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The landscape of genomic copy number alterations in colorectal cancer and their consequences on gene expression levels and disease outcome

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ABSTRACT

Aneuploidy, the unbalanced state of the chromosome content, represents a hallmark of most solid tumors, including colorectal cancer. Such aneuploidies result in tumor specific genomic imbalances, which emerge in premalignant precursor lesions. Moreover, increasing levels of chromosomal instability have been observed in adenocarcinomas and are maintained in distant metastases. A number of studies have systematically integrated copy number alterations with gene expression changes in primary carcinomas, cell lines, and experimental models of aneuploidy. In fact, chromosomal aneuploidies target a number of genes conferring a selective advantage for the metabolism of the cancer cell. Copy number alterations not only have a positive correlation with expression changes of the majority of genes on the altered genomic segment, but also have effects on the transcriptional levels of genes genome-wide. Finally, copy number alterations have been associated with disease outcome; nevertheless, the translational applicability in clinical practice requires further studies. Here, we (i) review the spectrum of genetic alterations that lead to colorectal cancer, (ii) describe the most frequent copy number alterations at different stages of colorectal carcinogenesis, (iii) exemplify their positive correlation with gene expression levels, and (iv) discuss copy number alterations that are potentially involved in disease outcome of individual patients.

1. Introduction to colorectal cancer genetics

1.1. Milestones in the genetic characterization of colorectal cancer

Cancer, colorectal cancer (CRC) included, is a disease marked by the aberrant behavior of cells that invade and destruct pre-existing tissues, both locally in the organ of origin and at a distance, i.e., metastatic sites. This selfish cell growth, and altered metabolic processes associated with it, can be lethal, despite of medical interventions. The aberrant behavior of tumor cells is driven by alterations in cell biology and affects critical processes such as proliferation, invasion, evasion to cellular death, and escape to immune surveillance, i.e., the so-called hallmarks of cancer (Hanahan and Weinberg, 2011). These alterations in cell biology, in turn, are the result of an evolutionary process by which gene mutations and somatically acquired copy number alterations (CNAs) accumulate and result in the selective advantage of cells that carry these alterations. The accumulation of genetic damage results in the expansion of initially benign (i.e., non-invasive) tumors, which, when untreated, form invasive subclones and lead to cancer. DNA alterations come in many flavors, namely, small nucleotide variants (SNVs), small insertions or deletions (Indels), structural variants (SVs) or epigenetic alterations, mostly promoter hypermethylation, and in chromosomal copy number alterations, i.e., aneuploidy. The role of chromosomal aneuploidy in tumorigenesis was for a long time underestimated despite observations by Hansemann in the 19th century (Hansemann, 1890), and work by Theodor Boveri in the 1920s

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(reviewed in Ried, 2009). In fact, in several hematological neoplasia and soft tissue tumors, specific chromosomal rearrangements had been found to be pathogenic (Rabbitts, 1994). Karyotyping of metaphases from tumor cells had been key to these observations, and this approach worked well in leukemias and soft tissue tumors, describing single or few chromosome rearrangements resulting in fusion genes (Heim and Mitelman, 2015). However, the role of specific chromosomal changes in the origin of solid tumors like CRC was harder to establish, mainly attributable to very complex and highly rearranged karyotypes. Yet, using methods like DNA flow cytometry and DNA image cytometry it had been clearly established that most CRCs showed abnormal nuclear DNA content, for which chromosomal copy number changes can be the only explanation (Giaretti et al., 2004; Steinbeck et al., 1994, 1993).

Groundbreaking work from Vogelstein and collaborators early in the 90's has shown that the accumulation of alterations in genes involved in key signaling pathways led to the neoplastic alteration of normal colonic epithelial cells, ultimately transforming into cancer (Fearon and Vogelstein, 1990). Accordingly, an early crucial event in the development of CRC is the disruption of the WNT-signaling pathway, leading to the formation of an adenoma, and this occurs in the majority of cases by disruption of the *APC* gene, either by truncating mutations or, less frequently, by loss of the long arm of chromosome 5 (5q), where *APC* is located. Subsequent accumulation of mutations in *KRAS* involved in the MAPK signaling pathway, losses on the long arm of chromosome 18, affecting TGF-β signaling pathway, and mutations in *TP53* or loss of the short arm of chromosome 17 (17p), where *TP53* is located, result in the formation of cancer (Fearon and Vogelstein, 1990).

With the introduction of comparative genomic hybridization (CGH) (du Manoir et al., 1993; Kallioniemi et al., 1992) and, later on, microarray CGH (array CGH) (Pinkel et al., 1998), it became possible to analyze CNAs genome-wide in one single experiment without the need for metaphase chromosome preparations from dividing cells. These molecular cytogenetic analyses confirmed and refined results established using conventional karyotype analysis (Bardi et al., 1993, 1991). This led to thorough characterization of non-random CNAs observed in microsatellite stable CRC, mainly gains of chromosomes 7, 8q, 13, and 20q as well as losses of 8p, 17p, and 18 (Camps et al., 2006; Douglas et al., 2004; Meijer et al., 1998; Nakao et al., 2004; Ried et al., 1996) Together with SNVs and CNAs, other alterations at the DNA or epigenome level turned out to be important as well. CpG island promoter methylation is a relevant mechanism for silencing gene expression (Baylin and Herman, 2000) and it is suggested to precede CNAs in the development of some CRC (Derks et al., 2006). In 2012, The Cancer Genome Atlas Network (TCGA) comprehensively characterized 276 CRCs at mutation, DNA copy number, hypermethylation and expression levels (Cancer Genome Atlas Network, 2012). They confirmed previously well-defined CNAs, including 1q, 7, 8q, 13q, and 20q gains and 1p, 4, 5q, 8p, 14q, 15q, 17p, and 18q losses as being frequently observed in CRC. These CNAs frequently co-occur within the same tumor, indicating potential interaction between loci within these chromosomes leading to a selective advantage (Hermsen et al., 2002; Takahashi et al., 2015).

1.2. Patho-biological classification of colorectal cancer

There are many ways in which CRCs can be patho-biologically classified. Two main genomic instability pathways are observed in CRC, the chromosomal instability (CIN) pathway, occurring in 85% of the cases, and the microsatellite instability (MSI) pathway, occurring in 15% of the cases (Lengauer et al., 1998). CIN CRCs are characterized by gross chromosomal aberrations (numerical and structural) while MSI CRCs are characterized by mutations at single nucleotide level in repetitive regions (microsatellites) resulting from a deficiency in the DNA mismatch repair mechanism (Boland and Goel, 2010).

CRCs can also be classified according to the levels of promoter hypermethylation (CpG Island Methylator Phenotype; CIMP), being divided in CIMP-high and CIMP-low CRCs (Toyota et al., 1999). There is a strong association of MSI phenotype with CIMP, due to hypermethylation of the mismatch repair gene *hMLH1* (Cancer Genome Atlas Network, 2012). Another classification based on the transcriptome has been put forward, including four different subtypes of CRC (Guinney et al., 2015). These Consensus Molecular Subtypes (CMS), however, are not completely discrete classes as there is some degree of overlap, reflecting a continuity of CRC transcriptomes (Ma et al., 2018). With the exception of CMS1 (MSI CRCs), all other 3 CMS groups (CMS2-4) present to a certain extent higher/lower degree of CNAs (CIN CRCs) (Guinney et al., 2015). Translation of CMS classification to preclinical models and clinical practice uncovered potential for targeted treatment (Sveen et al., 2018).

2. Temporal acquisition of CNAs throughout colorectal tumorigenesis

2.1. Adenoma to carcinoma progression

The disruption of the WNT-signaling pathway and the acquisition of chromosomal aneuploidy (e.g., extra copy of chromosome 7) might result in the formation of an adenoma, which progresses towards carcinoma through the accumulation of additional (epi)-genetic alterations in a background of genomic instability (Fearon and Vogelstein, 1990). Different precursor lesions, with different morphology, can lead to the development of CRC. These can be conventional (polypoid or flat) adenomas or sessile serrated polyps. Although the total amount of CNAs in advanced adenomas is low compared to carcinomas, the presence of chromosomal aneuploidies and genomic alterations in such premalignant lesions needs to be considered, contributing to the acquisition of relatively high levels of genetic heterogeneity (Cross et al., 2018). Furthermore, differences in the patterns of CNAs can be observed between different morphologies, namely, polypoid adenomas and non-polypoid adenomas. In a comparison of a large series of non-polypoid adenomas with polypoid adenomas, it was shown that non-polypoid adenomas more frequently presented 5q losses, but less 1p, 10q, 17p and 18q losses than polypoid adenomas (Voorham et al., 2012). Other precursors, such as sessile serrated polyps (IJspeert et al., 2015), progress to cancer via the MSI pathway, and therefore do not show the common CNAs observed in tumors arising via the CIN pathway (Bettington et al., 2017; Casorzo et al., 2015; Gaiser et al., 2013).

2.2. DNA copy number alterations associated with progression

Despite the fact that chromosomal aneuploidies can be observed in premalignant lesions, their appearance is more frequent in later stages during the transition to malignancy (Matano et al., 2015; Saito et al., 2018). Several studies showed that specific CNAs were associated with such transition (Hermsen et al., 2002; Meijer et al., 1998; Ried et al., 1996). It has been demonstrated that in adenomas in which a carcinoma is present (so-called progressed adenomas), the phenotypically benign part already contains most chromosomal aberrations present in the cancer, indicating a role of these aberrations, namely gains of 8q, 13q and 20q, and losses of 8p, 15q, 17p and 18q, in the adenoma-to-carcinoma progression (Carvalho et al., 2009; Hermsen et al., 2002) (Fig. 1). The presence of two or more of these CNAs has an accuracy of 80% for predicting the risk of an adenoma to progress to cancer (Hermsen et al., 2002). Moreover, studies modelling colorectal adenoma-to-carcinoma progression by perturbing organoids with gene mutations have shown that only organoids derived from adenomas with CNAs were able to form invasive tumors in mice, indicating the importance of these alterations (Drost et al., 2015; Matano et al., 2015). Genomic imbalances in distant metastases (e.g., liver) are of high similarity as those already present in the adenocarcinoma (Haan et al.,

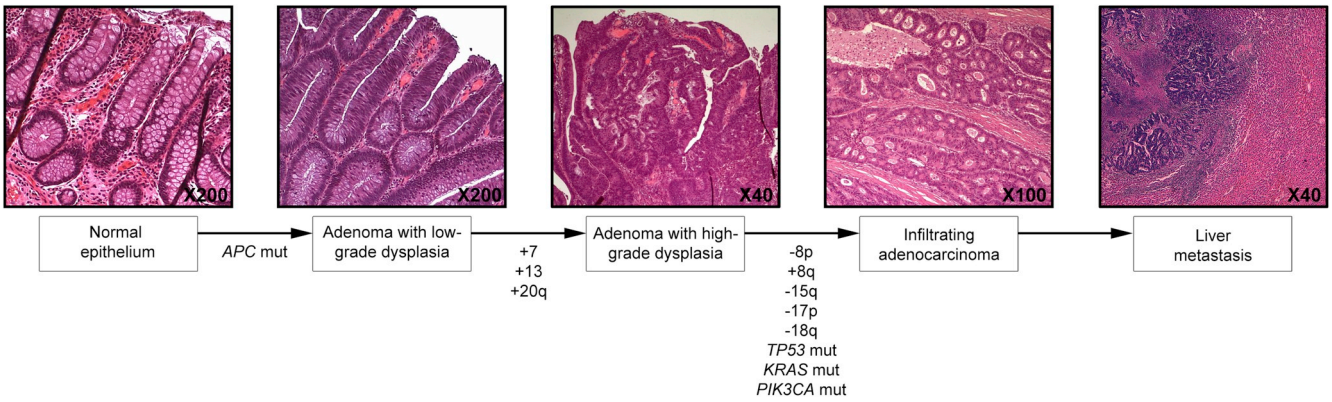

Fig. 1. Genomic alterations associated with the histopathological sequence of colorectal tumorigenesis. As shown, while *APC* mutation is present in most low-grade adenomas, chromosomal aneuploidies affecting chromosomes 7, 13 and 20q occur prior the transition from adenoma to adenocarcinoma. As indicated, additional genomic imbalances and single nucleotide variants result in the formation of the infiltrating adenocarcinoma providing the invasive phenotype. Histological sections are representative images stained with hematoxylin and eosin.

2014; Platzer et al., 2002; Xie et al., 2014). Altogether, these studies show the significance of CNAs with respect to the progression from colorectal adenomas to cancer.

2.3. High-risk adenoma versus advanced adenoma

Colorectal adenomas are a very common finding in elderly people with a prevalence of 18–35% reported in screening series (Imperiale et al., 2000; Lieberman et al., 2000). However, it is thought that only about 5% of colonic polyps removed at the time of endoscopy would have progressed to cancer (Shinya and Wolff, 1979). Indeed, histopathological features associated with the presence of focal cancer in adenomas include size ≥10 mm, high-grade dysplasia and villous histology (Muto et al., 1975; Shinya and Wolff, 1979). The presence of at least one of these histopathological features led to the concept of advanced adenoma (Winawer et al., 1993). However, the accuracy of these features to identify advanced adenomas that would be at risk to progress to cancer is low. Nonetheless, advanced adenomas are used in screening and surveillance programs as intermediate endpoints for CRC death. In a study evaluating the long-term effect of adenoma removal on reduction of CRC mortality, only a moderate effect was observed when advanced adenomas were removed (Løberg et al., 2014). This indicates that the use of advanced adenomas as intermediate endpoints associated with disease progression, leads to an overestimation of risk of dying from CRC, which translates to overdiagnosis. There is need for new intermediate endpoints that more precisely reflect the natural course of the disease, and more specifically identify adenomas at high risk of progressing to cancer (Sillars-Hardebol et al., 2012). In a recent pilot study, it has been shown that only 25% of advanced adenomas showed DNA CNAs associated with progression and that 2–4% of non-advanced adenomas already show these aberrations (Carvalho et al., 2018). This indicates that CNAs associated with progression from adenoma to cancer could more accurately identify adenomas at risk and therefore perform better as intermediate endpoints in CRC screening and surveillance programs.

