

## Serum adipocytokine levels in patients with colorectal cancer

### *Kolorektal kanserli hastalarda serum adipositokin düzeyleri*

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#### ABSTRACT

**Objectives:** Adipose tissue, besides its main function as an energy storage depot, is currently considered an endocrine organ that secretes several self-produced cytokines. Leptin and resistin play an important role in energy homeostasis, glucose, lipid metabolism and regulation of body weight. The aim of the current study was to determine the concentration of leptin and resistin in pre-operational and post-operational periods of patients diagnosed as colorectal cancer.

**Materials and methods:** The body mass index (BMI) and values of leptin and resistin in blood at diagnosis were measured in 12 colorectal cancer patients in pre- and post-operational periods and in 12 age- and sex-matched controls. Serum leptin and resistin concentrations were measured by ELISA method.

**Results:** Decreased leptin ( $1.95 \pm 0.62$  ng/ml) and resistin ( $4.32 \pm 1.83$  ng/ml) levels were found in pre-operational group compared with the control group (leptin:  $6.12 \pm 0.82$  ng/ml; resistin:  $10.75 \pm 1.46$  ng/ml) ( $p < 0.001$ ,  $p < 0.001$ , respectively). Leptin levels were increased in post-operational period ( $3.29 \pm 0.69$  ng/ml) compared with the pre-operational period ( $p < 0.001$ ). However, no difference was found in post-operative resistin levels ( $4.75 \pm 1.33$  ng/ml) compared with pre-operative period ( $p > 0.05$ ).

**Conclusion:** We conclude that serum concentration of leptin and resistin may have a role in patients with colorectal cancers. Further studies are needed to investigate the possible prognostic value of leptin and resistin in clinical practice of patients with colorectal cancers.

**Key words:** Colorectal neoplasm, leptin, resistin serum

#### ÖZET

**Amaç:** Yağ dokusu, bir enerji deposu olan ana işlevinin yanında, bazı sitokinler salgılayarak bir endokrin organ olarak kabul edilir. Leptin ve rezistin, enerji homeostazisinde, glikoz, lipid metabolizmasında ve vücut ağırlığının düzenlenmesinde önemli bir rol oynamaktadır. Bu çalışmanın amacı, kolorektal kanserli hastalarda operasyon öncesi ve operasyon sonrası dönemlerde leptin ve rezistin konsantrasyonlarının belirlenmesidir.

**Gereç ve yöntem:** Toplam 12 kolorektal kanserli hastada ameliyet öncesi ve sonrası dönemde ve yaş ve cinsiyet uyumlu 12 kişiden oluşan kontrol grubunda vücut kitle indeksi (VKİ) ve kan örneklerinde leptin ve rezistin düzeyleri ölçüldü. Serum leptin ve rezistin konsantrasyonları ELISA ile yöntemi ile ölçüldü.

**Bulgular:** Operasyon öncesi grup (leptin:  $1.95 \pm 0.62$  ng/ml; resistin  $4.32 \pm 1.83$  ng/ml) kontrol grubuna (leptin:  $6.12 \pm 0.82$  ng/ml; resistin:  $10.75 \pm 1.46$  ng/ml) göre karşılaştırıldığında leptin ve rezistin düzeylerinde azalma gözlemlendi (sırasıyla,  $p < 0.001$ ,  $p < 0.001$ ). Operasyon sonrası grupta ( $3.29 \pm 0.69$  ng/ml), operasyon öncesi gruba göre leptin düzeylerinde artış olduğu görüldü ( $p < 0.001$ ). Resistin düzeyleri yönünden operasyon sonrası ( $4.75 \pm 1.33$  ng/ml) dönemle operasyon öncesi dönem arasında anlamlı bir farklılık yoktu ( $p > 0.05$ ).

**Sonuç:** Çalışmamızda serum leptin ve rezistin konsantrasyonlarının kolorektal kanserde önemli rol oynayabileceği sonucuna varıldı. Ayrıca leptinin muhtemelen prognostik değeri klinikte faydalı olabilir ancak daha fazla araştırmanın yapılması gereklidir.

