

CHAPTER 5

Gain of chromosome 20q is
an indicator of poor prognosis
in colorectal cancer

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Abstract

Background: Approximately 50 percent of CRC patients ultimately develop metastatic disease and adjuvant chemotherapy for stage III CRC has proven to be beneficial. For stage II colorectal cancer adjuvant chemotherapy is not standard, although this group probably harbours a subgroup of patients that will develop metastatic disease and could benefit from adjuvant chemotherapy. The aim of this study was to investigate if certain chromosomal aberrations are related with patient's survival.

Material & Methods: Eighteen CRCs were analyzed by comparative genomic hybridization (CGH) and their DNA copy number profiles were correlated with the survival status of the patient.

Results and Conclusion: Gain of 20q was significantly associated with worse patient survival ($P=0.002$) and therefore may serve as an indicator of poor patient outcome, and especially in patients with stage II CRC this could have clinical implications.

Introduction

Colorectal cancer (CRC) is the second leading cause of cancer death in the Western world, and its incidence is still rising. Improved surgery and preoperative radiotherapy have resulted in reduced local recurrences for rectal cancer, but no dramatic improvements in clinical outcome of colorectal cancer overall have been achieved. Still approximately fifty percent ultimately develop metastatic disease, which generally is fatal despite systemic chemotherapy. Adjuvant chemotherapy for CRC patients with unfavourable characteristics, i.e. presence of lymph node metastasis (stage III cancer) has proven to be beneficial. Adjuvant chemotherapy for stage II colorectal cancer is not standard, although this group probably harbours a subgroup of patients that do develop metastatic disease and could benefit from adjuvant chemotherapy. In addition to heterogeneity in clinical behaviour, colorectal cancer is also a heterogeneous disease at the level of underlying genetic changes with chromosomal instability as one of the key features occurring in eighty five percent of colorectal cancers [1-4]. Within the large category of chromosomal instable colorectal cancer, different combinations of aberrations occur. In a series of colorectal adenomas and carcinomas, analysed by comparative genomic hybridisation (CGH), we showed that seven chromosomal aberrations (loss at 8p21-pter, 15q11-q21, 17p12-13 and 18q12-21, and gain at 8q23-qter, 13q14-31 and 20q13) were specifically associated with adenoma to carcinoma progression [5]. Hierarchic cluster analysis demonstrated the presence of three distinct subgroups of adenomas, marked by characteristic combinations of genetic aberrations in the adenomas; one cluster was marked by 17p12-13 loss and *KRAS* mutation, one by 8q23-qter and 13q14-31 gain, and one by 18q12-21 loss and 20q13 gain. Clustering of all carcinomas yielded two subgroups. One group showed among other abnormalities, a high frequency of 17p12-13 losses and a low frequency of 18q12-21 loss and 20q13 gain, while the other group showed a reverse pattern. So these chromosomal abnormalities occurred in specific combinations of a few abnormalities rather than as a mere accumulation of events, indicating the existence of multiple independent chromosomal instability pathways of colorectal cancer progression [5]. Some of these abnormalities, 18q loss and 20q gain, are thought to be involved in a more aggressive clinical behaviour [6,7]. Therefore, the aim of this study was to investigate which chromosomal aberrations in colorectal cancer are related with patient's survival.

Material and Methods

In a series of 40 CRC patients previously studied by CGH [5], we investigated if there is a correlation between some chromosomal aberrations and survival. Patients with TNM stage I tumors and patients that died within three months after surgery were excluded. In the end, eighteen CRC patients could be included in the analysis.

Mean follow up was 60.7 months (range 9-119). Eight patients were male and 10 were female, with a mean age of 67.9 years (range 45-85). Ten carcinomas were TNM stage II, 6 were stage III and 2 were stage IV. Nine tumors were located at the proximal colon and 9 at the distal colon. Two carcinomas were well, 14 moderately and 2 poor differentiated. DNA was isolated from formaldehyde fixed and paraffin embedded tumor samples and comparative genomic hybridization (CGH) was performed as described before [5].

Statistical Analysis

Statistical analysis was carried out using SPSS version 12.0 statistical software package (SPSS Inc., Chicago, IL). The method of Kaplan-Meier with the log-rank test was used to evaluate patient survival in comparison with the different chromosomal aberrations.