3. Comprehensive assessment of copy number alterations

3.1. Experimental strategies to analyze copy number alterations in cancer

Beyond conventional G-banding, molecular cytogenetic techniques, including FISH-related approaches (Cremer et al., 1988, 1986; Pinkel et al., 1988; Ried et al., 1992; Schröck et al., 1996; Speicher et al., 1996) and conventional and array CGH (du Manoir et al., 1993; Kallioniemi et al., 1992, 1992; Pinkel et al., 1998), improved the analysis of chromosome aberrations in both hematological and solid tumors. CGH allowed mapping of genomic imbalances in solid tumors to an unprecedented level by comparing the genomic DNA extracted from the tumor sample with a reference genome without the requirement of metaphase chromosome preparations. This allowed the use of formalin-fixed and paraffin embedded material for cytogenetic analyses (Ried et al., 1995; Speicher et al., 1993). The application of CGH provided evidence that genomic imbalances were responsible for tumor progression from dysplastic lesions to invasive disease (Heselmeyer et al., 1996). Later on, single nucleotide polymorphism (SNP)-based DNA microarrays allowed simultaneous measurement of the allele-specific copy number at many different single nucleotide polymorphic loci in the genome, thus resulting in a high resolution detection of LOH, a common event in tumorigenesis, in addition to the identification of CNAs at a resolution similar to that of array CGH (Beroukhim et al., 2006; Heinrichs and Look, 2007; Mullighan et al., 2007). This technology also allowed the identification of copy neutral LOH, a common feature affecting cancer, including colorectal (Torabi et al., 2019, 2015; Tuna et al., 2009).

3.2. Novel methodologies to assess copy number alterations based on next-generation sequencing

Besides array CGH and SNP-arrays, the development of massive parallel sequencing through next-generation sequencing platforms led to the development of many tools to infer CNAs from whole-exome (WES) and whole-genome sequencing (WGS). Consensus “best-practices” workflow for short-reads sequencing management has been possible, in part, due to the Genome Analysis Toolkit (GATK) (McKenna et al., 2010). In addition, many other calling tools have been included in the omicX database (https://omictools.com) (Henry et al., 2014), some of the most relevant being listed in Table 1. Four computational approaches to handle genomic sequencing have been described to detect structural variants of the genome: (i) *read pair*, which compares distances between mapped paired reads and average insert size of the genomic library; (ii) *split read*, to detect small insertions and deletions through analyzing alignment to the reference genome, taking special attention to continuous stretches of gaps in the reads; (iii) *assembly methods*, where reference-free reconstruction of the entire genome from a collection of reads is computed and compared to the reference genome by applying multiple software; and (iv) *read count* or *depth of coverage*, the most recent approach, which takes into account the number of reads mapping for each region in the genome and assuming uniform sequencing process, so the number of reads in a specific region would be proportional to its copy number (Magi et al., 2017; Yoon et al., 2009). Recent studies have attempted to benchmark and evaluate the performance of several methods, further emphasizing differences

Table 1
Computational tools and methods for detection of CNAs detection in massive sequencing platforms.

Name	Sequencing platform	Implementation	Programming language	Link	Reference
CNVkit	WES/WGS	Software	Python	https://cnvkit.readthedocs.io/en/stable/index.html	Talevich et al. (2016)
ExomeDepth	WES	CRAN package	R	https://cran.r-project.org/web/packages/ExomeDepth/index.html	Plagnol et al. (2012)
VarScan2	WES/WGS	Command-line software	Java	http://dkoboldt.github.io/varscan/	Koboldt et al. (2012)
ControlFreeC	WES/WGS	Software	C++	http://boevalab.com/FREEC/	Boeva et al. (2011)
ExomeCNV	WES	CRAN package	R	https://cran.r-project.org/src/contrib/Archive/ExomeCNV/	Sathirapongsasuti et al. (2011)
XHMM	WES	Software	C++	https://atgu.mgh.harvard.edu/xhmm/index.shtml	Fromer et al. (2012)
CoNIFER	WES	Command-line software	Python	http://conifer.sourceforge.net/index.html	Krumm et al. (2012)
Delly	WGS	Software	C++	https://tobiasrausch.com/delly/	Rausch et al. (2012)
XCAVATOR	WGS	Software	Perl, bash, R, Fortran	http://sourceforge.net/projects/xcavator/	Magi et al. (2017)
CNVnator	WGS	Software	C++	http://sv.gersteinlab.org/cnvnator	Abyzov et al. (2011)
CNV-seq	WGS	Package	R, perl	http://tiger.dbs.nus.edu.sg/cnv-seq/	Xie and Tammi (2009)
Pindel	WGS	Command-line software	C++	http://gmt.genome.wustl.edu/packages/pindel/quick-start.html	Ye et al. (2009)
CONTRA	WES	Software	Python/R	http://contra-cnv.sourceforge.net	Li et al. (2012)

WES, whole-exome sequencing; WGS, whole-genome sequencing.

between tools despite the common usage of a calling method for data segmentation (Alkodsi et al., 2015; Kadalayil et al., 2015; Liu et al., 2013; Nam et al., 2016; Trost et al., 2018; Zare et al., 2017). Tools such as GISTIC2.0 (Mermel et al., 2011), ConVaQ (Larsen et al., 2018) or CNApp (Franch-Exposito et al., 2018) allow cohort-level studies to integrate genomic CNA data with additional molecular and clinical features, and unravel new functional implications for these genomic events.

3.3. Copy number alteration signatures

To a certain extent, cancer genome SNVs and CNAs delineate the track of unrepaired genetic alterations that have accumulated during the lifetime of the tumor. In this sense, studies of SNV signatures identified mutational patterns originated by different types of nucleotide changes in a given tumor type, defined as mutational signatures (Alexandrov et al., 2013; Nesic et al., 2018). In contrast to SNVs, only the calling for the presence or absence of a chromosome alteration in tumor cells, but not the underlying mechanisms of such patterns, are described. Therefore, efforts to identify copy number signatures have been made considering different approaches and accounting for diverse features. By applying non-negative matrix factorization models to 32 rearrangement subclasses in whole genome sequencing derived breast cancer data, six rearrangement signatures based on their association with homologous recombination deficiency and microhomology-mediated end joining mechanisms were extracted (Nik-Zainal et al., 2016). Likewise, eight copy number signatures based on structural features obtained from copy number profiles were identified by using low-pass whole genome sequencing in high-grade serous ovarian cancer (Macintyre et al., 2018). These authors correlated CNA signatures with prognosis and response to treatment, suggesting their utility as clinical biomarkers. Finally, pan-cancer studies unraveled nine signatures responsible for the etiology of structural variants, suggesting that replication-based mechanisms generated varying chromosomal structures with low-level copy number gains and frequent inverted rearrangements across different tumor types, including CRC (Camps et al., 2008; Li et al., 2017).

4. Transcriptional consequences of copy number alterations

4.1. Positive correlation between copy number alterations and expression levels

It has been firmly established that CNAs are specific to the tissue of cancer origin, thus reflecting specific landscapes of genomic imbalances for each tumor type (Beroukhim et al., 2010; Ried et al., 2012). As mentioned earlier, in colorectal carcinomas, tumorigenesis requires specific CNAs, e.g., gains of chromosomes 7, 8q, 13, and 20q, and losses of 8p, 17p, and 18. Such observations trigger the question what are the consequences of these so dominantly selected aneuploidies with respect to the transcription levels of genes on the affected chromosomes. In fact, among several hypotheses as to why transcriptional programs are affected by arm-level and whole chromosome-level CNAs, the body of literature indicates that CNAs directly affect the expression of the majority of genes on the altered genomic segment; however, the extent to which genes other than oncogenes and tumor suppressors contribute to malignant transformation or maintenance of the transformed state remains unclear. The biological consequence of chromosomal aneuploidy is not limited to the affected chromosomal region, but may be due to effects on the transcriptional activity of genes residing in other areas of the genome. Naturally, a third possibility is that chromosomal aneuploidies only target a small or limited number of genes conferring a selective advantage for the metabolism of the cancer cell.

Cell lines derived from primary carcinomas have been extensively used to measure the effect of genomic CNAs on the expression of resident genes. Analysis of 15 CRC cell lines, including mismatch repair proficient and deficient lines, showed positive genome-wide correlation between CNAs determined by array CGH and corresponding average gene expression (Camps et al., 2009). Such correlations have been confirmed in many other tumor types. For example, Wolf and colleagues arrived at the conclusion that a correlation between copy number with gene expression levels became evident genome-wide in prostate cancer cell lines (Wolf et al., 2004). Using the NCI-60 cancer cell line panel, a correlation between gene expression and copy number for all genes showed an overall medium high correlation, with significantly higher correlations for the known tumor suppressor genes (Varma et al., 2014). Recently, Tao and colleagues reported that gene up-regulation or down-regulation is significantly correlated with genomic amplification or deletion events in a set of six cervical cancer cell lines (Yan et al., 2017).

The correlation of genomic copy number and average gene expression levels also applies to primary tumors. In fact, several authors have shown the influence of canonical arm and chromosome-level CNAs on gene expression levels in pre-malignant lesions and carcinomas of different origin (Camps et al., 2008; Carvalho et al., 2009; Fehrmann et al., 2015; Grade et al., 2007, 2006; Ortiz-Estevez et al., 2011; Pollack et al., 2002) (Fig. 2). In this set of studies, the authors have explored several cohorts of colon and rectal cancer samples and matched normal mucosa, and identified that upregulation of genes was highest for those located on chromosomes 7, 13 and 20, i.e., chromosomes that are consistently gained, while downregulated genes were consistently located at chromosome 18, and to a lesser extent, at

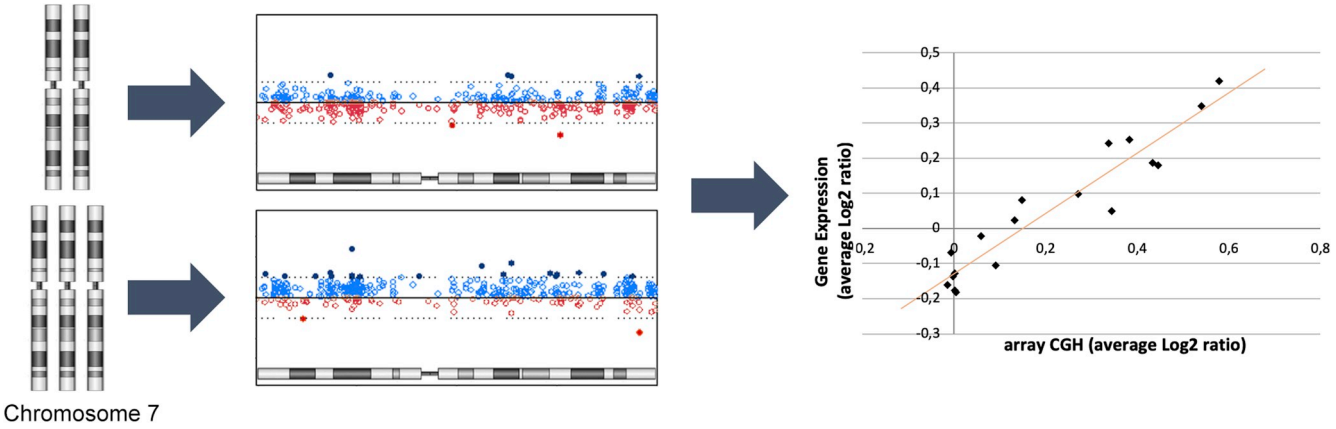

Fig. 2. Diagram depicting the positive correlation between genomic copy number alterations and levels of gene expression. The presence of an extra chromosome 7 results in the upregulation of most of the genes in this chromosome. Filled circles indicate genes that are considered over- (in blue) and under-expressed (in red) (middle panel) (adapted from Upender et al., 2004). Positive correlation between array CGH and gene expression for genes on chromosome 7 has been established for a set of 18 patients with colorectal cancer (R = 0.949) (right panel) (data source from Camps et al., 2013). (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

chromosomes 14 and 15, which are usually lost in CRC. By assessing the expression levels of normal colon epithelium, adenomas, carcinomas of different stages, and metastasis two independent studies provided further evidence of a copy number dependent transcriptional deregulation (Habermann et al., 2007; Tsafrir et al., 2006). Additionally, Sheffer and colleagues convincingly demonstrated that the pattern of gene expression profiles alone is sufficient to predict genomic CNAs in a comprehensive dataset of colorectal carcinomas (Sheffer et al., 2009). Recently, data derived from whole-genome sequencing and reported by The Cancer Genome Atlas consortium was used to map somatic structural changes, including CNAs, in 600 tumors of diverse origins, showing their contribution to altered gene expression in human cancer (Alaei-Mahabadi et al., 2016).

A gene-oriented assessment of such positive correlation between CNAs and gene expression has resulted in the discovery of novel cancer-related genes. In particular, in CRC, the gain of chromosome 13 and their associated overexpression of numerous genes, provided a unique chance to uncover several genes associated with tumorigenesis. Notably, these gene-dosage transcriptional dependent genes were mostly located at the proximal cytoband 13q12.12-q12.2. This included *CDK8*, *CDX2,* and *LNX2*, for which the overexpression was associated with WNT-pathway activity and tumorigenic features (Camps et al., 2013; Firestein et al., 2008; Salari et al., 2012). Similarly, but in a more distal region of the same chromosome, *DIS3*, *KLF5, IRS2,* and miRNA family miR17-92 were also chromosome 13 located cancer-related genes (Camps et al., 2003; Day et al., 2013; de Groen et al., 2014; Diosdado et al., 2009; Le et al., 2015). Besides overexpression of genes on chromosomes usually involved in recurrent CNAs, other regions of CRC genomes eventually involved in genomic imbalances do as well contain genes whose expression is correlated with gene dosage, such as the amplification and consequent overexpression of *IGF2* at 11p15.1 (Cancer Genome Atlas Network, 2012). Furthermore, an amplicon present in > 70% of colorectal tumor samples at 20q11-20q13 contained several cancer-related genes (*AHCY*, *POFUT1*, *RPN2*, *TH1L*, and *PRPF6*) that were upregulated and showed a significant linear correlation for gene dosage and gene expression (Loo et al., 2013). Also, *TPX2* (20q11.21) and *AURKA* (20q13.2), overexpressed as a consequence of gene-dosage, were shown to play a functional role as oncogenes driving this amplicon (Sillars-Hardebol et al., 2012). Similarly, copy number loss at 8p, a CNA associated with colorectal adenocarcinoma and poor prognosis, was observed in > 50% of the tumor samples and demonstrated a significant linear correlation for two potential tumor suppressor genes, *MTUS1* (8p22) and *PPP2CB* (8p12) (Loo et al., 2013). Additionally, focal deletions at 6q25-q27 reducing the expression of *PARK2* have been associated with *APC* deficiency in CRC (Poulogiannis et al., 2010b). Other integrative studies in CRC revealed that the genes *ARGLU1*, *UGGT2*, *CES2*, *FUT10,* and *PAOX* were the top five correlated genes in a set of 15 paired tumor-normal samples (Ali Hassan et al., 2014).