**Anahtar kelimeler:** Kolorektal kanser, leptin, rezistin, serum

## INTRODUCTION

Colorectal cancer is a colon disease, which is one of the most frequently observed cancer types in both men and women. It consists 15% of all cancer cases. In colorectal cancer patients, tumors may exist in any region of colon or rectum.<sup>1</sup>

The evidence from recent studies showed that apart from energy storing, adipose tissue also acts as an endocrine organ. Both excessive and deficient adipose tissue amount may cause endocrinologic metabolic complications. Preserving the balance between the levels of adipocytokines secreted from adipose tissue is important.<sup>2</sup> These adipocytokines- especially leptin and resistin are widely investigated. Because levels of these hormones alter at obesity and colorectal cancers usually together with cancer cachexia, evaluation of adipocytokine levels may be important in colorectal cancer studies.<sup>3</sup>

Leptin was thought to be expressed and secreted only by adipocytes, however their production in placenta, gastric, colorectal and mammary epithelial tissue has been documented<sup>4</sup>. Leptin and resistin play an important role in energy homeostasis, glucose, lipid metabolism and regulation of body weight.<sup>5,6</sup> Recent data have also revealed the functions of adipocytokines in immunity, cancer and bone formation<sup>2</sup>. Leptin increases the glucose uptake and glucose cycle in cells independent of insulin. Recent studies indicated that leptin is also associated with growth.<sup>7</sup> In vitro cancer studies demonstrated that leptin has mitogenic effects and increases migration and growth factors. Another study showed that leptin increases the risk of colon cancer by affecting colon cancer cells like a growth factor and stimulating epithelial cell proliferation<sup>8</sup>.

Resistin impairs intracellular glucose uptake stimulated by insulin and increases hepatic glucose synthesis. Serum resistin levels are elevated in obesity. Resistin is thought to be associated with inflammatory processes because it was expressed by human macrophages. Some studies suggest that elevated serum resistin levels are associated with increased cancer risk but there is no sufficient evidence for this suggestion.<sup>9</sup>

The aim of the present study was to determine the concentration of leptin and resistin in pre-operational and post-operational periods of patients diagnosed as colorectal cancer. To our best knowledge, this is the first study in which resistin and leptin

serum concentration was investigated both in pre-operational and post-operational in colorectal cancer patients.

## PATIENTS AND METHODS

The study comprised of three groups. First group was control group, second group was pre-operational and the third group was post-operational. The study population included newly diagnosed 12 adult men patients with colorectal cancer who were treated at the General Surgery Department of the Eskisehir Osmangazi University and control group was comprised of 12 men volunteers who didn't have diabetes and other chronic diseases and compatible with patient group in terms age and gender distribution. The level of body mass index (BMI) was measured in age- and sex-matched all groups.

The study protocol was approved by the Ethics Committee of the Eskisehir Osmangazi University and informed consent was obtained from all participants before their inclusion in the study. Exclusion criteria: treatment by chemotherapy, radiotherapy, or a major operation during the 6 months before recruitment, brain metastasis, acute or chronic infection, congestive heart failure and abnormal chronic obstructive pulmonary disease.

Blood samples were obtained in the morning after 12-h fasting from patients of all examined groups and controls. Blood samples were collected in colorectal cancer patients at the time of diagnosis and at post-operational periods. Serum samples were obtained after 30 min clotting and centrifugation at 1000 g for 15 min at 4°C. Serum was removed and stored frozen at -80°C. Fasting plasma leptin and resistin levels were determined in pre-operational and post-operational periods of patients diagnosed as colorectal cancer and control group. Quantitative parameters of serum resistin levels were measured by ELISA (Enzyme Linked Immunosorbent Assay) using AviBion Human Resistin (Orgenium Laboratories Division, Vantaa, FINLAND) ELISA test kits. Serum leptin levels were measured by ELISA (Enzyme Linked Immunosorbent Assay) using AviBion Human Leptin (Orgenium Laboratories Division, Vantaa, FINLAND) ELISA test kits.

