**UNIVERSITY OF MEDICINE AND PHARMACY
"CAROL DAVILA" BUCHAREST
FACULTY OF MEDICINE
DOCTORAL SCHOOL**

# KEY MOLECULES IN THE INITIATION AND PROGRESSION OF COLORECTAL CANCER
**PhD THESIS SUMMARY**

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**SUMMARY**

# I. GENERAL PART - CURRENT STATE OF KNOWLEDGE

Colorectal cancer represents one of the cancers with the highest morbidity and mortality rates among cancer patients, thus representing a major health problem worldwide.

In 2020, worldwide, colorectal cancer ranked third in terms of incidence, and among the Romanian population it is the third most common type of cancer in men and the second most common type of cancer in women. The risk of colorectal cancer is influenced by both environmental and genetic factors.

Despite significant efforts in prevention and early diagnosis, as well as advances in surgical and oncological treatment (radiotherapy and chemotherapy), the prognosis of colorectal cancer remains reserved. Moreover, medical staff frequently face relapses and metastases after performing the surgical intervention. It should also be mentioned the important role of screening programs, with a positive impact on the early diagnosis of colorectal cancer, which have gained momentum in many countries, as well as in Romania.

In the last decades, the scientific community has focused on the identification of biomarkers capable of detecting colorectal cancer in early stages and establishing with the greatest possible precision the prognosis, the response to treatment, as well as the risk of its recurrence and metastasis. Also, these biomarkers could be used to identify an increased susceptibility to colorectal cancer. Moreover, they could also represent some useful tools for the therapy of patients who do not respond to systemic treatment. Once these patients are identified, they could benefit from early active treatment, modification of the therapeutic regimen or the development of a strategic plan to prevent the occurrence of recurrences and metastases.

Improvements in molecular biology techniques as well as epigenetics and transcriptomics studies have led to a better understanding of the pathophysiology of colorectal cancer. In this regard, numerous types of RNA have been identified as potential biomarkers in colorectal cancer. In addition to the diagnostic or prognostic performance of a single biomarker, a combination of different molecules could improve the specificity and sensitivity of diagnostic, prognostic and predictive means.

# II. PERSONAL CONTRIBUTION

# 3. HYPOTHESIS AND GENERAL OBJECTIVES OF THE RESEARCH

The new discoveries regarding the genetic component of the colorectal carcinogenesis process and the involvement of metabolic processes or cellular processes, such as apoptosis and inflammation, give a new vision in the approach of this complex pathological entity. Molecules with the role of predictive and prognostic biomarkers are targeted, as well as the identification of new therapeutic targets with the help of which to develop new therapies with the aim of increasing the hope and quality of life of patients.

Taking into account the previously mentioned aspects, the studies within the present doctoral thesis addressed the following arguments:

1. the need for a better understanding of the molecular mechanisms underlying colorectal carcinogenesis, as well as the dysregulation of various metabolic pathways to identify the most reliable predictive and prognostic factors;

2. the need to identify new therapeutic targets, as well as establishing complementary therapeutic strategies to prevent the establishment of resistance to therapy.

The aim of the studies was to investigate some molecular mechanisms involved in the occurrence and progression of colorectal cancers and to identify new predictive biomarkers and new possible therapeutic targets.

The specific objectives of the studies were the following:

1. evaluating the expression of genes involved in the transport of fatty acids and the expression of microRNAs that target these genes;

2. evaluation of the expression of microRNAs belonging to the let-7 family and their involvement in the regulation of genes in the IGF1 signaling pathway;

3. evaluation of the expression of genes involved in apoptosis and interactions with microRNAs from the let-7 family.

The concept of molecular pathology undergoes dynamic changes with the passage of time, so it is necessary to understand the fine mechanisms involved in the onset, progression and dissemination of colorectal cancer, as well as the development of targeted therapies in accordance with new discoveries. It is necessary to align Romanian research in the field of molecular pathology with international research and develop new research perspectives for the entire scientific community.

# 4. GENERAL RESEARCH METHODOLOGY

In order to achieve the proposed objectives, the activities were oriented in four directions:

**Evaluation of the group of patients with CRC (N=39)**
- histopathological evaluation
- immunohistochemical evaluation
- KRAS, BRAF mutational status
- MSI status

**Study 1: molecules involved in fatty acid transport in CRC**

Evaluation of the expression of genes involved in fatty acid transport (39 CRC patients and control group with 18 patients) and the expression of micro-RNAs targeting these genes (25 CRC patients and control group with 18 patients)

**Study 2: IGF1 pathway and let-7 miR family in CRC**

Evaluation of the expression of let-7 family microRNAs and their involvement in the regulation of genes in the IGF1 signaling pathway (25 CRC patients)

**Study 3: apoptosis and the let-7 miR family in CRC**

Evaluation of the expression of genes involved in apoptosis and interactions with micro-RNAs from the let-7 family (18 patients with CRC and control group with 18 patients)

**Figure 1.** Scheme of the work plan

Patients were selected based on the histopathological diagnosis of colorectal adenocarcinoma. The following characteristics were recorded for each patient: socio-demographic parameters such as age, sex; risk factors such as tobacco or alcohol consumption; the presence of comorbidities; location of the tumor; TNM staging (tumor, lymph nodes, metastases) and biochemical parameters such as hemoglobin, leukocytes, platelets, INR, fibrinogen, albumin, cholesterol, triglycerides, ALT, AST, urea, creatinine, acid, uric, CEA, CA-19-9 and AFP.

The control group was represented by people who were screened for colorectal cancer by colonoscopy from which colonic mucosa was harvested by biopsy. Socio-demographic parameters such as age or gender were recorded for each individual.

The exclusion criteria for the control group were as follows:

1. the presence of colorectal cancer and/or inflammatory bowel diseases,

2. therapy with non-steroidal anti-inflammatory drugs in the last 3 months and

3. therapy with anticoagulant or antiplatelet drugs in the last 3 months.