4.2. Model systems to study aneuploidy-dependent changes in the transcriptome

In order to study the impact of CNAs on gene expression, several cell line or animal models have been developed. Despite the technical difficulties on generating stable models of aneuploidy, several groups have addressed the physiological and functional effects of aneuploidy-dependent transcriptional deregulation in model systems, such as cell lines (Habermann et al., 2007; Nawata et al., 2011; Stingele et al., 2012; Upender et al., 2004; Wangsa et al., 2019), budding yeast (Torres et al., 2007), and mouse models (Williams et al., 2008). These and other studies have further revealed that CNAs affect the expression not only of the genes located on the aneuploid chromosome, but also of multiple other genes across the entire genome, which influence protein expression as well (Gemoll et al., 2013; Sheltzer et al., 2012). This massive transcriptional deregulation triggers cellular pathways associated with environmental stress response, which at least in yeast, seems to be related to the aneuploidy-driven slow growth (Donnelly and Storchová, 2014; Torres et al., 2007). In addition, cellular responses similar to those observed in artificially induced trisomic cells are related to proteotoxic stress, most likely due to the increased amount of the protein load in the cell, which may induce autophagy (Chunduri and Storchová, 2019). Furthermore, at the physiological level several model systems have suggested compromised growth as a consequence of induced aneuploidy, leading to the hypothesis that single-chromosome gains can suppress transformation in newly generated trisomies; however, after cellular progression and associated genome destabilization, the same study showed that evolved complex karyotypes contributed to the aggressive growth of advanced malignancies (Sheltzer et al., 2017).

5. CNAs as biomarkers for clinical outcome

5.1. Association of CNAs with clinical-histological features and outcome

Only a few genetic biomarkers are currently used in clinical decisions related to CRC. These include *RAS* mutation, which is used routinely in CRC patients to guide the administration of anti-*EGFR* therapy. Similarly, the *BRAF* V600E mutation is a biomarker of poor prognosis in

Table 2
Summary of the key copy number alterations and their associated genes in relation to disease outcome in CRC.

Chromosomal region	CNA	Genes of interest	Cohort size	Clinical relevance	Reference
1p36.33-p36.32	Gain	*SKI*	159	Patients having *SKI* amplification had a worse OS (HR = 2.62, p = 0.012) and DFS (HR = 2.08, p = 0.049)	Buess et al. (2004b)
3p14.2	Loss	*FHIT*	269	Significantly higher frequency in Stage III (24.3%) patients when compared to Stage II (3.3%) (p < 0.01)	Xie et al. (2012)
5p14.3-p13.3	Gain	*RNASEN, C5orf22, GOLPH3, MTMR12, ZFR, SUB1,* and *TARS*	111	In the CAPIRI therapy cohort, gain of this region was associated with a shorter PFS (p = 0.047)	Haan et al. (2014)
5q12.1–q12.3	Loss	*SFRS12IP1*, *SDCCAG10*, *CENPK*, *PPWD1,* and *SFRS12*	105	In the CAP therapy cohort, loss of this region was associated with a shorter PFS (p = < 0.001)	Haan et al. (2014)
5q34	Loss	*CCNG1*	133	In the CAPOX-B therapy cohort, loss of this region was associated with a shorter PFS (p = 0.001)	Haan et al. (2014)
6q16.1–q16.3	Gain	*KIAA0776*, *C6orf66*, *C6orf167*, *FBXL4*, *SFRS18*, *CCNC, ASCC3*, *ATG5*, *QRSL1*, *6orf203*, *PDSS2*, *LACE1*, *CD164*, *SMPD2,* and *ZBTB24*	111	In the CAPIRI therapy cohort, gain of this region was associated with a shorter PFS (p = < 0.002)	Haan et al. (2014)
7p11.2	Gain	*EGFR*	34	Significantly associated with improved OS (p = 0.03)	Lenz et al. (2006)
7p11.2	Gain	*EGFR*	31	Significantly associated with response to anti-EGFR monoclonal antibodies (p < 0·0001)	Moroni et al. (2005)
7p11.2	Gain	*EGFR*	44	Patients having *EGFR* CN gain achieved a high percentage of partial remission (p = 0.02), while those patients without *EGFR* CN gain had progressive disease (p = 0.001). Furthermore, patients having high *EGFR* copy number had a longer TTP (p = 0.04)	Scartozzi et al. (2009)
7q22.1	Gain	*GAEC1*	79	Significantly associated with tumor perforation (p = 0.001) and later T stage (p = 0.048)	Lee et al. (2018)
7q31.2	Gain	*MET*	16	*MET* amplification associated with cetuximab-resistance in patients that are wild-type for *KRAS*, *BRAF*, *NRAS*, *PIK3CA*, and *HER2*. (p = 0.006)	Bardelli et al. (2013)
12p12.3	Gain	*STRAP*	166	Patients having *STRAP* amplification and not receiving 5-FU/MMC adjuvant chemotherapy have a better prognosis (HR = 0.26, p = 0.004). Patients having the amplification and receiving 5-FU/MMC adjuvant therapy have a worse prognosis (HR = 3.48, p = 0.019)	Buess et al. (2004a)
17q21-q21.3	Loss	*PSMB3, PIP4K2B, CCDC49, RPL23, LASP1, RPL19, FBXL20, MED1, CRKRS, NEUROD2, STARD3,TOP2A, SMARCE1, TMEM99, KRTAP3-3, KRTAP1-1, EIF1, NT5C3L, KLHL11, ACLY, NKIRAS2, KAT2A, COASY, MLX, EZH1, VPS25, CCDC56, BECN1, PSME3, RUNDC1, RPL27, BRCA1, NBR2, NBR1, DUSP3, TMEM101, LSM12, TMUB2, GPATCH8, CCDC43, EFTUD2, NMT1,* and *MAP3K14*	133	In the CAPOX-B therapy cohort, loss of this region was associated with a shorter PFS (p = 0.004)	Haan et al. (2014)
18p11.32	Loss	*USP14, THOC1, C18orf56, TYMS, ENOSF1,* and *YES1*	111	In the CAPIRI therapy cohort, loss of this region was associated with a longer PFS (p = 0.04)	Haan et al. (2014)
18p11.32-p11.21	Loss	*METTL4, NDC80, SMCHD1, EMILIN2, LPIN2, MRCL3, MRLC2, ZFP161, RAB12, KIAA0802, NDUFV2, ANKRD12, TWSG1, RALBP1, PPP4R1, VAPA,* and *NAPG*	133	In the CAPOX-B therapy cohort, loss of this region was associated with a longer PFS (p = 0.048)	Haan et al. (2014)
18p11.21	Loss	*CHMP1B, MPPE1, IMPA2, TUBB6, AFG3L2, CEP76, PSMG2, PTPN2, SEH1L, CEP192, C18orf19,* and *RNMT*	133	In the CAPOX-B therapy cohort, loss of this region was associated with a longer PFS (p = 0.039)	Haan et al. (2014)
18p11.32	Gain	*TYMS*	111	Amplification was significantly associated with worse median survival in patients receiving adjuvant 5-FU based chemotherapy (HR = 2.7, p = 0.027)	Watson et al. (2010)
18q11.2	Loss	*LAMA3*	133	In the CAPOX-B therapy cohort, loss of this region was associated with a longer PFS (p = 0.014)	Haan et al. (2014)
18q12.2	Loss	*BRUNOL4*	30	Significantly associated with a worse prognosis (p = 0.007)	Poulogiannis et al., 2009
18q21.1	Loss	*SMAD7*	264	*SMAD7* deletion was associated with better OS (HR = 0.43, p = 0.0012) and DFS (HR = 0.50, p = 0.0033)	Boulay et al. (2003)
18q21.1	Gain	*SMAD7*	264	*SMAD7* duplication was associated with worse OS (HR = 2.10, p = 0.020) and DFS (HR = 2.06, p = 0.015)	Boulay et al. (2003)
18q21.2	Loss	*SMAD4*	147	Significantly associated with tumor progression (p = 0.041)	Ma et al. (2014)
18q21	Loss	*CD226*	172	Patients having *CD226* deletion had a better OS when treated with 5 FU (HR = 0.51, p = 0.05), while no significant difference in OS was observed between 5-FU treated and untreated patients with *CD226* retention. Patients in the control group having loss of *CD226* had a worse OS (HR = 2.44, p = 0.01) and DFS (HR = 1.99, p = 0.03)	Storojeva et al. (2005)

(*continued on next page*)

Table 2 (*continued*)

Chromosomal region	CNA	Genes of interest	Cohort size	Clinical relevance	Reference
18q21	Loss	*CADH-7*	165	Patients in the control group having loss of *CADH-7* had a better OS (HR = 0.29, p = 0.01) and DFS (HR = 0.43, p = 0.03)	Storojeva et al. (2005)
18q21.33–q22	Loss	*MYO5B, MBD1, CXXC1, C18orf24.ME2, ELAC1, SMAD4, MEX3C, MBD2, POLI, RAB27B, CCDC68, TXNL1, WDR7, FECH, NARS, ATP8B1, ALPK2, MALT1, SEC11C, LMAN1, PMAIP1, RNF152, PIGN, KIAA1468, TNFRSF11A, ZCCHC2, PHLPP, BCL2, KDSR, VPS4B, SERPINB8, TMX3, RTTN, SOCS6, C18orf55,* and *CNDP2*	111	In the CAPIRI therapy cohort, loss of this region was associated with a longer PFS (p = 0.001)	Haan et al. (2014)
19 p13.12	Loss		269	Significantly associated with OS in Stage II/III tumors (p < 0.01)	Xie et al. (2012)
20q11-q13.3	Gain	*BCL2L1, ASXL1, SRC, DNMT3B, GNAS, TOP1, AURKA, PTPRT,* and *NCOA3*	354	Chromosome 20q gain and amplification are significantly associated with longer OS (p = 0.015; CI, 0.36–0.90), (p = 0.039; CI, 0.11–0.94), respectively	Ptashkin et al. (2017)
20q11.21-q13.33	Gain	*PTK6 and EEFIA2*	269	Significantly associated with better OS in Stage III tumors (p < 0.01)	Xie et al. (2012)
20q13.2	Gain	*CSE1L, NABC1, ZNF217,* and *STK15*	146	Chromosomal gain was associated with worse overall survival (p = 0.006) and faster tumor progression (p = 0.012)	Aust et al. (2004)

CN, copy number; DFS, disease-free survival; HR, hazard ratio; MMC, mitomycin C; OS, overall survival; PFS, progression-free survival; TTP, time to progression; 5-FU, 5-fluorouracil.

patients with metastatic CRC (mCRC) (Yokota et al., 2011). Another prognostic marker used in the clinic, albeit it is only listed as a recommendation in the clinical guidelines, is the mismatch repair status (Sepulveda et al., 2017). Additionally, this guideline concluded that there was insufficient evidence to recommend the following as predictive biomarkers in CRC outside of clinical trials: *BRAF* V600E mutation, *PIK3CA* mutational analysis and *PTEN* expression or deletion (Sepulveda et al., 2017).

To date, there are still unmet clinical needs in certain areas of CRC management, notably for prognostication in stage II colon cancer patients, who suffer from a lack of effective stratification parameters to inform who are most likely to recur (Dimitriou et al., 2018). In fact, the majority of current predictive biomarkers are only applied to stage IV CRC patients. Thus, many patients would benefit from improved prognostic and predictive stratification, which would also help with clinical trial design, for example in basket trials. CRC still lacks adequate predictive biomarkers compared to other cancers such as melanoma, leukemia, breast, ovarian, prostate and lung cancer (Sameek and Chinnaiyan, 2014). As molecular cytogenetic methodologies as well as next-generation sequencing-based techniques to assess CNAs can be applied to archived formalin-fixed paraffin-embedded (FFPE) material, the analysis of large series of CRCs with well annotated clinical follow-up became feasible allowing the analysis of the prognostic and predictive value of CNAs. Therefore, CNAs affecting genes such as *SMAD2*, *MYC*, *EGFR*, *STRAP*, *SMAD7,* and *SMAD4* are being investigated for prognostic or predictive significance (Briffa et al., 2015; Hu et al., 2018; Postma et al., 2009). Another study on the effect of CNAs in several cancers highlighted their effect on proliferation and glycolysis (Graham et al., 2017). Despite the fact that several studies have been published on the prognostic and predictive values of CNAs in CRC, in this review we have limited our discussion to non-controversial candidate biomarkers that have been investigated in prospective clinical trials. The candidate biomarkers with their corresponding clinical relevance are summarized in Table 2.

5.2. Prognostic biomarkers

In advanced CRC, increased *EGFR* copy number is associated with poor survival and might be an independent prognostic variable (Lenz et al., 2006). With regard to *PTEN*, the Molecular Biomarkers Guideline for the Evaluation of Colorectal Cancer states that more rigorous studies are required for *PTEN* deletion to be accepted as a prognostic/predictive biomarker in CRC (Sepulveda et al., 2017). *STRAP* amplification was reported in 22.8% of stage II and III CRC (n = 166) (Buess et al., 2004a). This gene is located on chromosome 12 and encodes a serine/threonine kinase receptor associated protein. Of interest, patients that did not receive adjuvant therapy (n = 95) showed better prognosis when *STRAP* was amplified. In another cohort of 354 CRC patients (stage IV, n = 240), both gain and amplification of chromosome 20q were associated with longer overall survival (OS) in mCRC patients. Amplification was observed in 7% of mCRCs, while chromosome 20q copy number gains were observed in 37% of mCRC patients. This chromosome arm harbors several CRC-related oncogenes that might explain its prognostic value, including *SRC*, *AURKA, TPX2*, and *BCL2L1* (Ptashkin et al., 2017). The authors concluded that chromosome 20q gain/amplification is an early tumorigenic event and that it is involved with distant metastases.

Another chromosome of interest in CRC is chromosome 18q, and its deletion is postulated to be a biomarker of poor prognosis (Storojeva et al., 2005). Deletion of *CD226*, a gene located on chromosome 18q that encodes for a glycoprotein expressed on the surface of NK cells, platelets, monocytes and a subset of T-cells, has been shown to be a biomarker of poor prognosis for 5-year OS and disease-free survival (DFS) (n = 94) (Ptashkin et al., 2017; Wagner et al., 2017). In the same CRC cohort (n = 97), deletion of *CDH-7* copy number was a biomarker of good outcome with respect to 5-year OS and DFS. On the contrary, in

a cohort of 147 patients, *SMAD4* copy number loss was detected in 34.7% of Chinese CRC patients, was more frequent in mCRC compared to early stage CRC, and was associated with tumor progression (Ma et al., 2014). Boulay and colleagues reported that *SMAD7* was deleted in 43% of the 264 CRC cases and amplified in 15%. In this study, patients having *SMAD7* deletion had a statistically significant better OS and progression free survival (PFS). On the other hand, *SMAD7* amplification was associated with a worse OS and PFS. The authors speculated that on the assumption that *SMAD7* regulates TGFβ signaling, copy number amplifications antagonize cell growth arrest and apoptosis (Boulay et al., 2003). Similarly, a multivariate analysis identified the loss of *BRUNOL4* located at 18q12.2 as an independent factor associated with poor prognosis in CRC (Poulogiannis et al., 2010a).