## Statistical Analysis

Statistical analyses were performed using the SPSS 16.0. All variables were checked for normal dis-

tribution by Kolmogorov-Smirnov test. Kruskal-Wallis was applied for multiple comparisons and Mann-Whitney U test for independent two samples. Pre- and post-operative comparisons were done by Wilcoxon test. Data are presented as mean  $\pm$  S.D. Differences were considered significant at  $p < 0.05$ .

## RESULTS

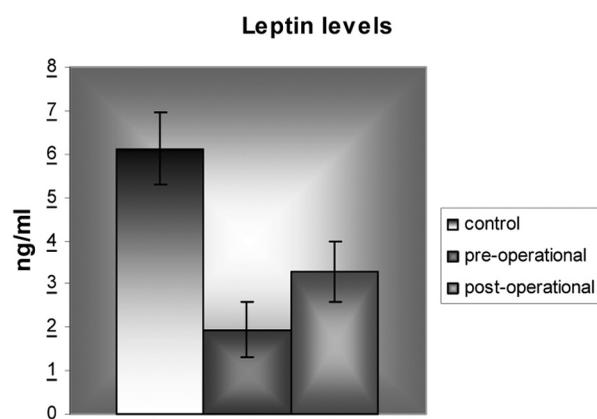
No significant difference was found between mean age of two groups (Table 1). Patients had significantly lower BMI value compared with control subjects ( $p < 0.01$ ) (Table 1).

Our results showed decreased leptin levels in pre-operational group ( $1.95 \pm 0.62$  ng/ml) compared control group ( $6.12 \pm 0.82$  ng/ml) ( $p < 0,001$ ) (Figure 1). The leptin levels was increased in post-operational group ( $3.29 \pm 0.69$  ng/ml) compared in pre-operational group ( $1.95 \pm 0.62$  ng/ml) ( $p < 0,001$ ). The resistin levels was decreased in pre-operational group ( $4.32 \pm 1.83$  ng/ml) compared in control group ( $10.75 \pm 1.46$  ng/ml) ( $p < 0.001$ ) and resistin levels was increased in post-operational group compared in pre-operational group but there was no important significance ( $p > 0.05$ ) (Figure 2). Leptin and resistin levels were decreased in post-operational group compared control group (respectively  $p < 0.001$ ,  $p < 0.001$ ).

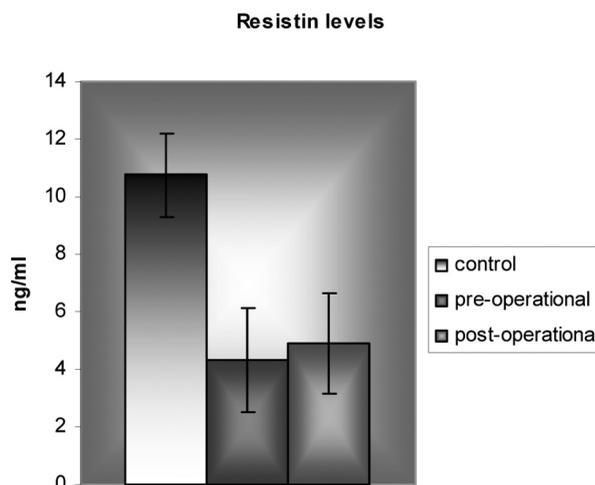
**Table 1.** Clinical features of patient and control groups

	Controls (n=12)	Pre-operation (n=12)	Post-operation (n=12)
Age (Year)	57.0 (53.0-59.0) <sup>a</sup>	60.0 (58.0-62.8)	60.0 (58.0-62.8)
BMI (kg/m <sup>2</sup> )	27.0 $\pm$ 2.4 <sup>b</sup>	21.5 $\pm$ 2.2	21.5 $\pm$ 2.2

BMI: Body mass index; a: not significant; b:  $p < 0.01$  (Controls vs. patients)



**Figure 1.** Serum leptin levels of patient and control groups



**Figure 2.** Serum resistin levels of patient and control groups

## DISCUSSION

Adipose tissue, besides its main function as an energy storage depot, is currently considered an endocrine organ that secretes several self-produced cytokines. These cytokines, which are named adipokines, are proteins with paracrine, autocrine and endocrine function. The three best-studied adipokines are leptin, adiponectin and resistin<sup>10</sup>. Adipokines exert significant effects on metabolism and lipogenesis as well as in regulation of human inflammatory responses.<sup>11</sup>