The evaluation of the study group allowed the histopathological, immunohistochemical, mutational and MSI characterization of the tumor tissues included in the study. The study group was represented by 39 patients diagnosed with colorectal adenocarcinoma. For each tumor we checked the degree of tumor invasion, the presence of perineural and lymphovascular invasion. With the help of immunohistochemical tests we verified the presence of microsatellite instability (MSI) by testing the expression of proteins involved in the repair process of DNA replication errors (MMR). These proteins are mainly represented by: MLH1, MSH2, MSH6 and PMS2. MSI status was also determined by molecular biology and the results obtained by the two methods were compared in order to identify the performance and value for money of the two methods. The mutational status of the KRAS gene was determined by PCR-RFLP for the detection of mutations at codons 12 and 13, and that of the BRAF gene by reverse hybridization PCR for codons 600 and 601.

The present work combined several approaches, the clinical data of the patients being integrated and correlated with the histopathological, immunohistochemical and molecular biology data. In addition, the biostatistical approach allowed the integration of the obtained results with existing information in public databases using bioinformatics analysis. More specifically, they used:

✧ histopathological techniques for the initial evaluation of colorectal tumors, as well as the confirmation of normal tissues,

✧ immunohistochemical techniques for evaluating the protein expression of CD36 and the status of microsatellite instability (MSI),

✧ molecular biology techniques for the detection of KRAS, BRAF mutations, the status of microsatellite instability, as well as the evaluation of gene expression and microRNAs in the tissues included in each individual study,

✋ dedicated biostatistical analysis programs to identify significant differences in case-control studies and correlations between all the results obtained,

✋ bioinformatics techniques for integrating the results obtained with results from the specialized literature and for identifying interactions of the microRNA – mRNA type.

# 5. EVALUATION OF THE EXPRESSION OF GENES INVOLVED IN FATTY ACID TRANSPORT AND MICRORNAS TARGETING THESE GENES

The first study assessed the expression of genes involved in fatty acid transport and the expression of microRNAs that target these genes.

The expression of the genes coding for the molecules CD36, FASN, GPC4, SLC27A3, SLC27A4 and BIRC5 was identified. Based on the bioinformatic analysis, we selected a panel of microRNAs involved in regulating the expression of the analyzed genes: miR-155-5p, miR-16-5p, miR-27a-3p, miR-26b-5p, miR-29a-3p, miR -107 and miR-195-5p. Gene and microRNA expression level in tumor tissue was analyzed for the entire cohort of patients (39 patients) and a subset of patients (25 patients) compared to matched peritumoral tissue or normal colonic mucosa harvested from healthy individuals (18 individuals). We also performed the statistical analysis to identify correlations between the expression level, clinical parameters, histopathological, immunohistochemical characteristics, MSI status and tumor mutational status.

### Results

Out of the total of 39 patients with a histopathological diagnosis of colorectal adenocarcinoma, 18 (46.15%) were male and 21 (53.85%) were female.

Out of the total of 18 healthy individuals, 8 (44.44%) were male and 10 (55.56%) were female.

Regarding tumor location, 61.54% of patients (24 patients) presented with colon adenocarcinoma, 23.08% of patients (9 patients) presented with recto-sigmoid junction adenocarcinoma and 15.38% of patients (6 patients) presented with rectal adenocarcinoma.

Regarding TNM staging of the studied tumors: 15.38% of tumors (6 tumors) had stage T2, 71.79% of tumors (28 tumors) had stage T3 and 12.82% of tumors (5 tumors) had T4 stage (2 stage T4A and 3 stage T4B).

The grade of tumor invasion for the studied patients was: G1 for 15.38% of tumors (6 tumors), G2 for 66.67% of tumors (26 tumors), and G3 for 17.95% of tumors (7 tumors).

Lymphovascular invasion was present in 10 patients (25.64% of patients enrolled in the study), while lymphovascular invasion was absent in 19 patients (74.36% of patients).

Perineural invasion was present in 6 patients (15.38% of patients enrolled in the study), while perineural invasion was absent in 33 patients (84.62% of patients).

Through the IHC method, 4 cases (10.25%) were identified negative for at least two studied markers (MSI-H). The results obtained for the 4 patients with MSI identified by IHC overlapped the results obtained by PCR.

Mutations identified by PCR-RFLP at codons 12 and 13 were present in 17 patients from the group of patients diagnosed with colorectal adenocarcinoma (43.59% of patients).

Regarding BRAF gene mutations, four cases (10.26% of tumors) presented V600E mutation, identified by the reverse PCR-hybridization technique.