Recently, Lee and colleagues showed that *GAEC1* (gene amplified in esophageal cancer 1), a putative oncogene located on chromosome 7 was amplified in 24.1% of the Australian CRC cohort (n = 79) (Lee et al., 2018). Moreover, copy number gain was associated with worse prognosis due to increased tumor aggressiveness. Another prognostic CNA is *SKI*, located on chromosome 1 and a repressor of TGF-β signaling. In a cohort of 533 stage II and III CRC cases, *SKI* gene copy number could be successfully measured in 159 patients and was amplified in 10.1% (Buess et al., 2004b). *SKI* amplification was associated with a worse OS and DFS as compared to patients with no copy number increase or deletions of this gene.

In 2002, a study on an early-stage CRC cohort (n = 180) investigated allelic imbalance of chromosomes 8p and 18q with respect to disease recurrence. Patients with Dukes' A stage tumors showing allelic imbalance in both chromosome arms were more likely to present with a recurrence when compared to Dukes' B patients without allelic imbalances (Zhou et al., 2002). Focal chromosomal CNAs have also been implicated in prognosis of CRC patients. A study on 46 stage II CRC concluded that loss of 5q34 was associated with worse OS and DFS and gain of 13q.22.1 was associated with worse OS (Brosens et al., 2010). Another study evaluated the effect of CNAs in a cohort (consisting of 89.1% MSS cases) of 302 stage II and III CRC patients with OS and progression-free survival (Xie et al., 2012). Deletion at 10p15.3-p14 and 19p13.12 in both stages were associated with poor OS. Furthermore, deletions of 10p15.3-p14 were also significantly associated with a worse RFS. On the contrary, in stage III patients, gain of 20q was associated with better OS. At the chromosome-arm level, loss of chromosome arm 4q (Brosens et al., 2011) and gain of chromosome arm 20q (Aust et al., 2004) were associated with poor prognosis. Also, both 8q gain and 20q gain were shown to be more frequently present in metastasized tumors than in tumors without metastases (Buffart et al., 2005; Hidaka et al., 2000).

5.3. Predictive biomarkers

Several CNAs have been associated with response to treatment in CRC. *TYMS* amplification was investigated in a cohort of mCRC patients treated with 5-fluorouracil (5-FU) based therapies, where it was associated with resistance to 5-FU (Watson et al., 2010). On the contrary, copy number deletion of the negative prognostic marker *CD226* is associated with better OS following 5-FU-based therapy (Storojeva et al., 2005). Buess and colleagues reported a study on stage II and III CRC patients (n = 166) and demonstrated that *STRAP* amplification results in a worse response to 5-FU based therapy which was observed with the patients having a higher relapse and death rate when compared to patients without amplification (Buess et al., 2004a). In 2008, Postma and colleagues carried out an investigation on CNA and response to capecitabine plus irinotecan (XELIRI regimen), in a cohort of treatment-naïve mCRC patients (n = 39). This study concluded that responders showed loss of chromosomal bands 18p11.32-q11.2 and 18q12.1-q23 (Postma et al., 2009). In the *KRAS* wild-type mCRC sub-group, only 13–17% benefit from single-agent anti-*EGFR* therapy (Bertotti et al., 2011). Additionally, in this sub-group increased *EGFR* copy numbers are associated with improved response rates to irinotecan-cetuximab therapy and longer time to progression (Scartozzi et al., 2009). Metastatic CRC patients with *EGFR* amplifications as confirmed by FISH are more likely to respond to monoclonal antibodies targeting EGFR (Moroni et al., 2005). In 2013, Jiang and colleagues reported a meta-analysis of 13 studies representing 1,174 mCRC patients treated with either cetuximab or panitumumab. Their results showed that *EGFR* copy number gains in this population were associated with improved OS and PFS (Jiang et al., 2013).

The mechanisms to understand why *KRAS* amplification in mCRC may be involved in resistance to EGFR-targeted therapies has been investigated (Valtorta et al., 2013). Yaeger and colleagues hypothesized that *RAS* amplification in *KRAS* wild-type CRCs induces acquired resistance to RAF inhibitors through the formation of drug-resistant RAF dimers, although the authors recognized that more studies are required to confirm this hypothesis (Yaeger et al., 2017). *MET* amplification is more frequent in mCRC cases that have acquired resistance to anti-*EGFR* therapy compared to *de novo* mCRC (Bardelli et al., 2013; Pietrantonio et al., 2016). Therefore, *MET* may be an ideal on-treatment monitoring candidate biomarker, especially since CNAs can be detected in cfDNA (Raghav et al., 2016).

In addition to its role in breast cancer, *HER2* amplification is an emerging biomarker in advanced CRC. Sartore-Bianchi and colleagues argued that *KRAS* exon 2 wild-type mCRCs should be tested for *HER2* amplification because, in this sub-group of patients, *HER2* amplifications may indicate responses to trastuzumab and pertuzumab; however, *KRAS*-mutant patients may be intrinsically resistant to anti-HER2 therapy even in the presence of *HER2* amplification (Sartore-Bianchi et al., 2018). Therefore, the overall genetic context matters, and individual CNAs alone may not fully indicate the treatment response of a particular tumor. In fact, Corcoran and colleagues reported *BRAF* amplification in a *BRAF* V600E mutated CRC and speculated that *BRAF* amplification in this cohort might lead to acquired resistance to MEK and BRAF inhibitors (Corcoran et al., 2010).

The presence of other specific CNAs has been associated with the response to treatment in mCRC. In a comprehensive analysis of CNAs in 349 tumors resected from patients participating in the CAIRO and CAIRO2 clinical trials, it was observed that specific chromosomal regions, mainly gain of 6q and loss of 18q, were associated with significant difference in progression-free survival (PFS) between treatment arms with and without irinotecan (Haan et al., 2014). Furthermore, van Dijk and colleagues recently reported that the loss of chromosome 18q11.2-q12.1 in mCRC patients was indicative of good prognosis as these patients achieved prolonged PFS, better OS and better overall response rate when receiving becacizumab (van Dijk et al., 2018). Similarly, Smeets and colleagues investigated the likelihood of other CNAs as candidate predictive biomarkers to guide therapy with bevacizumab in a cohort of mCRC patients (Smeets et al., 2018). In this study, the authors took a novel approach, where initially they clustered the CNA profiles of 908 CRC patients using unsupervised hierarchical clustering, resulting in three different clusters. The first cluster was characterized by tumors having a considerable amount of somatic mutations, including tumors having *POLE* and *POLD1* mutations and MSI classified tumors. Additionally, this cluster had only 10.2% advanced CRC patients and presented with few CNAs when compared with the other clusters. The authors proceeded with analyzing the response of the mCRC cohort with respect to chemotherapy alone versus chemotherapy in combination with bevacizumab. Patients in cluster 2 and 3 (intermediate to high instability) being administered chemotherapy in combination with bevacizumab had a better PFS and OS when compared to those receiving chemotherapy alone. On the other hand, patients in cluster 1 did not benefit from the addition of bevacizumab. The authors proceeded with comparing the CNA-based clusters to the current CMS classification system. CNA-based clusters overlapped with CMS sub-types: cluster 1 (low CNAs) was akin to CMS1

and 3, cluster 2 and 3 were similar to CMS 2 and 4. As a result, it was concluded that in mCRC patients, copy number load might be a novel predictive marker for the efficacy of bevacizumab combination therapy.

Besides gene dosage alterations, CIN can also lead to structural rearrangements, like breakpoints and translocations leading to fusions of different genes. Kloosterman and collegaues, in a systematic analysis of 278 CRCs, identified several novel oncogenic gene fusions in CRC that may drive malignant development and offer new targets for personalized therapy (Kloosterman et al., 2017). Chromosomal breakpoints in *MACROD2*, leading to loss of its expression, occurs frequently in advanced CRC (van den Broek et al., 2015). In addition, loss of expression of MACROD2 was associated with poor prognosis in stage II and III CRCs and with poor response to 5FU-based chemotherapy in stage III CRCs (van den Broek et al., 2018).

6. Conclusions and future challenges

There is a positive correlation between genomic CNAs and resident gene expression levels in cancer, resulting in a massive deregulation affecting not only genes on aneuploid chromosomes but also genes throughout the genome. This phenomenon is, naturally, associated with the fact that CNAs are cancer-type specific, which defines aneuploidy-dependent cancer type transcriptional programs. Taking this into consideration, several questions arise: (i) are all genes residing on a chromosome affected by such positive correlation or, alternatively, some genes escape this dependency? (ii) to what extent are CNAs the result of positively selected regions of the genome containing oncogenes and tumor suppressor genes? (iii) are those gene expression networks defined in carcinomas already functional in premalignant lesions? It remains to be clarified how exactly aneuploidy shapes the transcriptome of cancer cells and why cancer cells require such aneuploidy-specific deregulated transcriptional networks.

From the clinical point of view, the translational significance of genomic CNAs into clinical practice requires further studies. Eventually, understanding CNAs will allow stratification of patients based on biological and genetic features to improve disease prognostication, follow-up and to guide therapeutic strategies.

Conflicts of interest

The authors declare no conflicts of interest.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.mam.2019.07.007.

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References

Abyzov, A., Urban, A.E., Snyder, M., Gerstein, M., 2011. CNVnator: an approach to discover, genotype, and characterize typical and atypical CNVs from family and population genome sequencing. Genome Res. 21, 974–984. https://doi.org/10.1101/gr.114876.110.

Alaei-Mahabadi, B., Bhadury, J., Karlsson, J.W., Nilsson, J.A., Larsson, E., 2016. Global analysis of somatic structural genomic alterations and their impact on gene expression in diverse human cancers. Proc. Natl. Acad. Sci. U.S.A. 113, 13768–13773. https://doi.org/10.1073/pnas.1606220113.

Alexandrov, L.B., Nik-Zainal, S., Wedge, D.C., Aparicio, S.A.J.R., Behjati, S., Biankin, A.V., Bignell, G.R., Bolli, N., Borg, A., Børresen-Dale, A.-L., Boyault, S., Burkhardt, B., Butler, A.P., Caldas, C., Davies, H.R., Desmedt, C., Eils, R., Eyfjörd, J.E., Foekens, J.A., Greaves, M., Hosoda, F., Hutter, B., Ilicic, T., Imbeaud, S., Imielinski, M., Jäger, N., Jones, D.T.W., Jones, D., Knappskog, S., Kool, M., Lakhani, S.R., López-Otín, C., Martin, S., Munshi, N.C., Nakamura, H., Northcott, P.A., Pajic, M., Papaemmanuil, E., Paradiso, A., Pearson, J.V., Puente, X.S., Raine, K., Ramakrishna, M., Richardson, A.L., Richter, J., Rosenstiel, P., Schlesner, M., Schumacher, T.N., Span, P.N., Teague, J.W., Totoki, Y., Tutt, A.N.J., Valdés-Mas, R., van Buuren, M.M., van 't Veer, L., Vincent-Salomon, A., Waddell, N., Yates, L.R., Zucman-Rossi, J., Andrew Futreal, P., McDermott, U., Lichter, P., Meyerson, M., Grimmond, S.M., Siebert, R., Campo, E., Shibata, T., Pfister, S.M., Campbell, P.J., Stratton, M.R., Campo, E., Shibata, T., Pfister, S.M., Campbell, P.J., Stratton, M.R., 2013. Signatures of mutational processes in human cancer. Nature 500, 415–421. https://doi.org/10.1038/nature12477.

Ali Hassan, N.Z., Mokhtar, N.M., Kok Sin, T., Mohamed Rose, I., Sagap, I., Harun, R., Jamal, R., 2014. Integrated analysis of copy number variation and genome-wide expression profiling in colorectal cancer tissues. PLoS One 9, e92553. https://doi.org/10.1371/journal.pone.0092553.

Alkodsi, A., Louhimo, R., Hautaniemi, S., 2015. Comparative analysis of methods for identifying somatic copy number alterations from deep sequencing data. Briefings Bioinf. 16, 242–254. https://doi.org/10.1093/bib/bbu004.

Aust, D.E., Muders, M., Köhler, A., Schmidt, M., Diebold, J., Müller, C., Löhrs, U., Waldman, F.M., Baretton, G.B., 2004. Prognostic relevance of 20q13 gains in sporadic colorectal cancers: a FISH analysis. Scand. J. Gastroenterol. 39, 766–772. https://doi.org/10.1080/00365520410003191.

Bardelli, A., Corso, S., Bertotti, A., Hobor, S., Valtorta, E., Siravegna, G., Sartore-Bianchi, A., Scala, E., Cassingena, A., Zecchin, D., Apicella, M., Migliardi, G., Galimi, F., Lauricella, C., Zanon, C., Perera, T., Veronese, S., Corti, G., Amatu, A., Gambacorta, M., Diaz, L.A., Sausen, M., Velculescu, V.E., Comoglio, P., Trusolino, L., Di Nicolantonio, F., Giordano, S., Siena, S., 2013. Amplification of the MET receptor drives resistance to anti-EGFR therapies in colorectal cancer. Cancer Discov. 3, 658–673. https://doi.org/10.1158/2159-8290.CD-12-0558.

Bardi, G., Johansson, B., Pandis, N., Bak-Jensen, E., Orndal, C., Heim, S., Mandahl, N., Andrén-Sandberg, A., Mitelman, F., 1993. Cytogenetic aberrations in colorectal adenocarcinomas and their correlation with clinicopathologic features. Cancer 71, 306–314.

Bardi, G., Johansson, B., Pandis, N., Heim, S., Mandahl, N., Andrén-Sandberg, A., Hägerstrand, I., Mitelman, F., 1991. Trisomy 7 in short-term cultures of colorectal adenocarcinomas. Genes Chromosomes Cancer 3, 149–152.

Baylin, S.B., Herman, J.G., 2000. DNA hypermethylation in tumorigenesis: epigenetics joins genetics. Trends Genet. 16, 168–174.

Beroukhim, R., Lin, M., Park, Y., Hao, K., Zhao, X., Garraway, L.A., Fox, E.A., Hochberg, E.P., Mellinghoff, I.K., Hofer, M.D., Descazeaud, A., Rubin, M.A., Meyerson, M., Wong, W.H., Sellers, W.R., Li, C., 2006. Inferring loss-of-heterozygosity from unpaired tumors using high-density oligonucleotide SNP arrays. PLoS Comput. Biol. 2, e41. https://doi.org/10.1371/journal.pcbi.0020041.

