from Fisher exact test.

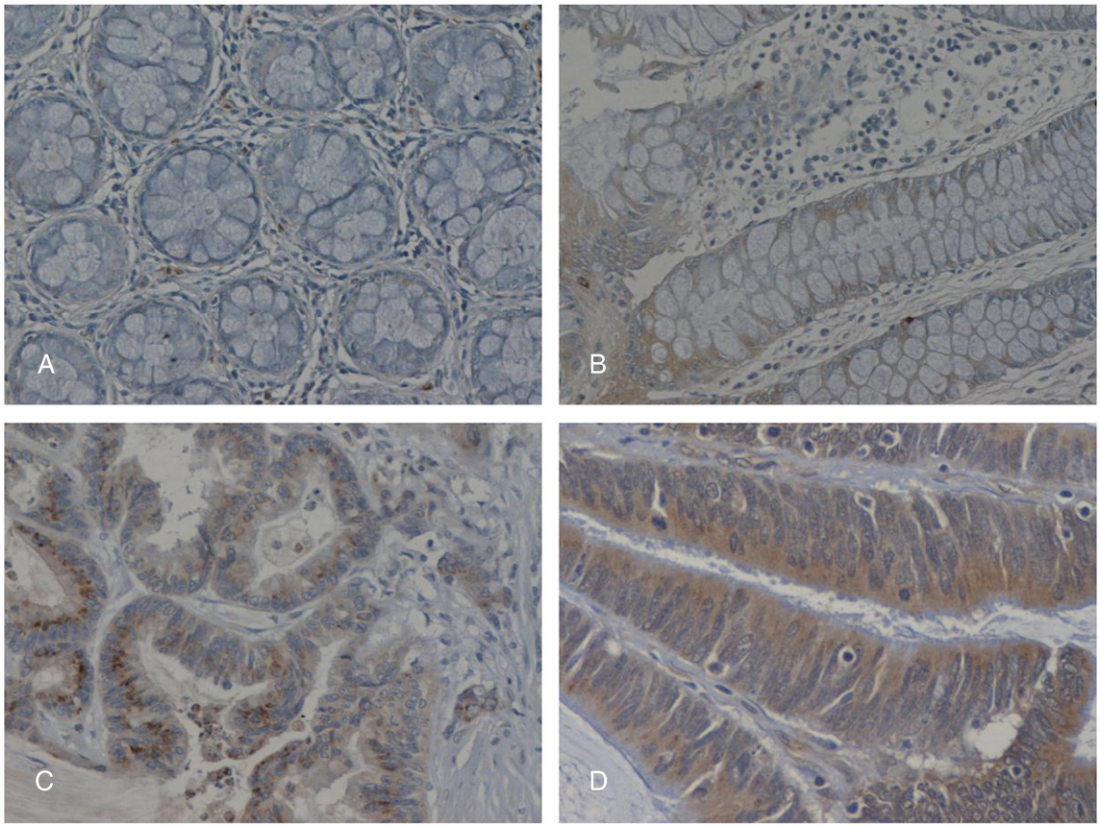

Fig. 1 – Immunohistochemical expression of adiponectin receptors AdipoR1 (A and C) and AdipoR2 (B and D) in nontumor (A and B) and tumor (C and D) colorectal specimens.

kinase, thereby downregulating the mammalian target of rapamycin pathway [19]. In glucose-deprived medium, however, adiponectin was recently shown to support CRC cell survival, with an increase in AdipoR1 and AdipoR2 expression, through enhancement of autophagic response [41].

To our knowledge, this is the first study to suggest an inverse relation of adiponectin with CRC grade. The same observation has been reported for prostate [21,22] and endometrial cancer [23], along with a borderline statistically significant association for renal cancer [14]. Regarding other CRC characteristics, no association was found in the present investigation. Two prior studies showed an inverse relation of adiponectin with CRC stage [25,26], but the finding was not confirmed in a third one [27]. Power limitations may be an underlying factor for these discrepancies. The topic is of interest, however; and inverse associations have already been reported for other cancer types by stage of disease, notably lung cancer [24]. Prospective studies are also needed to define whether low circulating adiponectin levels are the cause or the effect of poor colorectal adenocarcinoma differentiation.

Table 5 – Distribution of 71 AdipoR1 and AdipoR2 immunohistochemical expression by TNM stage and grade along with P values derived from multiple logistic regression analysis, controlling for age, sex, BMI at diagnosis, WHR, weight change during the last 2 months, and adiponectin

Variable	AdipoR1 expression				AdipoR2 expression			
	None (0) n (%)	Positive (+) n (%)	Strong positive (++) n (%)	P value	None (0) n (%)	Positive (+) n (%)	Strong positive (++) n (%)	P value
TNM stage				.31				.01
O, I	4 (18.2)	5 (16.7)	4 (21.1)		6 (40.1)	4 (13.3)	3 (11.5)	
II	4 (18.2)	13 (43.3)	8 (42.0)		5 (33.3)	10 (33.3)	10 (38.5)	
III	7 (31.8)	7 (23.3)	4 (21.1)		2 (13.3)	10 (33.3)	6 (23.1)	
IV	7 (31.8)	5 (16.7)	3 (15.8)		2 (13.3)	6 (20.1)	7 (26.9)	
Grade				.12				.25
Well differentiated	2 (9.1)	3 (10.0)	1 (5.3)		1 (6.7)	4 (13.3)	1 (3.8)	
Moderately differentiated	13 (59.1)	24 (80.0)	16 (84.2)		11 (73.3)	24 (80.0)	18 (69.3)	
Poorly differentiated	7 (31.8)	3 (10.0)	2 (10.5)		3 (20.0)	2 (6.7)	7 (26.9)	