Within the group of patients with a histopathological diagnosis of colorectal adenocarcinoma, the immunohistochemical expression of CD36 was evaluated for all 39 pairs of tumor tissue - peritumoral tissue.
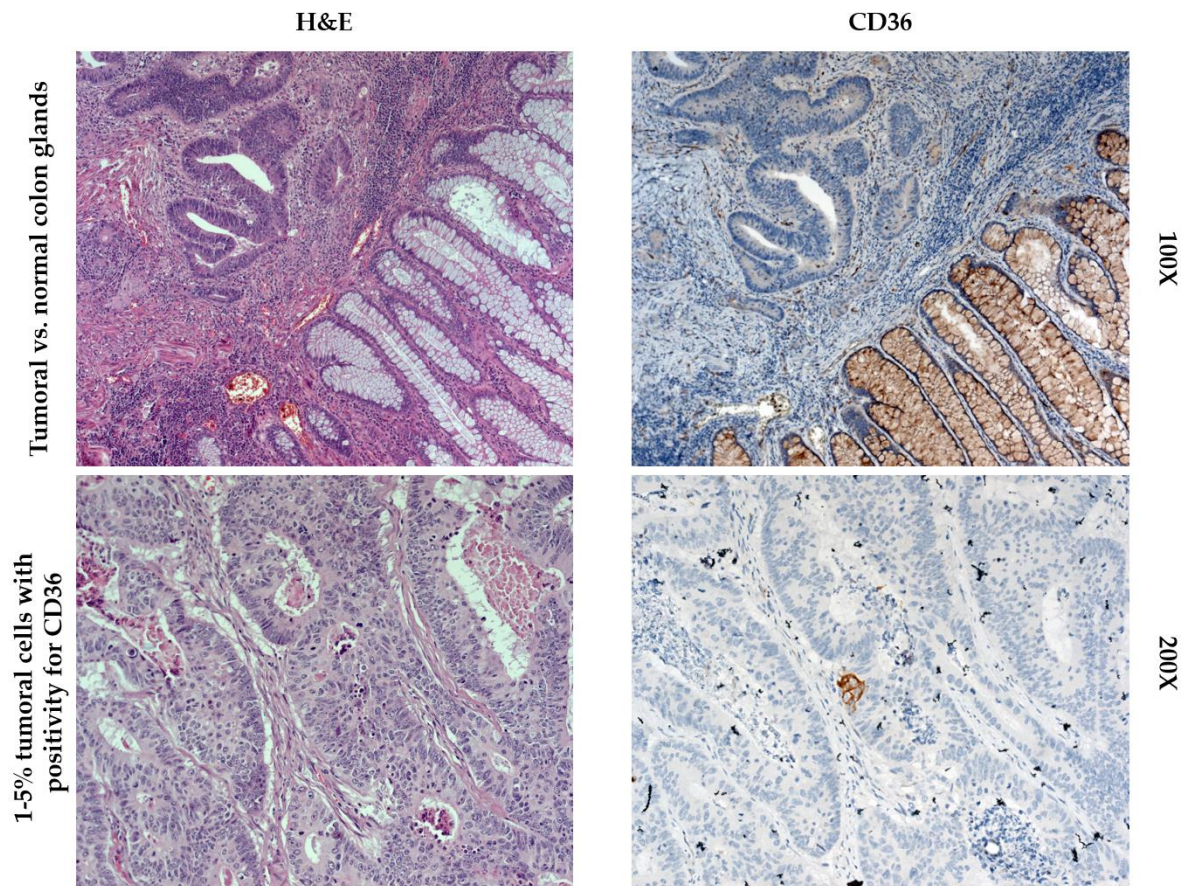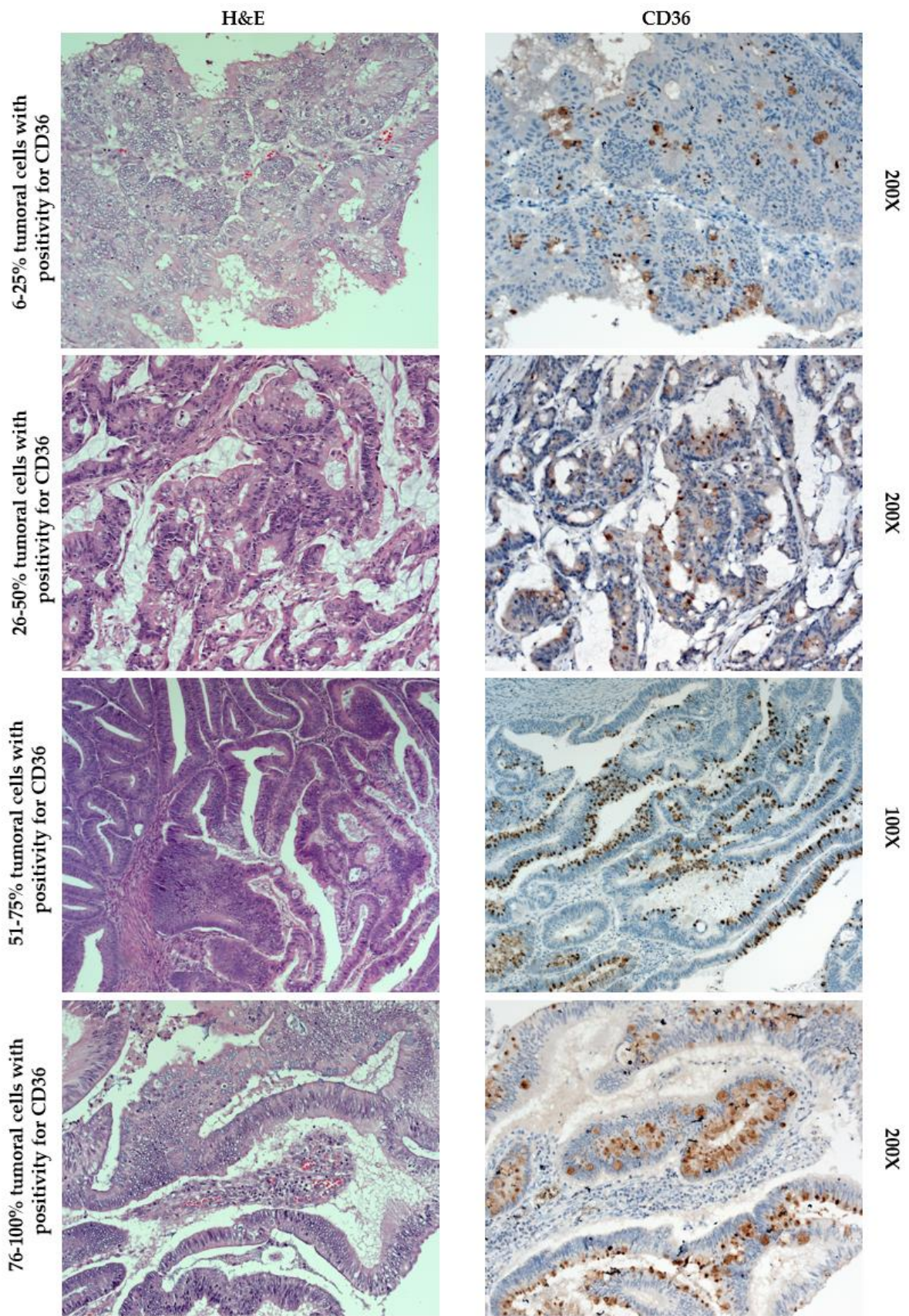
**Figure 2.** Differential immunohistochemical expression of CD36 in tumor and peritumoral tissues