Beroukhim, R., Mermel, C.H., Porter, D., Wei, G., Raychaudhuri, S., Donovan, J., Barretina, J., Boehm, J.S., Dobson, J., Urashima, M., Mc Henry, K.T., Pinchback, R.M., Ligon, A.H., Cho, Y.-J., Haery, L., Greulich, H., Reich, M., Winckler, W., Lawrence, M.S., Weir, B.A., Tanaka, K.E., Chiang, D.Y., Bass, A.J., Loo, A., Hoffman, C., Prensner, J., Liefeld, T., Gao, Q., Yecies, D., Signoretti, S., Maher, E., Kaye, F.J., Sasaki, H., Tepper, J.E., Fletcher, J.A., Tabernero, J., Baselga, J., Tsao, M.-S., Demichelis, F., Rubin, M.A., Janne, P.A., Daly, M.J., Nucera, C., Levine, R.L., Ebert, B.L., Gabriel, S., Rustgi, A.K., Antonescu, C.R., Ladanyi, M., Letai, A., Garraway, L.A., Loda, M., Beer, D.G., True, L.D., Okamoto, A., Pomeroy, S.L., Singer, S., Golub, T.R., Lander, E.S., Getz, G., Sellers, W.R., Meyerson, M., 2010. The landscape of somatic copy-number alteration across human cancers. Nature 463, 899–905. https://doi.org/10.1038/nature08822.

Bertotti, A., Migliardi, G., Galimi, F., Sassi, F., Torti, D., Isella, C., Corà, D., Di Nicolantonio, F., Buscarino, M., Petti, C., Ribero, D., Russolillo, N., Muratore, A., Massucco, P., Pisacane, A., Molinaro, L., Valtorta, E., Sartore-Bianchi, A., Risio, M., Capussotti, L., Gambacorta, M., Siena, S., Medico, E., Sapino, A., Marsoni, S., Comoglio, P.M., Bardelli, A., Trusolino, L., 2011. A molecularly annotated platform of patient-derived xenografts ("xenopatients") identifies HER2 as an effective therapeutic target in cetuximab-resistant colorectal cancer. Cancer Discov. 1, 508. 23. https://doi.org/10.1158/2159-8290.CD-11-0109.

Bettington, M., Walker, N., Rosty, C., Brown, I., Clouston, A., McKeone, D., Pearson, S.-A., Leggett, B., Whitehall, V., 2017. Clinicopathological and molecular features of sessile serrated adenomas with dysplasia or carcinoma. Gut 66, 97–106. https://doi.org/10.1136/gutjnl-2015-310456.

Boeva, V., Zinovyev, A., Bleakley, K., Vert, J.P., Janoueix-Lerosey, I., Delattre, O., Barillot, E., 2011. Control-free calling of copy number alterations in deep-sequencing data using GC-content normalization. Bioinformatics 27, 268–269. https://doi.org/10.1093/bioinformatics/btq635.

Boland, C.R., Goel, A., 2010. Microsatellite instability in colorectal cancer. Gastroenterology 138, 2073–2087. e3. https://doi.org/10.1053/j.gastro.2009.12.064.

Boulay, J.-L., Mild, G., Lowy, A., Reuter, J., Lagrange, M., Terracciano, L., Laffer, U., Herrmann, R., Rochlitz, C., 2003. SMAD7 is a prognostic marker in patients with colorectal cancer. Int. J. Cancer 104, 446–449. https://doi.org/10.1002/ijc.10908.

Briffa, R., Um, I., Faratian, D., Zhou, Y., Turnbull, A.K., Langdon, S.P., Harrison, D.J., 2015. Multi-scale genomic, transcriptomic and proteomic analysis of colorectal cancer cell lines to identify novel biomarkers. PLoS One 10, e0144708. https://doi.org/10.1371/journal.pone.0144708.

Brosens, R.P.M., Belt, E.J.T.H., Haan, J.C., Buffart, T.E., Carvalho, B., Grabsch, H., Quirke, P., Cuesta, M.A., Engel, A.F., Ylstra, B., Meijer, G.A., 2011. Deletion of chromosome 4q predicts outcome in Stage II colon cancer patients. Cell. Oncol. 34, 215–223. https://doi.org/10.1007/s13402-011-0042-8.

Brosens, R.P.M., Haan, J.C., Carvalho, B., Rustenburg, F., Grabsch, H., Quirke, P., Engel, A.F., Cuesta, M.A., Maughan, N., Flens, M., Meijer, G.A., Ylstra, B., 2010. Candidate driver genes in focal chromosomal aberrations of stage II colon cancer. J. Pathol. 221, 411–424. https://doi.org/10.1002/path.2724.

Buess, M., Terracciano, L., Reuter, J., Ballabeni, P., Boulay, J.-L., Laffer, U., Metzger, U., Herrmann, R., Rochlitz, C., 2004a. STRAP is a strong predictive marker of adjuvant chemotherapy benefit in colorectal cancer. Neoplasia 6, 813–820. https://doi.org/10.1593/neo.04307.

Buess, M., Terracciano, L., Reuter, J., Ballabeni, P., Boulay, J.-L., Laffer, U., Metzger, U., Herrmann, R., Rochlitz, C.F., 2004b. Amplification of SKI is a prognostic marker in early colorectal cancer. Neoplasia 6, 207–212. https://doi.org/10.1593/neo.03442.

Buffart, T.E., Coffa, J., Hermsen, M.A.J.A., Carvalho, B., van der Sijp, J.R.M., Ylstra, B., Pals, G., Schouten, J.P., Meijer, G.A., 2005. DNA copy number changes at 8q11-24 in metastasized colorectal cancer. Cell. Oncol. 27, 57–65.

Camps, J., Armengol, G., del Rey, J., Lozano, J.J., Vauhkonen, H., Prat, E., Egozcue, J., Sumoy, L., Knuutila, S., Miró, R., 2006. Genome-wide differences between microsatellite stable and unstable colorectal tumors. Carcinogenesis 27, 419–428. https://doi.org/10.1093/carcin/bgi244.

Camps, J., Grade, M., Nguyen, Q.T., Hörmann, P., Becker, S., Hummon, A.B., Rodriguez, V., Chandrasekharappa, S., Chen, Y., Difilippantonio, M.J., Becker, H., Ghadimi, B.M., Ried, T., 2008. Chromosomal breakpoints in primary colon cancer cluster at sites of structural variants in the genome. Cancer Res. 68, 1284–1295. https://doi.org/10.1158/0008-5472.CAN-07-2864.

Camps, J., Nguyen, Q.T., Padilla-Nash, H.M., Knutsen, T., McNeil, N.E., Wangsa, D., Hummon, A.B., Grade, M., Ried, T., Difilippantonio, M.J., 2009. Integrative genomics reveals mechanisms of copy number alterations responsible for transcriptional deregulation in colorectal cancer. Genes Chromosomes Cancer 48, 1002–1017. https://doi.org/10.1002/gcc.20699.

Camps, J., Pitt, J.J., Emons, G., Hummon, A.B., Case, C.M., Grade, M., Jones, T.L., Nguyen, Q.T., Ghadimi, B.M., Beissbarth, T., Difilippantonio, M.J., Caplen, N.J., Ried, T., 2013. Genetic amplification of the NOTCH modulator LNX2 upregulates the WNT/-catenin pathway in colorectal cancer. Cancer Res. 73, 2003–2013. https://doi.org/10.1158/0008-5472.CAN-12-3159.

Cancer Genome Atlas Network, 2012. Comprehensive molecular characterization of human colon and rectal cancer. Nature 487, 330–337. https://doi.org/10.1038/nature11252.

Carvalho, B., Diosdado, B., Terhaar Sive Droste, J.S., Bolijn, A.S., Komor, M.A., de Wit, M., Bosch, L.J.W., van Burink, M., Dekker, E., Kuipers, E.J., Coupé, V.M.H., van Grieken, N.C.T., Fijneman, R.J.A., Meijer, G.A., 2018. Evaluation of cancer-associated DNA copy number events in colorectal (advanced) adenomas. Cancer Prev. Res. 11, 403–412. https://doi.org/10.1158/1940-6207.CAPR-17-0317.

Carvalho, B., Postma, C., Mongera, S., Hopmans, E., Diskin, S., van de Wiel, M.A., van Criekinge, W., Thas, O., Matthäi, A., Cuesta, M.A., Terhaar Sive Droste, J.S., Craanen, M., Schröck, E., Ylstra, B., Meijer, G.A., 2009. Multiple putative oncogenes at the chromosome 20q amplicon contribute to colorectal adenoma to carcinoma progression. Gut 58, 79–89. https://doi.org/10.1136/gut.2007.143065.

Casorzo, L., Dell'Aglio, C., Sarotto, I., Risio, M., 2015. Aurora kinase A gene copy number is associated with the malignant transformation of colorectal adenomas but not with the serrated neoplasia progression. Hum. Pathol. 46, 411–418. https://doi.org/10.1016/j.humpath.2014.11.016.

Chunduri, N.K., Storchová, Z., 2019. The diverse consequences of aneuploidy. Nat. Cell Biol. 21, 54–62. https://doi.org/10.1038/s41556-018-0243-8.

Corcoran, R.B., Dias-Santagata, D., Bergethon, K., Iafrate, A.J., Settleman, J., Engelman, J.A., 2010. BRAF gene amplification can promote acquired resistance to MEK inhibitors in cancer cells harboring the BRAF V600E mutation. Sci. Signal. 3 ra84. https://doi.org/10.1126/scisignal.2001148.

Cremer, T., Landegent, J., Brückner, A., Scholl, H.P., Schardin, M., Hager, H.D., Devilee, P., Pearson, P., van der Ploeg, M., 1986. Detection of chromosome aberrations in the human interphase nucleus by visualization of specific target DNAs with radioactive and non-radioactive in situ hybridization techniques: diagnosis of trisomy 18 with probe L1.84. Hum. Genet. 74, 346–352.

Cremer, T., Lichter, P., Borden, J., Ward, D.C., Manuelidis, L., 1988. Detection of chromosome aberrations in metaphase and interphase tumor cells by in situ hybridization using chromosome-specific library probes. Hum. Genet. 80, 235–246.

Cross, W., Kovac, M., Mustonen, V., Temko, D., Davis, H., Baker, A.-M., Biswas, S., Arnold, R., Chegwidden, L., Gatenbee, C., Anderson, A.R., Koelzer, V.H., Martinez, P., Jiang, X., Domingo, E., Woodcock, D.J., Feng, Y., Kovacova, M., Maughan, T., Jansen, M., Rodriguez-Justo, M., Ashraf, S., Guy, R., Cunningham, C., East, J.E., Wedge, D.C., Wang, L.M., Palles, C., Heinimann, K., Sottoriva, A., Leedham, S.J., Graham, T.A., Tomlinson, I.P.M., Tomlinson, I.P.M., 2018. The evolutionary landscape of colorectal tumorigenesis. Nat. Ecol. Evol. 2, 1661–1672. https://doi.org/10.1038/s41559-018-0642-z.

Day, E., Poulogiannis, G., McCaughan, F., Mulholland, S., Arends, M.J., Ibrahim, A.E.K., Dear, P.H., 2013. IRS2 is a candidate driver oncogene on 13q34 in colorectal cancer. Int. J. Exp. Pathol. 94, 203–211. https://doi.org/10.1111/iep.12021.

de Groen, F.L.M., Krijgsman, O., Tijssen, M., Vriend, L.E.M., Ylstra, B., Hooijberg, E., Meijer, G.A., Steenbergen, R.D.M., Carvalho, B., 2014. Gene-dosage dependent overexpression at the 13q amplicon identifies DIS3 as candidate oncogene in colorectal cancer progression. Genes Chromosomes Cancer 53, 339–348. https://doi.org/10.1002/gcc.22144.

Derks, S., Postma, C., Moerkerk, P.T.M., van den Bosch, S.M., Carvalho, B., Hermsen, M.A.J.A., Giaretti, W., Herman, J.G., Weijenberg, M.P., de Bruïne, A.P., Meijer, G.A., van Engeland, M., 2006. Promoter methylation precedes chromosomal alterations in colorectal cancer development. Cell. Oncol. 28, 247–257.

Dimitriou, N., Arandjelović, O., Harrison, D.J., Caie, P.D., 2018. A principled machine learning framework improves accuracy of stage II colorectal cancer prognosis. NPJ Digit. Med. 1, 52. https://doi.org/10.1038/s41746-018-0057-x.

Diosdado, B., van de Wiel, M.A., Terhaar Sive Droste, J.S., Mongera, S., Postma, C., Meijerink, W.J.H.J., Carvalho, B., Meijer, G.A., 2009. MiR-17-92 cluster is associated with 13q gain and c-myc expression during colorectal adenoma to adenocarcinoma progression. Br. J. Canc. 101, 707–714. https://doi.org/10.1038/sj.bjc.6605037.

Donnelly, N., Storchová, Z., 2014. Dynamic karyotype, dynamic proteome: buffering the effects of aneuploidy. Biochim. Biophys. Acta Mol. Cell Res. 1843, 473–481. https://doi.org/10.1016/j.bbamcr.2013.11.017.

Douglas, E.J., Fiegler, H., Rowan, A., Halford, S., Bicknell, D.C., Bodmer, W., Tomlinson, I.P.M., Carter, N.P., 2004. Array comparative genomic hybridization analysis of colorectal cancer cell lines and primary carcinomas. Cancer Res. 64, 4817–4825. https://doi.org/10.1158/0008-5472.CAN-04-0328.

Drost, J., van Jaarsveld, R.H., Ponsioen, B., Zimberlin, C., van Boxtel, R., Buijs, A., Sachs, N., Overmeer, R.M., Offerhaus, G.J., Begthel, H., Korving, J., van de Wetering, M., Schwank, G., Logtenberg, M., Cuppen, E., Snippert, H.J., Medema, J.P., Kops, G.J.P.L., Clevers, H., 2015. Sequential cancer mutations in cultured human intestinal stem cells. Nature 521, 43–47. https://doi.org/10.1038/nature14415.

du Manoir, S., Speicher, M.R., Joos, S., Schröck, E., Popp, S., Döhner, H., Kovacs, G., Robert-Nicoud, M., Lichter, P., Cremer, T., 1993. Detection of complete and partial chromosome gains and losses by comparative genomic in situ hybridization. Hum. Genet. 90, 590–610.

Fearon, E.R., Vogelstein, B., 1990. A genetic model for colorectal tumorigenesis. Cell 61, 759–767.

Fehrmann, R.S.N., Karjalainen, J.M., Krajewska, M., Westra, H.-J., Maloney, D., Simeonov, A., Pers, T.H., Hirschhorn, J.N., Jansen, R.C., Schultes, E.A., van Haagen, H.H.H.B.M., de Vries, E.G.E., te Meerman, G.J., Wijmenga, C., van Vugt, M.A.T.M., Franke, L., 2015. Gene expression analysis identifies global gene dosage sensitivity in cancer. Nat. Genet. 47, 115–125. https://doi.org/10.1038/ng.3173.