Both cancerous and normal colorectal specimens showed a membranous as well as cytoplasmic localization of adiponectin receptors; similarly to previously studies [28,42], stronger immunoreactivity was observed in colorectal adenocarcinoma than in epithelial cells lining the adjacent normal mucosa. Yoneda et al [20] reported analogous results, though without reference to any significant difference between cancerous and healthy colorectal tissue. Apart from CRC, 2 other gastrointestinal malignancies, namely, pancreatic [43] and gastric [44] cancer, have been shown to express both adiponectin receptors, and a stronger expression in cancerous compared with normal gastric epithelium has been reported. We have previously reported the expression of adiponectin receptors in cancerous but not in normal lung tissue [24], whereas a stronger expression of AdipoR1 and AdipoR2 in chondrosarcoma in comparison with normal cartilage tissue, both in chondrosarcoma patients and cell lines, was found [45].

Although the different histological features might explain the overexpression of adiponectin receptors in adenocarcinoma in comparison with normal colorectal tissue, an upregulation of AdipoR1 and AdipoR2 expression, induced by lower serum adiponectin levels or differences in obesity status, cannot be excluded. However, expression of adiponectin receptors was related neither to circulating adiponectin nor to anthropometric variables in the study herein (data not shown).

In multivariate analysis, a statistically significant positive association of AdipoR2 expression with TNM stage as well as decreased AdipoR1 immunoreactivity in tumor specimens derived from patients with lymph node metastasis were noted. Byeon et al [17] reported an inverse relation of CRC T stage and grade with the immunohistochemical expression of both receptor types, without adjusting, however, for possible confounding factors, such as anthropometric characteristics and serum adiponectin levels.

Increased BMI, WHR, or simply waist circumference has been linked to an increased risk of CRC [3,6,7]. Most patients with gastrointestinal cancer experience—upon diagnosis—weight loss or cachexia [46], which is why BMI measurements among CRC patients were on average lower than those among controls, as also indicated by the profound weight loss of cases during the last 2 months before diagnosis. By contrast, as expected, a sizeable and statistically significant association of central obesity with the disease was evident in all alternative models of the analysis.

Apart from the negative association of adiponectin levels with BMI and WHR among controls in our study, as previously published [47,48], the protective role of physical exercise and the adverse effect of red meat consumption were confirmed, whereas diabetes mellitus was more prevalent among CRC patients [49]. Plant food consumption was negatively associated with CRC risk, although literature is currently inconclusive [49,50]. A recent meta-analysis implicates both alcohol consumption and smoking as CRC risk factors [49], but our results are rather showing a null effect. This may be due to drinking patterns and high smoking prevalence in the Greek population or, alternatively, to potential recall bias.

Strengths of the study include inclusion of genetically homogeneous population and use of state-of-the-art techniques for laboratory measurements, which were performed by personnel blinded to the study hypothesis with analyses of samples performed under code. The process does not exclude the possibility of random error. Even if this were the case, however, random error may impact negatively on an effect estimate, but does not influence the generation of the statistically significant associations reported herein. Multivariate analysis has been used to control for possible confounding factors, yet the results should be interpreted with caution because of power limitations and the possibility of chance findings due to multiple comparisons.

Moreover, the case-control design of the study does not allow but stipulations on the etiological sequence among central obesity, plasma adiponectin concentrations, expression of adiponectin receptors, and CRC risk or the relation with clinicopathological characteristics of the disease. Lastly, BMI at diagnosis or other covariates in the study may have been influenced by the significant weight loss due to cancer cachexia or other pathophysiological mechanisms on account of the disease process, making it difficult to disentangle a possible role in the disease etiology in contrast to a disease-associated condition. Thus, prospective and possibly interventional studies, incorporating the time sequence criterion for causality, are needed.

In this first study that simultaneously explores the association of adiponectin and its receptors with the etiology as well as multiple disease characteristics, there is a suggestion for a negative association of serum adiponectin with CRC grade, an attenuated expression of AdipoR1 among patients with lymph node metastasis, and a positive association of AdipoR2 expression with TNM stage of the disease. The above findings along with the inverse relation of serum adiponectin with CRC stage [26], the higher expression of adiponectin receptors in cancerous compared with normal colorectal tissue [28,42], and the increased immunoreactivity with advancing tumor stage and grade [17] are consistent with the hypothesis that adiponectin affects not only the development but also the progression of CRC. Novel, fertile areas of research for the identification of cancer therapeutic targets emerge, ceramide-related pathways being one of them [51]. Indeed, adiponectin has been shown to stimulate a ceramidase activity, associated with AdipoR1 and AdipoR2 receptors, and to enhance ceramide catabolism and formation of its antiapoptotic metabolite, sphingosine-1-phosphate [52], thus raising the hypothesis that this adipokine could eventually serve as a tentative therapeutic target for CRC.

In conclusion, the findings of our case-control study further exemplify the role of an adipose tissue–related hormone, namely, adiponectin, and its receptors, in colorectal carcinogenesis and advancement. Large prospective studies are warranted to confirm these observations, further exploring the potential role of adiponectin and its receptors as therapeutic targets along with their clinicopathological implications in CRC survival.

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Conflicts of Interest

None to declare.

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