Relative gene expression analysis of tumor tissue versus peritumoral tissue, tumor tissue versus normal colonic mucosa, peritumoral tissue versus normal colonic mucosa is shown in Figure 3.
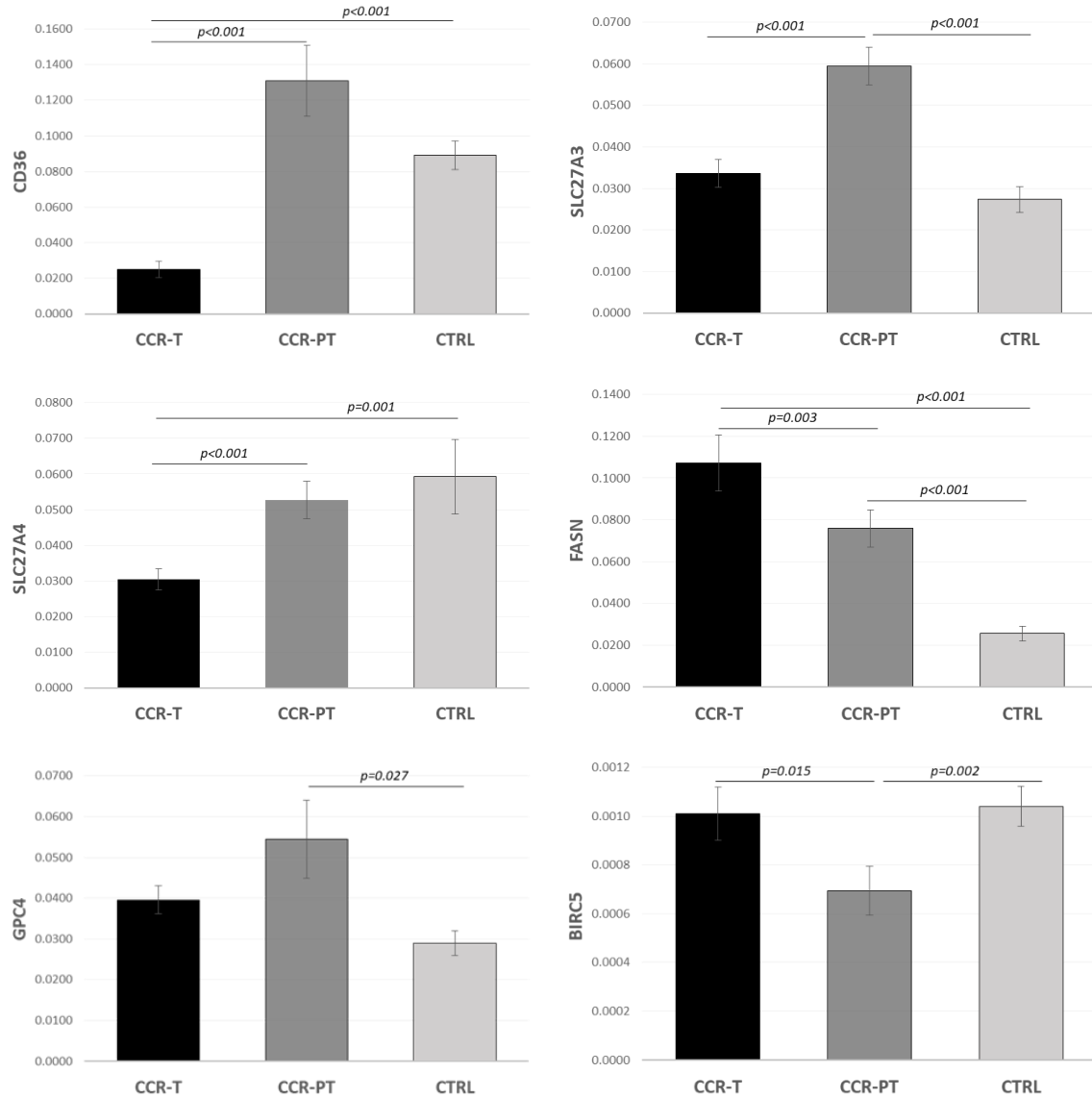


**Figure 3.** Expression of CD36, SLC27A3, SLC27A4, FASN, GPC4 and BIRC5 in tumor, peritumoral and normal tissues. Expression levels are presented as mean values of $2^{-\Delta CT} \pm$ standard error of the mean (SEM).

In silico analysis to identify interactions between microRNAs and messenger RNAs of interest led to seven candidate microRNAs that were subsequently validated using data identified in the literature. Based on these selections, the expression of the following

microRNAs was analyzed: miR-155-5p, miR-16-5p, miR-27a-3p, miR-26b-5p, miR-29a-3p, miR-107 and miR- 195-5 p.