Firestein, R., Bass, A.J., Kim, S.Y., Dunn, I.F., Silver, S.J., Guney, I., Freed, E., Ligon, A.H., Vena, N., Ogino, S., Chheda, M.G., Tamayo, P., Finn, S., Shrestha, Y., Boehm, J.S., Jain, S., Bojarski, E., Mermel, C., Barretina, J., Chan, J.A., Baselga, J., Tabernero, J., Root, D.E., Fuchs, C.S., Loda, M., Shivdasani, R.A., Meyerson, M., Hahn, W.C., 2008. CDK8 is a colorectal cancer oncogene that regulates β-catenin activity. Nature 455, 547–551. https://doi.org/10.1038/nature07179.

Franch-Exposito, S., Bassaganyas, L., Vila-Casadesus, M., Hernandez-Illan, E., Esteban-Fabro, R., Diaz-Gay, M., Lozano, J.J., Castells, A., Llovet, J.M., Castellvi-Bel, S., Camps, J., 2018. CNApp: a web-based tool for integrative analysis of genomic copy number alterations in cancer. bioRxiv 479667. https://doi.org/10.1101/479667.

Fromer, M., Moran, J.L., Chambert, K., Banks, E., Bergen, S.E., Ruderfer, D.M., Handsaker, R.E., McCarroll, S.A., O'Donovan, M.C., Owen, M.J., Kirov, G., Sullivan, P.F., Hultman, C.M., Sklar, P., Purcell, S.M., 2012. Discovery and statistical genotyping of copy-number variation from whole-exome sequencing depth. Am. J. Hum. Genet. 91, 597–607. https://doi.org/10.1016/j.ajhg.2012.08.005.

Gaiser, T., Meinhardt, S., Hirsch, D., Killian, J.K., Gaedcke, J., Jo, P., Ponsa, I., Miró, R., Rüschoff, J., Seitz, G., Hu, Y., Camps, J., Ried, T., 2013. Molecular patterns in the evolution of serrated lesion of the colorectum. Int. J. Cancer 132, 1800–1810. https://doi.org/10.1002/ijc.27869.

Gemoll, T., Habermann, J.K., Becker, S., Szymczak, S., Upender, M.B., Bruch, H.-P., Hellman, U., Ried, T., Auer, G., Jörnvall, H., Roblick, U.J., 2013. Chromosomal aneuploidy affects the global proteome equilibrium of colorectal cancer cells. Anal. Cell Pathol. 36, 149–161. https://doi.org/10.3233/ACP-140088.

Giaretti, W., Molinu, S., Ceccarelli, J., Prevosto, C., 2004. Chromosomal instability, aneuploidy, and gene mutations in human sporadic colorectal adenomas. Cell. Oncol. 26, 301–305.

Grade, M., Ghadimi, B.M., Varma, S., Simon, R., Wangsa, D., Barenboim-Stapleton, L., Liersch, T., Becker, H., Ried, T., Difilippantonio, M.J., 2006. Aneuploidy-dependent massive deregulation of the cellular transcriptome and apparent divergence of the Wnt/beta-catenin signaling pathway in human rectal carcinomas. Cancer Res. 66, 267–282. https://doi.org/10.1158/0008-5472.CAN-05-2533.

Grade, M., Hörmann, P., Becker, S., Hummon, A.B., Wangsa, D., Varma, S., Simon, R., Liersch, T., Becker, H., Difilippantonio, M.J., Ghadimi, B.M., Ried, T., 2007. Gene expression profiling reveals a massive, aneuploidy-dependent transcriptional deregulation and distinct differences between lymph node-negative and lymph node-positive colon carcinomas. Cancer Res. 67, 41–56. https://doi.org/10.1158/0008-5472.

CAN-06-1514.
Graham, N.A., Minasyan, A., Lomova, A., Cass, A., Balanis, N.G., Friedman, M., Chan, S., Zhao, S., Delgado, A., Go, J., Beck, L., Hurtz, C., Ng, C., Qiao, R., Ten Hoeve, J., Palaskas, N., Wu, H., Müschen, M., Multani, A.S., Port, E., Larson, S.M., Schultz, N., Braas, D., Christofk, H.R., Mellinghoff, I.K., Graeber, T.G., 2017. Recurrent patterns of DNA copy number alterations in tumors reflect metabolic selection pressures. Mol. Syst. Biol. 13, 914. https://doi.org/10.15252/msb.20167159.
Guinney, J., Dienstmann, R., Wang, X., de Reyniès, A., Schlicker, A., Soneson, C., Marisa, L., Roepman, P., Nyamundanda, G., Angelino, P., Bot, B.M., Morris, J.S., Simon, I.M., Gerster, S., Fessler, E., De Sousa E Melo, F., Missiaglia, E., Ramay, H., Barras, D., Homicsko, K., Maru, D., Manyam, G.C., Broom, B., Boige, V., Perez-Villamil, B., Laderas, T., Salazar, R., Gray, J.W., Hanahan, D., Tabernero, J., Bernards, R., Friend, S.H., Laurent-Puig, P., Medema, J.P., Sadanandam, A., Wessels, L., Delorenzi, M., Kopetz, S., Vermeulen, L., Tejpar, S., 2015. The consensus molecular subtypes of colorectal cancer. Nat. Med. 21, 1350–1356. https://doi.org/10.1038/nm.3967.
Haan, J.C., Labots, M., Rausch, C., Koopman, M., Tol, J., Mekenkamp, L.J.M., van de Wiel, M.A., Israeli, D., van Essen, H.F., van Grieken, N.C.T., Voorham, Q.J.M., Bosch, L.J.W., Qu, X., Kabbarah, O., Verheul, H.M.W., Nagtegaal, I.D., Punt, C.J.A., Ylstra, B., Meijer, G.A., 2014. Genomic landscape of metastatic colorectal cancer. Nat. Commun. 5, 5457. https://doi.org/10.1038/ncomms6457.
Habermann, J.K., Paulsen, U., Roblick, U.J., Upender, M.B., McShane, L.M., Korn, E.L., Wangsa, D., Krüger, S., Duchrow, M., Bruch, H.-P., Auer, G., Ried, T., 2007. Stage-specific alterations of the genome, transcriptome, and proteome during colorectal carcinogenesis. Genes Chromosomes Cancer 46, 10–26. https://doi.org/10.1002/gcc.20382.
Hanahan, D., Weinberg, R.A., 2011. Hallmarks of cancer: the next generation. Cell 144, 646–674. https://doi.org/10.1016/j.cell.2011.02.013.
Hansemann, D., 1890. Über asymmetrische Zellteilung in Epithelkrebsen und deren biologische Bedeutung. Virchows Arch. Pathol. 119, 299–326.
Heim, S., Mitelman, F., 2015. Cancer Cytogenetics: Chromosomal and Molecular Genetic Abberations of Tumor Cells, fourth ed. Wiley-Blackwell.
Heinrichs, S., Look, A.T., 2007. Identification of structural aberrations in cancer by SNP array analysis. Genome Biol. 8, 219. https://doi.org/10.1186/gb-2007-8-7-219.
Henry, V.J., Bandrowski, A.E., Pepin, A.-S., Gonzalez, B.J., Desfeux, A., 2014. OMICtools: an informative directory for multi-omic data analysis. Database (Oxford). 2014. bau069–bau069. https://doi.org/10.1093/database/bau069.
Hermsen, M., Postma, C., Baak, J., Weiss, M., Rapallo, A., Sciutto, A., Roemen, G., Arends, J.-W., Williams, R., Giaretti, W., De Goeij, A., Meijer, G., 2002. Colorectal adenoma to carcinoma progression follows multiple pathways of chromosomal instability. Gastroenterology 123, 1109–1119.
Heselmeyer, K., Schröck, E., du Manoir, S., Blegen, H., Shah, K., Steinbeck, R., Auer, G., Ried, T., 1996. Gain of chromosome 3q defines the transition from severe dysplasia to invasive carcinoma of the uterine cervix. Proc. Natl. Acad. Sci. U.S.A. 93, 479–484.
Hidaka, S., Yasutake, T., Takeshita, H., Kondo, M., Tsuji, T., Nanashima, A., Sawai, T., Yamaguchi, H., Nakagoe, T., Ayabe, H., Tagawa, Y., 2000. Differences in 20q13.2 copy number between colorectal cancers with and without liver metastasis. Clin. Cancer Res. 6, 2712–2717.
Hu, Y., Gaedcke, J., Emons, G., Beissbarth, T., Grade, M., Jo, P., Yeager, M., Chanock, S.J., Wolff, H., Camps, J., Ghadimi, B.M., Ried, T., 2018. Colorectal cancer susceptibility loci as predictive markers of rectal cancer prognosis after surgery. Genes Chromosomes Cancer 57. https://doi.org/10.1002/gcc.22512.
IJspeert, J.E.G., Medema, J.P., Dekker, E., 2015. Colorectal neoplasia pathways. Gastrointest. Endosc. Clin. N. Am. 25, 169–182. https://doi.org/10.1016/j.giec.2014.11.004.
Imperiale, T.F., Wagner, D.R., Lin, C.Y., Larkin, G.N., Rogge, J.D., Ransohoff, D.F., 2000. Risk of advanced proximal neoplasms in asymptomatic adults according to the distal colorectal findings. N. Engl. J. Med. 343, 169–174. https://doi.org/10.1056/NEJM200007203430302.
Jiang, Z., Li, C., Li, F., Wang, X., 2013. EGFR gene copy number as a prognostic marker in colorectal cancer patients treated with cetuximab or panitumumab: a systematic review and meta analysis. PLoS One 8, e56205. https://doi.org/10.1371/journal.pone.0056205.
Kadalayil, L., Rafiq, S., Rose-Zerilli, M.J.J., Pengelly, R.J., Parker, H., Oscier, D., Strefford, J.C., Tapper, W.J., Gibson, J., Ennis, S., Collins, A., 2015. Exome sequence read depth methods for identifying copy number changes. Briefings Bioinf. 16, 380–392. https://doi.org/10.1093/bib/bbu027.
Kallioniemi, A., Kallioniemi, O.P., Sudar, D., Rutovitz, D., Gray, J.W., Waldman, F., Pinkel, D., 1992. Comparative genomic hybridization for molecular cytogenetic analysis of solid tumors. Science 258, 818–821.
Kloosterman, W.P., Coebergh van den Braak, R.R.J., Pieterse, M., van Roosmalen, M.J., Sieuwerts, A.M., Stangl, C., Brunekreef, R., Lalmahomed, Z.S., Ooft, S., van Galen, A., Smid, M., Lefebvre, A., Zwartkruis, F., Martens, J.W.M., Foekens, J.A., Biermann, K., Koudijs, M.J., Ijzermans, J.N.M., Voest, E.E., 2017. A systematic analysis of oncogenic gene fusions in primary colon cancer. Cancer Res. 77, 3814–3822. https://doi.org/10.1158/0008-5472.CAN-16-3563.
Koboldt, D.C., Zhang, Q., Larson, D.E., Shen, D., McLellan, M.D., Lin, L., Miller, C.A., Mardis, E.R., Ding, L., Wilson, R.K., 2012. VarScan 2: somatic mutation and copy number alteration discovery in cancer by exome sequencing. Genome Res. 22, 568–576. https://doi.org/10.1101/gr.129684.111.
Krumm, N., Sudmant, P.H., Ko, A., O'Roak, B.J., Malig, M., Coe, B.P., NHLBI Exome Sequencing Project, N.N.E.S., Quinlan, A.R., Nickerson, D.A., Eichler, E.E., 2012. Copy number variation detection and genotyping from exome sequence data. Genome Res. 22, 1525–1532. https://doi.org/10.1101/gr.138115.112.
Larsen, S.J., do Canto, L.M., Rogatto, S.R., Baumbach, J., 2018. CoNVaQ: a web tool for copy number variation-based association studies. BMC Genomics 19, 369. https://doi.org/10.1186/s12864-018-4732-8.
Le, D.T., Uram, J.N., Wang, H., Bartlett, B.R., Kemberling, H., Eyring, A.D., Skora, A.D., Luber, B.S., Azad, N.S., Laheru, D., Biedrzycki, B., Donehower, R.C., Zaheer, A., Fisher, G.A., Crocenzi, T.S., Lee, J.J., Duffy, S.M., Goldberg, R.M., de la Chapelle, A., Koshiji, M., Bhaijee, F., Huebner, T., Hruban, R.H., Wood, L.D., Cuka, N., Pardoll, D.M., Papadopoulos, N., Kinzler, K.W., Zhou, S., Cornish, T.C., Taube, J.M., Anders, R.A., Eshleman, J.R., Vogelstein, B., Diaz, L.A., 2015. PD-1 blockade in tumors with mismatch-repair deficiency. N. Engl. J. Med. 372, 2509–2520. https://doi.org/10.1056/NEJMoa1500596.
Lee, K.T.-W., Gopalan, V., Islam, F., Wahab, R., Mamoori, A., Lu, C., Smith, R.A., Lam, A.K., 2018. GAEC1 mutations and copy number aberration is associated with biological aggressiveness of colorectal cancer. Eur. J. Cell Biol. 97, 230–241. https://doi.org/10.1016/j.ejcb.2018.03.002.
Lengauer, C., Kinzler, K.W., Vogelstein, B., 1998. Genetic instabilities in human cancers. Nature 396, 643–649. https://doi.org/10.1038/25292.
Lenz, H.-J., Van Cutsem, E., Khambata-Ford, S., Mayer, R.J., Gold, P., Stella, P., Mirtsching, B., Cohn, A.L., Pippas, A.W., Azarnia, N., Tsuchihashi, Z., Mauro, D.J., Rowinsky, E.K., 2006. Multicenter phase II and translational study of cetuximab in metastatic colorectal carcinoma refractory to irinotecan, oxaliplatin, and fluoropyrimidines. J. Clin. Oncol. 24, 4914–4921. https://doi.org/10.1200/JCO.2006.06.7595.
Li, J., Lupat, R., Amarasinghe, K.C., Thompson, E.R., Doyle, M.A., Ryland, G.L., Tothill, R.W., Halgamuge, S.K., Campbell, I.G., Gorringe, K.L., 2012. CONTRA: copy number analysis for targeted resequencing. Bioinformatics 28, 1307–1313. https://doi.org/10.1093/bioinformatics/bts146.
Li, Y., Roberts, N., Weischenfeldt, J., Wala, J.A., Shapira, O., Schumacher, S., Khurana, E., Korbel, J.O., Imielinski, M., Beroukhim, R., Campbell, P., 2017. Patterns of Structural Variation in Human Cancer. bioRxiv 181339. https://doi.org/10.1101/181339.
Lieberman, D.A., Weiss, D.G., Bond, J.H., Ahnen, D.J., Garewal, H., Harford, W.V., Provenzale, D., Sontag, S., Schnell, T., Durbin, T.E., Nelson, D.B., Ewing, S.L., Triadafilopoulos, G., Ramirez, F.C., Lee, J.G., Collins, J.F., Fennerty, M.B., Johnston, T.K., Corless, C.L., McQuaid, K.R., Sampliner, R.E., Morales, T.G., Fass, R., Smith, R., Maheshwari, Y., Chejfec, G., 2000. Use of colonoscopy to screen asymptomatic adults for colorectal cancer. N. Engl. J. Med. 343, 162–168. https://doi.org/10.1056/NEJM200007203430301.
Liu, B., Morrison, C.D., Johnson, C.S., Trump, D.L., Qin, M., Conroy, J.C., Wang, J., Liu, S., 2013. Computational methods for detecting copy number variations in cancer genome using next generation sequencing: principles and challenges. Oncotarget 4, 1868–1881. https://doi.org/10.18632/oncotarget.1537.
Løberg, M., Kalager, M., Holme, Ø., Hoff, G., Adami, H.-O., Bretthauer, M., 2014. Long-term colorectal-cancer mortality after adenoma removal. N. Engl. J. Med. 371, 799–807. https://doi.org/10.1056/NEJMoa1315870.
Loo, L.W.M., Tiirikainen, M., Cheng, I., Lum-Jones, A., Seifried, A., Church, J.M., Gryfe, R., Weisenberger, D.J., Lindor, N.M., Gallinger, S., Haile, R.W., Duggan, D.J., Thibodeau, S.N., Casey, G., Le Marchand, L., 2013. Integrated analysis of genome-wide copy number alterations and gene expression in microsatellite stable, CpG island methylator phenotype-negative colon cancer. Genes Chromosomes Cancer 52, 450–466. https://doi.org/10.1002/gcc.22043.
Ma, X., Ezer, D., Adryan, B., Stevens, T.J., 2018. Canonical and single-cell Hi-C reveal distinct chromatin interaction sub-networks of mammalian transcription factors. Genome Biol. 19, 174. https://doi.org/10.1186/s13059-018-1558-2.
Ma, Y., Yan, F., Li, L., Liu, L., Sun, J., 2014. Deletion and down-regulation of SMAD4 gene in colorectal cancers in a Chinese population. Chin. J. Canc. Res. 26, 525–531. https://doi.org/10.3978/j.issn.1000-9604.2014.09.02.
Macintyre, G., Goranova, T.E., De Silva, D., Ennis, D., Piskorz, A.M., Eldridge, M., Sie, D., Lewsley, L.-A., Hanif, A., Wilson, C., Dowson, S., Glasspool, R.M., Lockley, M., Brockbank, E., Montes, A., Walther, A., Sundar, S., Edmondson, R., Hall, G.D., Clamp, A., Gourley, C., Hall, M., Fotopoulou, C., Gabra, H., Paul, J., Supernat, A., Millan, D., Hoyle, A., Bryson, G., Nourse, C., Mincarelli, L., Sanchez, L.N., Ylstra, B., Jimenez-Linan, M., Moore, L., Hofmann, O., Markowetz, F., McNeish, I.A., Brenton, J.D., 2018. Copy number signatures and mutational processes in ovarian carcinoma. Nat. Genet. 50, 1262–1270. https://doi.org/10.1038/s41588-018-0179-8.
Magi, A., Pippucci, T., Sidore, C., 2017. XCAVATOR: accurate detection and genotyping of copy number variants from second and third generation whole-genome sequencing experiments. BMC Genomics 18, 747. https://doi.org/10.1186/s12864-017-4137-0.
Matano, M., Date, S., Shimokawa, M., Takano, A., Fujii, M., Ohta, Y., Watanabe, T., Kanai, T., Sato, T., 2015. Modeling colorectal cancer using CRISPR-Cas9-mediated engineering of human intestinal organoids. Nat. Med. 21, 256–262. https://doi.org/10.1038/nm.3802.
McKenna, A., Hanna, M., Banks, E., Sivachenko, A., Cibulskis, K., Kernytsky, A., Garimella, K., Altshuler, D., Gabriel, S., Daly, M., DePristo, M.A., 2010. The Genome Analysis Toolkit: a MapReduce framework for analyzing next-generation DNA sequencing data. Genome Res. 20, 1297–1303. https://doi.org/10.1101/gr.107524.110.
Meijer, G.A., Hermsen, M.A., Baak, J.P., van Diest, P.J., Meuwissen, S.G., Beliën, J.A., Hoovers, J.M., Joenje, H., Snijders, P.J., Walboomers, J.M., 1998. Progression from colorectal adenoma to carcinoma is associated with non-random chromosomal gains as detected by comparative genomic hybridisation. J. Clin. Pathol. 51, 901–909.
Mermel, C.H., Schumacher, S.E., Hill, B., Meyerson, M.L., Beroukhim, R., Getz, G., 2011. GISTIC2.0 facilitates sensitive and confident localization of the targets of focal somatic copy-number alteration in human cancers. Genome Biol. 12, R41. https://doi.org/10.1186/gb-2011-12-4-r41.
Moroni, M., Veronese, S., Benvenuti, S., Marrapese, G., Sartore-Bianchi, A., Di Nicolantonio, F., Gambacorta, M., Siena, S., Bardelli, A., 2005. Gene copy number for epidermal growth factor receptor (EGFR) and clinical response to antiEGFR treatment in colorectal cancer: a cohort study. Lancet Oncol. 6, 279–286. https://doi.org/10.1016/S1470-2045(05)70102-9.