For testing significance of differences in distribution of categorical variables, cross tables were analyzed with the two-sided Fisher Exact's test and Pearson Chi-square test. P values <0.05 were considered significant.

Results

CGH data

Mean numbers of losses and gains were 6.0 (range 0-16) and 3.8 (range 0-9), respectively. Most frequent losses (i.e. >40%) concerned 8p (44%), 17p (56%) and 18q (61%), and most frequent gains 8q (50%), 13q (67%) and 20q (50%).

Chromosomal changes in comparison with survival data

When we compared all chromosomal aberrations found to the survival status of the 18 CRC patients, only gain of 20q was associated with a significantly worse patient survival ($P=0.002$) (Figure 1), in fact all six patients that died of disease had tumors with 20q gain, out of a total of nine with 20q gain.

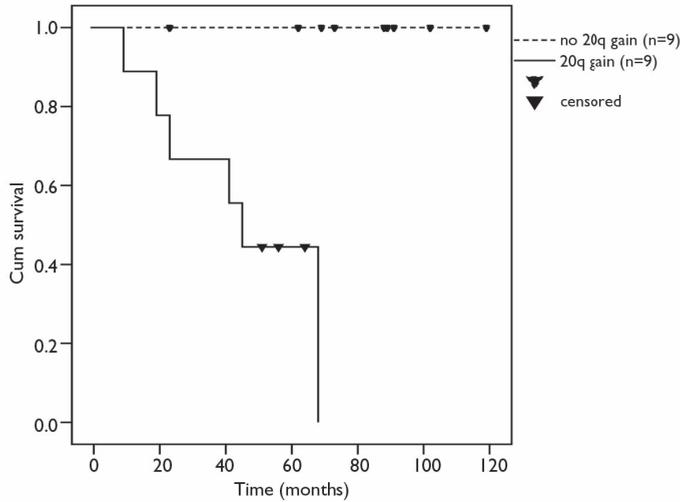


Figure 1. Cancer related survival in 18 colorectal cancer patients with and without gain of chromosome 20q detected by CGH. Patients with 20q gain positive colorectal cancer have a significantly ($P=0.002$) worse outcome.

Fewer patients with TNM stage II disease (40%) had 20q gain in their tumor than patients with stage III or IV disease (63%), but this was not significant in the present series. Also no correlations were found between presence of 20q gain and other clinico-pathological parameters (Table I).

Table I. Correlation between 20q gain and histological grade, stage, tumor size, and localization in the large intestine, respectively.

	Grade				TNM stage		
	Well	Moderate	Poor	Total no.	II	III + IV	Total no.
20q gain	1 (50%)	7 (50%)	1 (50%)	9	4 (40%)	5 (63%)	9
No 20q gain	1 (50%)	7 (50%)	1 (50%)	9	6 (60%)	3 (37%)	9
Total no.	2	14	2	18	10	8	18
	$(P=1)$				$(P=0.6)$		
	Size (cm)			Localization			
	<4	>4	Total no.	Proximal	Distal	Total no.	
20q gain	3 (50%)	6 (50%)	9	3 (33%)	6 (67%)	9	
No 20q gain	3 (50%)	6 (50%)	9	6 (67%)	3 (33%)	9	
Total no.	6	12	18	9	9	18	
	$(P=1)$			$(P=0.3)$			

In the present small series, there was a trend ($P=0.09$) for stage being associated with death of disease (DOD) (data not shown).

Discussion

Outcome of patients with colon cancer has improved by adjuvant chemotherapy, especially in those with TNM stage III. Although stage II carcinomas are not treated with chemotherapy, some will eventually develop metastases and would benefit from chemotherapy.

In this study we found that carcinomas of the colon with gain of 20q had a significant ($P=0.002$) worse patient survival than tumors without 20q gain. These data are consistent with other observations, either by CGH or FISH, in which 20q gain in colorectal cancer was associated with poor survival [6,7]. Gain of 20q studied by FISH or CGH has also been reported to be associated with metastatic CRC, which is an intermediate endpoint of survival [8-10]. Chromosome 20q contains many genes and a number of these have been implicated as driver genes, including ZNF217 [11-13]. However, also other candidate oncogenes may be involved, and further functional studies will be needed to settle this issue.

In conclusion, gain of 20q is an indicator of poor patient outcome in colorectal cancer, and especially in patients with stage II colorectal cancer this could have clinical implications.

Acknowledgements

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