Through molecular biology techniques, we identified a significant impairment of these microRNAs in the patient cohort analyzed as follows: underexpression of miR-16-5p, miR-26b-5p, miR-107 and miR-195-5p in tissues tumor compared to peritumoral tissues and miR-195-5p underexpression and miR-27a-3p overexpression in tumor tissues compared to normal tissues.
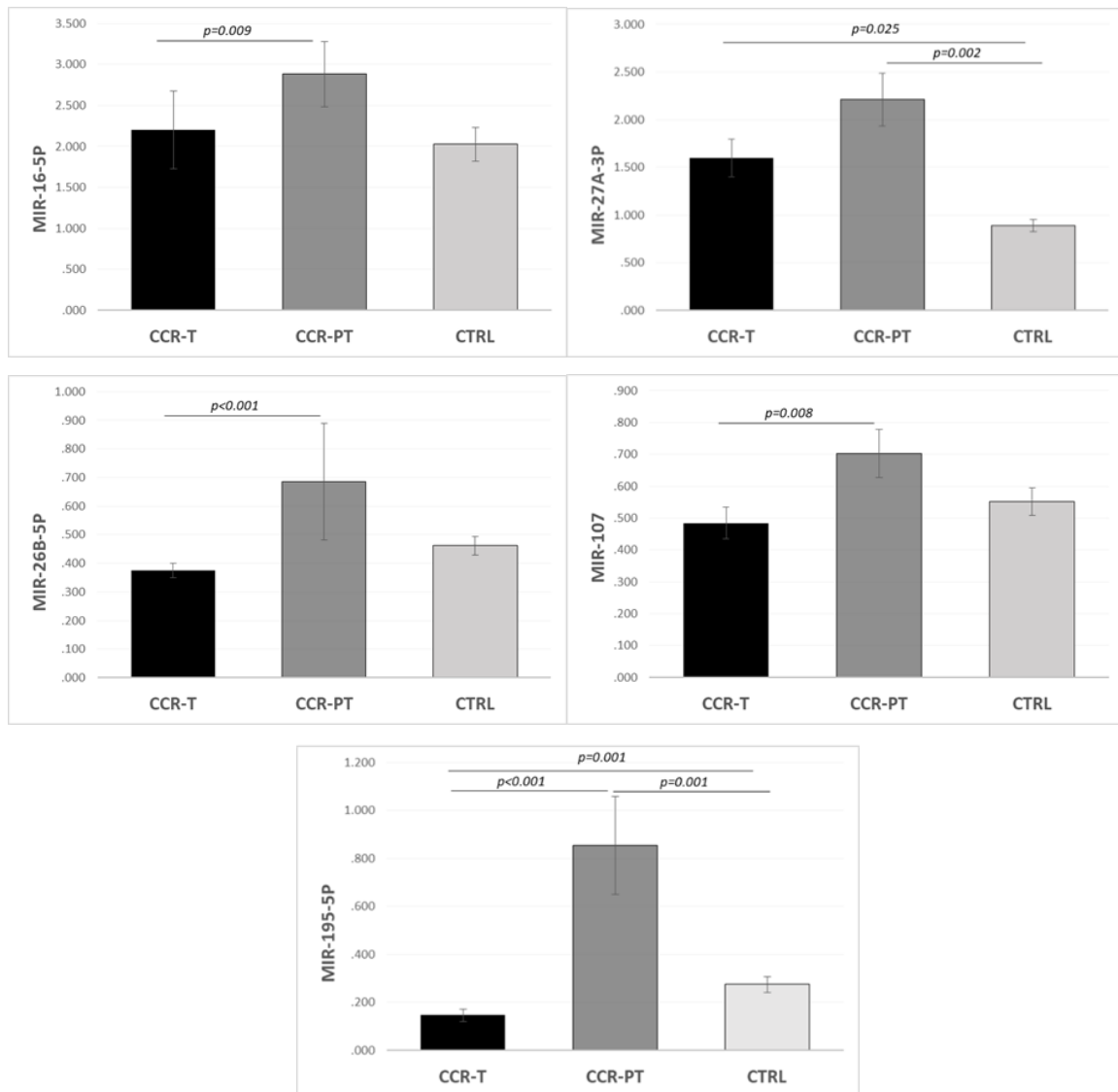


**Figure 4.** Expression of miR-16-5p, miR-27a-3p, miR-26b-5p, miR-107 and miR-195-5p in tumor, peritumoral and normal tissues. Expression levels are presented as mean values of $2^{-\Delta Ct} \pm$ standard error of the mean (SEM).

When we compared the levels of microRNAs in tumor tissues from 11 patients with lymph node metastases (LNI+) versus patients without lymph node involvement, we found that miR-27a-3p was overexpressed (FR = 1.84; p = 0.018) . Also, in the 11 patients with loco-regional lymph node involvement, we identified a negative linear correlation between miR-27a-3p and CD36 expression (p = 0.011, r = -0.730) and a positive correlation between miR-195-5p and FASN (p = 0.023, r = 0.673).

Comparing tumor tissues from the 6 patients with perineural invasion to the 19 patients without invasion we identified miR-27a-3p overexpression (FR = 1.75; p = 0.014). We identified no other differences in the expression of microRNAs and messenger RNAs according to perineural invasion.

# 6. EXPRESSION OF LET-7 FAMILY MICRORNAS: IMPLICATIONS IN COLORECTAL CANCER BY TARGETING THE IGF1 AXIS AND POSSIBLE ASSOCIATION WITH PERINEURAL INVASION

In this study we identified the expression of microRNAs of the let-7 family as the main regulators of genes encoding receptors and substrates involved in the IGF1 signaling pathway. The expression of the following microRNAs was studied: hsa-let-7a-5p, hsa-let-7b-5p, hsa-let-7c-5p, hsa-let-7d-5p, hsa-let-7e-5p, hsa -let-7f -5p, hsa-let-7g-5p and hsa-let-7i-5p.

The study aimed to identify the expression level of microRNAs in the tumor tissue compared to the paired peritumoral tissue. We also performed the statistical analysis to identify correlations between the expression level of microRNAs, clinical parameters and histopathological, immunohistochemical characteristics, MSI and mutational status of the tumors. The analysis was performed on a group of 25 patients with primary colorectal cancer.

### Results

Of the total of 25 patients with a histopathological diagnosis of colorectal adenocarcinoma, 12 (46.15%) were male and 13 (53.85%) were female.

Regarding tumor location, 61.54% of patients (24 patients) presented with colon adenocarcinoma, 23.08% of patients (9 patients) presented with recto-sigmoid junction adenocarcinoma and 15.38% of patients (6 patients) presented with rectal adenocarcinoma.