Mullighan, C.G., Goorha, S., Radtke, I., Miller, C.B., Coustan-Smith, E., Dalton, J.D., Girtman, K., Mathew, S., Ma, J., Pounds, S.B., Su, X., Pui, C.-H., Relling, M.V., Evans, W.E., Shurtleff, S.A., Downing, J.R., 2007. Genome-wide analysis of genetic alterations in acute lymphoblastic leukaemia. Nature 446, 758–764. https://doi.org/10.1038/nature05690.

Muto, T., Bussey, H.J., Morson, B.C., 1975. The evolution of cancer of the colon and rectum. Cancer 36, 2251–2270.

Nakao, K., Mehta, K.R., Fridlyand, J., Moore, D.H., Jain, A.N., Lafuente, A., Wiencke, J.W., Terdiman, J.P., Waldman, F.M., 2004. High-resolution analysis of DNA copy number alterations in colorectal cancer by array-based comparative genomic hybridization. Carcinogenesis 25, 1345–1357. https://doi.org/10.1093/carcin/bgh134.

Nam, J.-Y., Kim, N.K.D., Kim, S.C., Joung, J.-G., Xi, R., Lee, S., Park, P.J., Park, W.-Y., 2016. Evaluation of somatic copy number estimation tools for whole-exome sequencing data. Briefings Bioinf. 17, 185–192. https://doi.org/10.1093/bib/bbv055.

Nawata, H., Kashino, G., Tano, K., Daino, K., Shimada, Y., Kugoh, H., Oshimura, M., Watanabe, M., 2011. Dysregulation of gene expression in the artificial human trisomy cells of chromosome 8 associated with transformed cell phenotypes. PLoS One 6, e25319. https://doi.org/10.1371/journal.pone.0025319.

Nesic, K., Wakefield, M., Kondrashova, O., Scott, C.L., McNeish, I.A., 2018. Targeting DNA repair: the genome as a potential biomarker. J. Pathol. 244, 586–597. https://doi.org/10.1002/path.5025.

Nik-Zainal, S., Davies, H., Staaf, J., Ramakrishna, M., Glodzik, D., Zou, X., Martincorena, I., Alexandrov, L.B., Martin, S., Wedge, D.C., Van Loo, P., Ju, Y.S., Smid, M., Brinkman, A.B., Morganella, S., Aure, M.R., Lingjærde, O.C., Langerød, A., Ringnér, M., Ahn, S.-M., Boyault, S., Brock, J.E., Broeks, A., Butler, A., Desmedt, C., Dirix, L., Dronov, S., Fatima, A., Foekens, J.A., Gerstung, M., Hooijer, G.K.J., Jang, S.J., Jones, D.R., Kim, H.-Y., King, T.A., Krishnamurthy, S., Lee, H.J., Lee, J.-Y., Li, Y., McLaren, S., Menzies, A., Mustonen, V., O'Meara, S., Pauporté, I., Pivot, X., Purdie, C.A., Raine, K., Ramakrishnan, K., Rodríguez-González, F.G., Romieu, G., Sieuwerts, A.M., Simpson, P.T., Shepherd, R., Stebbings, L., Stefansson, O.A., Teague, J., Tommasi, S., Treilleux, I., Van den Eynden, G.G., Vermeulen, P., Vincent-Salomon, A., Yates, L., Caldas, C., van't Veer, L., Tutt, A., Knappskog, S., Tan, B.K.T., Jonkers, J., Borg, Å., Ueno, N.T., Sotiriou, C., Viari, A., Futreal, P.A., Campbell, P.J., Span, P.N., Van Laere, S., Lakhani, S.R., Eyfjord, J.E., Thompson, A.M., Birney, E., Stunnenberg, H.G., van de Vijver, M.J., Martens, J.W.M., Børresen-Dale, A.-L., Richardson, A.L., Kong, G., Thomas, G., Stratton, M.R., 2016. Landscape of somatic mutations in 560 breast cancer whole-genome sequences. Nature 534, 47–54. https://doi.org/10.1038/nature17676.

Ortiz-Estevez, M., De Las Rivas, J., Fontanillo, C., Rubio, A., 2011. Segmentation of genomic and transcriptomic microarrays data reveals major correlation between DNA copy number aberrations and gene-loci expression. Genomics 97, 86–93. https://doi.org/10.1016/j.ygeno.2010.10.008.

Pietrantonio, F., Oddo, D., Gloghini, A., Valtorta, E., Berenato, R., Barault, L., Caporale, M., Busico, A., Morano, F., Gualeni, A.V., Alessi, A., Siravegna, G., Perrone, F., Di Bartolomeo, M., Bardelli, A., de Braud, F., Di Nicolantonio, F., 2016. MET-driven resistance to dual EGFR and BRAF blockade may Be overcome by switching from EGFR to MET inhibition in BRAF-mutated colorectal cancer. Cancer Discov. 6, 963–971. https://doi.org/10.1158/2159-8290.CD-16-0297.

Pinkel, D., Landegent, J., Collins, C., Fuscoe, J., Segraves, R., Lucas, J., Gray, J., 1988. Fluorescence in situ hybridization with human chromosome-specific libraries: detection of trisomy 21 and translocations of chromosome 4. Proc. Natl. Acad. Sci. U.S.A. 85, 9138–9142.

Pinkel, D., Segraves, R., Sudar, D., Clark, S., Poole, I., Kowbel, D., Collins, C., Kuo, W.L., Chen, C., Zhai, Y., Dairkee, S.H., Ljung, B.M., Gray, J.W., Albertson, D.G., 1998. High resolution analysis of DNA copy number variation using comparative genomic hybridization to microarrays. Nat. Genet. 20, 207–211. https://doi.org/10.1038/2524.

Plagnol, V., Curtis, J., Epstein, M., Mok, K.Y., Stebbings, E., Grigoriadou, S., Wood, N.W., Hambleton, S., Burns, S.O., Thrasher, A.J., Kumararatne, D., Doffinger, R., Nejentsev, S., 2012. A robust model for read count data in exome sequencing experiments and implications for copy number variant calling. Bioinformatics 28, 2747–2754. https://doi.org/10.1093/bioinformatics/bts526.

Platzer, P., Upender, M.B., Wilson, K., Willis, J., Lutterbaugh, J., Nosrati, A., Willson, J.K.V., Mack, D., Ried, T., Markowitz, S., 2002. Silence of chromosomal amplifications in colon cancer. Cancer Res. 62, 1134–1138.

Pollack, J.R., Sørlie, T., Perou, C.M., Rees, C.A., Jeffrey, S.S., Lonning, P.E., Tibshirani, R., Botstein, D., Børresen-Dale, A.-L., Brown, P.O., 2002. Microarray analysis reveals a major direct role of DNA copy number alteration in the transcriptional program of human breast tumors. Proc. Natl. Acad. Sci. U.S.A. 99, 12963–12968. https://doi.org/10.1073/pnas.162471999.

Postma, C., Koopman, M., Buffart, T.E., Eijk, P.P., Carvalho, B., Peters, G.J., Ylstra, B., van Krieken, J.H., Punt, C.J.A., Meijer, G.A., 2009. DNA copy number profiles of primary tumors as predictors of response to chemotherapy in advanced colorectal cancer. Ann. Oncol. Off. J. Eur. Soc. Med. Oncol. 20, 1048–1056. https://doi.org/10.1093/annonc/mdn738.

Poulogiannis, G., Ichimura, K., Hamoudi, R.A., Luo, F., Leung, S.Y., Yuen, S.T., Harrison, D.J., Wyllie, A.H., Arends, M.J., 2010a. Prognostic relevance of DNA copy number changes in colorectal cancer. J. Pathol. 220, 338–347. https://doi.org/10.1002/path.2640.

Poulogiannis, G., McIntyre, R.E., Dimitriadi, M., Apps, J.R., Wilson, C.H., Ichimura, K., Luo, F., Cantley, L.C., Wyllie, A.H., Adams, D.J., Arends, M.J., 2010b. PARK2 deletions occur frequently in sporadic colorectal cancer and accelerate adenoma development in Apc mutant mice. Proc. Natl. Acad. Sci. U.S.A. 107, 15145–15150. https://doi.org/10.1073/pnas.1009941107.

Ptashkin, R.N., Pagan, C., Yaeger, R., Middha, S., Shia, J., O'Rourke, K.P., Berger, M.F., Wang, L., Cimera, R., Wang, J., Klimstra, D.S., Saltz, L., Ladanyi, M., Zehir, A., Hechtman, J.F., 2017. Chromosome 20q amplification defines a subtype of microsatellite stable, left-sided colon cancers with wild-type RAS/RAF and better overall survival. Mol. Cancer Res. 15, 708–713. https://doi.org/10.1158/1541-7786.MCR-16-0352.

Rabbitts, T.H., 1994. Chromosomal translocations in human cancer. Nature 372, 143–149. https://doi.org/10.1038/372143a0.

Raghav, K., Morris, V., Tang, C., Morelli, P., Amin, H.M., Chen, K., Manyam, G.C., Broom, B., Overman, M.J., Shaw, K., Meric-Bernstam, F., Maru, D., Menter, D., Ellis, L.M., Eng, C., Hong, D., Kopetz, S., 2016. MET amplification in metastatic colorectal cancer: an acquired response to EGFR inhibition, not a de novo phenomenon. Oncotarget 7, 54627–54631. https://doi.org/10.18632/oncotarget.10559.

Rausch, T., Zichner, T., Schlattl, A., Stütz, A.M., Benes, V., Korbel, J.O., 2012. DELLY: structural variant discovery by integrated paired-end and split-read analysis. Bioinformatics 28, i333–i339. https://doi.org/10.1093/bioinformatics/bts378.