Adipocytokines were shown to participate to some extent in the process of carcinogenesis, however most if not all of these positive data come from in vitro studies on cancer cell lines. It is well documented that obesity increases the risk of some types of cancer such as that of the colon, breast and prostate. Multiple recent studies have indicated that some of adipose tissue-derived hormones may significantly influence the growth and proliferation of tumorous stroma and malignant cells within.<sup>2</sup> Leptin and resistin play a physiologic role in the reduction of food intake, energy homeostasis, and regulation of body weight. Leptin primarily controls body fat stores and have roles in promoting cellular proliferation, inhibiting cellular apoptosis, and inducing angiogenesis.

Over the years, the association between leptin levels and the risk of colorectal cancer or adenoma has remained controversial. The expression of the leptin receptor in normal human colon mucosa, ad-

enomas, and cancers suggests that a direct effect of leptin may be involved in carcinogenesis.<sup>12</sup> Some authors suggested that low leptin serum concentration can be connected with weight loss observed in cachectic cancer patients.<sup>13</sup> Simons et al. and Wallace et al. suggested that low-serum leptin concentration in their studies could be related to decreased body fat mass which is similar to our results.<sup>14,15</sup> We observed, to our knowledge, for the first time, lower leptin concentration in patients who presented newly diagnosed with colorectal cancer. We also found that highest serum concentration of leptin in post-operational period, than in pre-operational period. Based on our observations, we suspect that weight loss is a causative factor of hypoleptinemia and some unknown mechanisms may be responsible for low serum leptin levels in colorectal cancer patients.

The authors suspect that leptin represents the growth factor for colonic epithelial cells and can stimulate proliferation of the colon mucosa cells.<sup>16</sup> Garofalo et al. suggested that leptin not only stimulates cell growth via ERK1/2 pathway in colorectal cancer but also reduces cell apoptosis.<sup>17</sup> In our study, we observed significantly lower serum leptin concentration in colorectal cancer patients than control group however, higher serum leptin concentration in post-operational group than in pre-operational group.

In the present study, we observed that lowest resistin serum concentration in colorectal cancer patients and highest in control group. In our study, we found higher serum resistin concentration in post-operational group than in control group but it did not observe a statistically significant difference. The adipocytokine resistin has been demonstrated to be involved in inflammatory states corresponding to its predominant expression in mononuclear cells, particularly in atherosclerosis.<sup>18</sup> As for its correlation with cancer, two case-control studies for the risk of MDS or multiple myeloma have been reported.<sup>19</sup> Dalamaga et al. demonstrated a decreased resistin level in MDS patients, and speculated that it was due to a compensatory response to the up-regulation of other inflammatory factors etiologically linked to myelodysplasia. They also reported a decreased level of resistin in patients with multiple myeloma.<sup>20</sup>

In other studies, data also observed higher serum resistin concentration in colorectal cancer

group in patients than control group. These data can explain the highest concentration of resistin in colorectal cancer, which is strongly connected with inflammation.<sup>21</sup> Based on our observations, we suspect that inflammation is not a causative factor of lower resistin levels and some unknown mechanisms may be responsible for low serum resistin in colorectal cancer patients. To date, the role of resistin in colorectal cancer is not fully elucidated yet however further research are necessary.

To our best knowledge, this is the first study in which resistin and leptin serum concentration was investigated both in pre-operational and post-operational in colorectal cancer patients. Based on our data the decrease of serum leptin and resistin concentrations in pre-operational period in colorectal cancer patients, however highest serum leptin and resistin concentrations in post-operational period in colorectal cancer patients may be dependent on decrease in BMI and weight loss. We conclude that serum concentration of leptin and resistin may play an important role in colorectal cancer. We also assume that leptin may possibly have the prognostic value useful in clinical practice. Further studies are needed to investigate the possible prognostic value of leptin and resistin in clinical practice of patients with colorectal cancers.

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#### Conflict of Interest

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