Regarding the TNM staging of the studied tumors: 12.00% of tumors (3 tumors) were detected in stage T2, 72.00% of tumors (18 tumors) were detected in stage T3 and 16.00% of tumors (4 tumors) were detected at stage T4.

Loco-regional lymph node invasion was absent in 13 cases (52% of patients) and present in 12 cases (48% of patients), 7 patients being stage N1 and 5 patients stage N2.

The grade of tumor differentiation for the studied patients was: G1 for 16.00% of tumors (4 tumors), G2 for 56.00% of tumors (14 tumors), and G3 for 28.00% of tumors (7 tumors).

Lymphovascular invasion was present in 9 patients (36.00% of patients enrolled in the study), while lymphovascular invasion was absent in 16 patients (64.00% of patients).

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Perineural invasion was present in 6 patients (24.00% of patients enrolled in the study), while perineural invasion was absent in 19 patients (76.00% of patients).

Microsatellite instability was present in 2 patients (8.00% of patients enrolled in the study). The results obtained for the 2 patients with MSI identified by IHC overlapped the results obtained by PCR.

Mutations identified by PCR-RFLP at the level of codons 12 and 13 were present in 12 patients from the group of patients diagnosed with colorectal adenocarcinoma (48.00% of patients). Regarding BRAF gene mutations, two cases (8.00% of tumors) showed V600E mutation.

In silico analysis to evaluate the microRNA–target gene interaction between the eight microRNAs belonging to the let-7 family and the genes involved in the IGF1 signaling pathway showed that all eight microRNAs were computationally predicted or experimentally validated as regulators of selected genes except IGF2R and IRS1.

The study of the relative expression of microRNAs between the tumor tissue (T) and the paired peritumoral mucosa (PT) showed that seven of the eight analyzed microRNAs were statistically significantly underexpressed in the tumor tissue: let-7a-5p, let- 7b-5p, let-7c-5p, let-7d-5p, let-7e-5p, let-7f-5p and let-7g-5p. Let-7i-5p was moderately underexpressed in tumor tissue, but without reaching statistical significance.
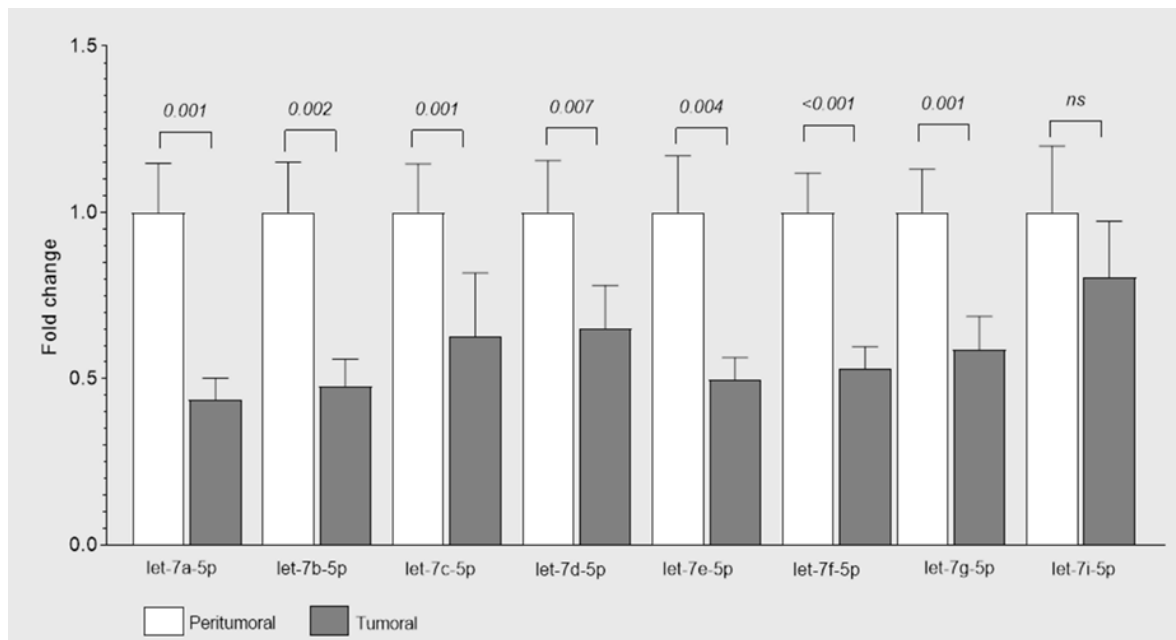


**Figure 5.** Expression levels of let-7 family microRNAs in T and PT tissues from 25 CRC patients. Bars represent mean expression ± SEM. Statistical significance was calculated with the Wilcoxon test; ns = not significant.

Comparing tumor tissues with perineural invasion (IPN+) to those without perineural invasion (IPN-), we identified overexpression in (IPN+) samples of the following microRNAs: let-7a-5p (FC=2.30, p=0.014 ), let-7b-5p (FC=2.87, p=0.006), let-7c-5p (FC=3.65, p=0.011), let-7d-5p (FC=2.71, p= 0.009) and let-7i- 5p (FC=2.63, p=0.036).

The comparative study of let-7 family microRNAs according to the mutational status of KRAS and BRAF genes revealed expression changes in tumor tissues with mutations. Thus in the 12 tumor tissues with KRAS mutations (codon 12 or codon 13) we found that miR-let-7e-5p was overexpressed (FC=1.78, p=0.040) compared to the 13 tumor tissues without KRAS mutations. This increase was also observed comparing the peritumoral tissues of the two groups (FC=2.15; p=0.004) even though none of the PT tissues showed mutations.

# 7. INTERRELATIONSHIPS OF LET-7 FAMILY MICRORNAS AND GENES INVOLVED IN APOPTOSIS AND NF-KB SIGNALING PATHWAY

In this study we examined the expression of eight target genes of microRNAs from the let-7 family involved in apoptosis and the NF-κB signaling pathway. Genes were identified as target by miRWalk analysis and were represented by: BCL2A1, BCL2L1, CASP8, CFLAR, TNFRSF10A, TNFRSF10B, TNFRSF1A and TRAF2.

The study aimed to identify the expression level of selected genes in tumor tissue compared to normal colonic mucosa. We also performed the statistical analysis to identify the correlations between the level of gene expression and the location and histopathological characteristics of the tumors.

### Results

Of the total of 18 patients with a histopathological diagnosis of colorectal adenocarcinoma, 10 (55.56%) were male and 8 (44.44%) were female.

Out of the total of 18 healthy individuals, 8 (44.44%) were male and 10 (55.56%) were female.

Regarding tumor location: 66.67% of patients (12 patients) presented with colon adenocarcinoma, 27.78% of patients (5 patients) presented with recto-sigmoid junction adenocarcinoma and 5.56% of patients (1 patient) presented with rectal adenocarcinoma.

Regarding TNM staging of the studied tumors, 5.56% of tumors (1 tumor) had stage T2, 77.78% of tumors (14 tumors) had stage T3 and 16.67% of tumors (3 tumors) had had stage T4.

The grade of tumor differentiation for the studied patients was: G1 for 11.11% of tumors (2 tumors), G2 for 66.67% of tumors (12 tumors), and G3 for 22.22% of tumors (4 tumors).

The relative gene expression study between tumor tissue (T) and normal colonic mucosa (CTRL) revealed statistically significant overexpression in tumor tissue of all studied genes.
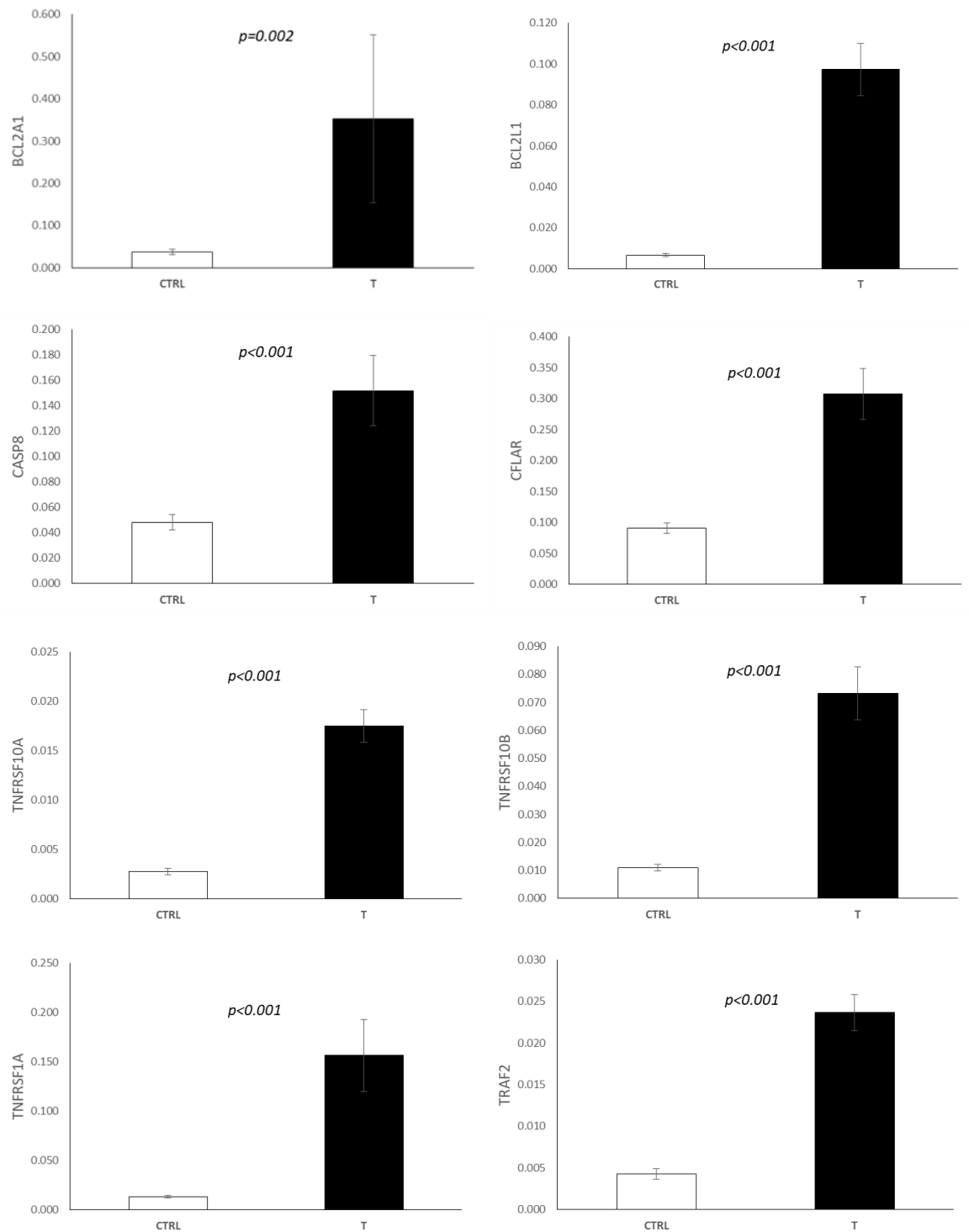
**Figure 6.** Expression level of BCL2A1, BCL2L1, CASP8, CFLAR, TNFRSF10A, TNFRSF10B, TNFRSF1A and TRAF2 in tumor tissues from 18 colorectal cancer patients (CCR) and in normal colonic mucosa from 18 healthy individuals (CTRL). Bars represent mean expression ± SEM. Statistical significance was calculated with the Mann Whitney test

Analyzing the levels of gene expression according to the degree of tumor invasion, we found an underexpression of the BCL2L1 and CASP8 genes in poorly differentiated tumors (G3) compared to moderately differentiated ones (G2). Well-differentiated tumors (G1) did not show statistically significant different expression level.

# 8. CONCLUSIONS AND PERSONAL CONTRIBUTIONS

The main objective of this PhD thesis was to identify molecular mechanisms of colorectal carcinogenesis and new prognostic and predictive biomarkers for targeted therapy in colorectal cancer.

More specifically, the studies targeted three lines of research: fatty acid transport, the IGF1 signaling pathway, and apoptosis.

For each research direction, the obtained results validated the existing results in the specialized literature and highlighted novel elements of the molecular profile of patients with colorectal cancer in Romania.

### 1. Evaluation of the expression of genes involved in fatty acid transport and the expression of microRNAs targeting these genes

In this study we identified a general impairment of the expression of key genes involved in fatty acid metabolism and a set of microRNAs targeting these genes in the colorectal mucosa of patients with colorectal adenocarcinoma compared to adjacent non-tumorous mucosa and normal colonic mucosa.

The expression level of CD36, FASN, GPC4, BIRC5, SLC27A3 and SLC27A4 genes was comparatively analyzed in paired samples collected from 39 patients with colorectal cancer (tumor tissue and peritumoral tissue) and 18 normal tissues collected from the colonic mucosa of some individuals without a diagnosis of colorectal adenocarcinoma.

Seven microRNAs targeting the CD36 gene and most of the analyzed genes were evaluated in 25 patients and all analyzed controls.

A significant impairment of the expression of all analyzed genes, except GPC4, was identified in tumor tissues compared to peritumoral ones. More specifically, we identified underexpression of CD36, SLC27A3, and SLC27A4 and overexpression of FASN and BIRC5. Comparing gene expression between tumor and control tissues, only CD36 and SLC27A4 were underexpressed in tumor tissue, while FASN was overexpressed. No changes were observed in the levels of CD36 and SLC27A4 in peritumoral tissues compared with normal tissue, while SLC27A3, FASN and GPC4 were overexpressed and BIRC5 was underexpressed.

In silico analysis identified seven microRNAs that target CD36: miR-155-5p, miR-16-5p, miR-27a-3p, miR-26b-5p, miR-29a-3p, miR-107 and miR-195 -5 p.

Comparing the expression of these microRNAs between tumor and peritumoral tissues we identified the underexpression of miR-16-5p, miR-26b-5p, miR-107 and miR-195-5p. By comparing the expression of microRNAs between tumor and normal tissues we identified the underexpression of miR-195-5p and the overexpression of miR-27a-3p. Both microRNAs were overexpressed in peritumoral tissue compared with normal tissue.

The analysis of Pearson correlations between the expression level of the analyzed genes and microRNAs and the socio-demographic, clinical and paraclinical parameters, as well as the morphofunctional characteristics of the tumor tissues revealed the following:

&#8626; miR-27a-3p was overexpressed in tumor tissues from 11 patients with lymph node metastases; in the same patients we identified a negative linear correlation between miR-27a-3p and CD36 expression and a positive linear correlation between miR-195-5p and FASN;

&#8626; miR-27a-3p was overexpressed in tumor tissues from 6 patients with perineural invasion.

We did not observe significant changes in the expression level of genes and microRNAs according to tumor location, TNM stage or degree of tumor invasion.

There were no correlations between the values of hematological, biochemical parameters, tumor markers and the expression level of the studied genes.

The mutational status of KRAS, BRAF genes and MSI status did not significantly influence the level of gene expression and microRNAs.


**2. Expression of let-7 family microRNAs: implications in colorectal cancer by targeting the IGF1 axis and possible association with perineural invasion**

We identified a general impairment of the expression of microRNAs of the let7 family in colorectal mucosa harvested from 25 patients with colorectal cancer compared to adjacent non-tumorous mucosa. Thus, let-7a-5p, let-7b-5p, let-7c-5p, let-7d-5p, let-7e-5p, let-7f-5p and let-7g-5p were statistically significantly underexpressed, and let-7i-5p was moderately underexpressed in tumor tissue, but without reaching statistical significance.

In tumor tissues with perineural invasion let-7a-5p, let-7b-5p, let-7c-5p, let-7d-5p and let-7i-5p were overexpressed compared to tumor tissues without perineural invasion.

In tumor tissues with KRAS mutations (codon 12 or codon 13) we found that miR-let-7e-5p was overexpressed. This increase was also observed comparing the peritumoral tissues of the two groups, although the peritumoral tissues did not show mutations.

No significant differences were observed in the expression level of the microRNAs analyzed according to the characteristics of the tumors (location, degree of tumor differentiation, lymphovascular invasion, microsatellite instability) or of the patients (age, sex, moderate alcohol consumption and hereditary history) for cancer).

### 3. Interrelationships of let-7 family microRNAs and genes involved in apoptosis and NF-κB signaling pathway

By means of bioinformatic analysis, we identified interactions between let7 family microRNAs and genes involved in the apoptosis process and in the NF-κB signaling pathway: BCL2A1, BCL2L1, CASP8, CFLAR, TNFRSF10A, TNFRSF10B, TNFRSF1A and TRAF2.

Analysis of gene expression on a group of 18 patients with colorectal cancer and 18 controls revealed the overexpression of the eight studied genes.

BCL2L1 and CASP8 genes were overexpressed in moderately differentiated tumor tissues (G2) compared to poorly differentiated tumor tissues (G3).

We identified no differences when we stratified patients according to sex, age, presence of comorbidities, tumor location, or TNM stage.

As a limitation of these studies, we could mention the low number of patients enrolled in the studies of this doctoral thesis and the lack of experimental evidence of microRNA – target gene interactions.

As a research direction for future studies we aim to deepen these in larger cohorts of patients and controls, as well as to validate the interactions in different colorectal tumor cell lines.

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# PUBLIC LIST OF WORKS

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