Ried, T., 2009. Homage to Theodor Boveri (1862-1915): Boveri's theory of cancer as a disease of the chromosomes, and the landscape of genomic imbalances in human carcinomas. Environ. Mol. Mutagen. 50, 593–601. https://doi.org/10.1002/em.20526.

Ried, T., Hu, Y., Difilippantonio, M.J., Ghadimi, B.M., Grade, M., Camps, J., 2012. The consequences of chromosomal aneuploidy on the transcriptome of cancer cells. Biochim. Biophys. Acta Gene Regul. Mech. 1819, 784–793. https://doi.org/10.1016/j.bbagrm.2012.02.020.

Ried, T., Just, K.E., Holtgreve-Grez, H., du Manoir, S., Speicher, M.R., Schröck, E., Latham, C., Blegen, H., Zetterberg, A., Cremer, T., 1995. Comparative genomic hybridization of formalin-fixed, paraffin-embedded breast tumors reveals different patterns of chromosomal gains and losses in fibroadenomas and diploid and aneuploid carcinomas. Cancer Res. 55, 5415–5423.

Ried, T., Knutzen, R., Steinbeck, R., Blegen, H., Schröck, E., Heselmeyer, K., du Manoir, S., Auer, G., 1996. Comparative genomic hybridization reveals a specific pattern of chromosomal gains and losses during the genesis of colorectal tumors. Genes Chromosomes Cancer 15, 234–245. https://doi.org/10.1002/(SICI)1098-2264(199604)15:4 < 234::AID-GCC5 > 3.0.CO;2-2.

Ried, T., Lengauer, C., Cremer, T., Wiegant, J., Raap, A.K., van der Ploeg, M., Groitl, P., Lipp, M., 1992. Specific metaphase and interphase detection of the breakpoint region in 8q24 of Burkitt lymphoma cells by triple-color fluorescence in situ hybridization. Genes Chromosomes Cancer 4, 69–74.

Saito, T., Niida, A., Uchi, R., Hirata, H., Komatsu, H., Sakimura, S., Hayashi, S., Nambara, S., Kuroda, Y., Ito, S., Eguchi, H., Masuda, T., Sugimachi, K., Tobo, T., Nishida, H., Daa, T., Chiba, K., Shiraishi, Y., Yoshizato, T., Kodama, M., Okimoto, T., Mizukami, K., Ogawa, R., Okamoto, K., Shuto, M., Fukuda, K., Matsui, Y., Shimamura, T., Hasegawa, T., Doki, Y., Nagayama, S., Yamada, K., Kato, M., Shibata, T., Mori, M., Aburatani, H., Murakami, K., Suzuki, Y., Ogawa, S., Miyano, S., Mimori, K., 2018. A temporal shift of the evolutionary principle shaping intratumor heterogeneity in colorectal cancer. Nat. Commun. 9, 2884. https://doi.org/10.1038/s41467-018-05226-0.

Salari, K., Spulak, M.E., Cuff, J., Forster, A.D., Giacomini, C.P., Huang, S., Ko, M.E., Lin, A.Y., van de Rijn, M., Pollack, J.R., 2012. CDX2 is an amplified lineage-survival oncogene in colorectal cancer. Proc. Natl. Acad. Sci. U.S.A. 109, E3196–E3205. https://doi.org/10.1073/pnas.1206004109.

Sameek, R., Chinnaiyan, A.M., 2014. Translating genomics for precision cancer medicine. Annu. Rev. Genom. Hum. Genet. 15, 395–415. https://doi.org/10.1146/annurev-genom-090413-025552.

Sartore-Bianchi, A., Marsoni, S., Siena, S., 2018. Human epidermal growth factor receptor 2 as a molecular biomarker for metastatic colorectal cancer. JAMA Oncol. 4, 19. https://doi.org/10.1001/jamaoncol.2017.3323.

Sathirapongsasuti, J.F., Lee, H., Horst, B.A.J., Brunner, G., Cochran, A.J., Binder, S., Quackenbush, J., Nelson, S.F., 2011. Exome sequencing-based copy-number variation and loss of heterozygosity detection: ExomeCNV. Bioinformatics 27, 2648–2654. https://doi.org/10.1093/bioinformatics/btr462.

Scartozzi, M., Bearzi, I., Mandolesi, A., Pierantoni, C., Loupakis, F., Zaniboni, A., Negri, F., Quadri, A., Zorzi, F., Galizia, E., Berardi, R., Biscotti, T., Labianca, R., Masi, G., Falcone, A., Cascinu, S., 2009. Epidermal Growth Factor Receptor (EGFR) gene copy number (GCN) correlates with clinical activity of irinotecan-cetuximab in K-RAS wild-type colorectal cancer: a fluorescence in situ (FISH) and chromogenic in situ hybridization (CISH) analysis. BMC Canc. 9, 303. https://doi.org/10.1186/1471-2407-9-303.

Schröck, E., du Manoir, S., Veldman, T., Schoell, B., Wienberg, J., Ferguson-Smith, M.A., Ning, Y., Ledbetter, D.H., Bar-Am, I., Soenksen, D., Garini, Y., Ried, T., 1996. Multicolor spectral karyotyping of human chromosomes. Science 273, 494–497.

Sepulveda, A.R., Hamilton, S.R., Allegra, C.J., Grody, W., Cushman-Vokoun, A.M., Funkhouser, W.K., Kopetz, S.E., Lieu, C., Lindor, N.M., Minsky, B.D., Monzon, F.A., Sargent, D.J., Singh, V.M., Willis, J., Clark, J., Colasacco, C., Rumble, R.B., Temple-Smolkin, R., Ventura, C.B., Nowak, J.A., 2017. Molecular biomarkers for the evaluation of colorectal cancer: guideline from the American society for clinical pathology, college of American pathologists, association for molecular pathology, and the American society of clinical oncology. J. Clin. Oncol. 35, 1453–1486. https://doi.org/10.1200/JCO.2016.71.9807.

Sheffer, M., Bacolod, M.D., Zuk, O., Giardina, S.F., Pincas, H., Barany, F., Paty, P.B., Gerald, W.L., Notterman, D.A., Domany, E., 2009. Association of survival and disease progression with chromosomal instability: a genomic exploration of colorectal cancer. Proc. Natl. Acad. Sci. 106, 7131–7136. https://doi.org/10.1073/pnas.0902232106.

Sheltzer, J.M., Ko, J.H., Replogle, J.M., Habibe Burgos, N.C., Chung, E.S., Meehl, C.M., Sayles, N.M., Passerini, V., Storchova, Z., Amon, A., 2017. Single-chromosome gains commonly function as tumor suppressors. Cancer Cell 31, 240–255. https://doi.org/10.1016/j.ccell.2016.12.004.

Sheltzer, J.M., Torres, E.M., Dunham, M.J., Amon, A., 2012. Transcriptional

consequences of aneuploidy. Proc. Natl. Acad. Sci. U.S.A. 109, 12644–12649. https://doi.org/10.1073/pnas.1209227109.
Shinya, H., Wolff, W.I., 1979. Morphology, anatomic distribution and cancer potential of colonic polyps. Ann. Surg. 190, 679–683.
Sillars-Hardebol, A.H., Carvalho, B., van Engeland, M., Fijneman, R.J., Meijer, G.A., 2012. The adenoma hunt in colorectal cancer screening: defining the target. J. Pathol. 226, 1–6. https://doi.org/10.1002/path.3012.
Smeets, D., Miller, I.S., O'Connor, D.P., Das, S., Moran, B., Boeckx, B., Gaiser, T., Betge, J., Barat, A., Klinger, R., van Grieken, N.C.T., Cremolini, C., Prenen, H., Mazzone, M., Depreeuw, J., Bacon, O., Fender, B., Brady, J., Hennessy, B.T., McNamara, D.A., Kay, E., Verheul, H.M., Maarten, N., Gallagher, W.M., Murphy, V., Prehn, J.H.M., Koopman, M., Punt, C.J.A., Loupakis, F., Ebert, M.P.A., Ylstra, B., Lambrechts, D., Byrne, A.T., 2018. Copy number load predicts outcome of metastatic colorectal cancer patients receiving bevacizumab combination therapy. Nat. Commun. 9, 4112. https://doi.org/10.1038/s41467-018-06567-6.
Speicher, M.R., Ballard, S.G., Ward, D.C., 1996. Karyotyping human chromosomes by combinatorial multi-fluor FISH. Nat. Genet. 12, 368–375. https://doi.org/10.1038/ng0496-368.
Speicher, M.R., du Manoir, S., Schröck, E., Holtgreve-Grez, H., Schoell, B., Lengauer, C., Cremer, T., Ried, T., 1993. Molecular cytogenetic analysis of formalin-fixed, paraffin-embedded solid tumors by comparative genomic hybridization after universal DNA-amplification. Hum. Mol. Genet. 2, 1907–1914.
Steinbeck, R.G., Heselmeyer, K.M., Auer, G.U., 1994. DNA ploidy in human colorectal adenomas. Anal. Quant. Cytol. Histol. 16, 196–202.
Steinbeck, R.G., Heselmeyer, K.M., Neugebauer, W.F., Falkmer, U.G., Auer, G.U., 1993. DNA ploidy in human colorectal adenocarcinomas. Anal. Quant. Cytol. Histol. 15, 187–194.
Stingele, S., Stoehr, G., Peplowska, K., Cox, J., Mann, M., Storchova, Z., 2012. Global analysis of genome, transcriptome and proteome reveals the response to aneuploidy in human cells. Mol. Syst. Biol. 8. https://doi.org/10.1038/msb.2012.40.
Storojeva, I., Boulay, J.-L., Ballabeni, P., Buess, M., Terracciano, L., Laffer, U., Mild, G., Herrmann, R., Rochlitz, C., 2005. Prognostic and predictive relevance of DNAM-1, SOCS6 and CADH-7 genes on chromosome 18q in colorectal cancer. Oncology 68, 246–255. https://doi.org/10.1159/000086781.
Sveen, A., Bruun, J., Eide, P.W., Eilertsen, I.A., Ramirez, L., Murumägi, A., Arjama, M., Danielsen, S.A., Kryeziu, K., Elez, E., Tabernero, J., Guinney, J., Palmer, H.G., Nesbakken, A., Kallioniemi, O., Dienstmann, R., Lothe, R.A., 2018. Colorectal cancer consensus molecular subtypes translated to preclinical models uncover potentially targetable cancer cell dependencies. Clin. Cancer Res. 24, 794–806. https://doi.org/10.1158/1078-0432.CCR-17-1234.
Takahashi, Y., Sheridan, P., Niida, A., Sawada, G., Uchi, R., Mizuno, H., Kurashige, J., Sugimachi, K., Sasaki, S., Shimada, Y., Hase, K., Kusunoki, M., Kudo, S., Watanabe, M., Yamada, K., Sugihara, K., Yamamoto, H., Suzuki, A., Doki, Y., Miyano, S., Mori, M., Mimori, K., 2015. The AURKA/TPX2 axis drives colon tumorigenesis co-operatively with MYC. Ann. Oncol. 26, 935–942. https://doi.org/10.1093/annonc/mdv034.
Talevich, E., Shain, A.H., Botton, T., Bastian, B.C., 2016. CNVkit: genome-wide copy number detection and visualization from targeted DNA sequencing. PLoS Comput. Biol. 12, e1004873. https://doi.org/10.1371/journal.pcbi.1004873.
Torabi, K., Erola, P., Alvarez-Mora, M.I., Díaz-Gay, M., Ferrer, Q., Castells, A., Castellví-Bel, S., Milà, M., Lozano, J.J., Miró, R., Ried, T., Ponsa, I., Camps, J., 2019. Quantitative analysis of somatically acquired and constitutive uniparental disomy in gastrointestinal cancers. Int. J. Cancer 144, 513–524. https://doi.org/10.1002/ijc.31936.
Torabi, K., Miró, R., Fernández-Jiménez, N., Quintanilla, I., Ramos, L., Prat, E., del Rey, J., Pujol, N., Killian, J.K., Meltzer, P.S., Fernández, P.L., Ried, T., Lozano, J.J., Camps, J., Ponsa, I., 2015. Patterns of somatic uniparental disomy identify novel tumor suppressor genes in colorectal cancer. Carcinogenesis 36, 1103–1110. https://doi.org/10.1093/carcin/bgv115.
Torres, E.M., Sokolsky, T., Tucker, C.M., Chan, L.Y., Boselli, M., Dunham, M.J., Amon, A., 2007. Effects of aneuploidy on cellular physiology and cell division in haploid yeast. Science 317, 916–924. https://doi.org/10.1126/science.1142210.
Toyota, M., Ahuja, N., Ohe-Toyota, M., Herman, J.G., Baylin, S.B., Issa, J.P., 1999. CpG island methylator phenotype in colorectal cancer. Proc. Natl. Acad. Sci. U.S.A. 96, 8681–8686.
Trost, B., Walker, S., Wang, Z., Thiruvahindrapuram, B., MacDonald, J.R., Sung, W.W.L., Pereira, S.L., Whitney, J., Chan, A.J.S., Pellecchia, G., Reuter, M.S., Lok, S., Yuen, R.K.C., Marshall, C.R., Merico, D., Scherer, S.W., 2018. A comprehensive workflow for read depth-based identification of copy-number variation from whole-genome sequence data. Am. J. Hum. Genet. 102, 142–155. https://doi.org/10.1016/j.ajhg.2017.12.007.
Tsafrir, D., Bacolod, M., Selvanayagam, Z., Tsafrir, I., Shia, J., Zeng, Z., Liu, H., Krier, C., Stengel, R.F., Barany, F., Gerald, W.L., Paty, P.B., Domany, E., Notterman, D.A., 2006. Relationship of gene expression and chromosomal abnormalities in colorectal cancer. Cancer Res. 66, 2129–2137. https://doi.org/10.1158/0008-5472.CAN-05-2569.
Tuna, M., Knuutila, S., Mills, G.B., 2009. Uniparental disomy in cancer. Trends Mol. Med. 15, 120–128. https://doi.org/10.1016/j.molmed.2009.01.005.
Upender, M.B., Habermann, J.K., McShane, L.M., Korn, E.L., Barrett, J.C., Difilippantonio, M.J., Ried, T., 2004. Chromosome transfer induced aneuploidy results in complex dysregulation of the cellular transcriptome in immortalized and cancer cells. Cancer Res. 64, 6941–6949. https://doi.org/10.1158/0008-5472.CAN-04-0474.
Valtorta, E., Misale, S., Sartore-Bianchi, A., Nagtegaal, I.D., Paraf, F., Lauricella, C., Dimartino, V., Hobor, S., Jacobs, B., Ercolani, C., Lamba, S., Scala, E., Veronese, S., Laurent-Puig, P., Siena, S., Tejpar, S., Mottolese, M., Punt, C.J.A., Gambacorta, M., Bardelli, A., Di Nicolantonio, F., 2013. KRAS gene amplification in colorectal cancer and impact on response to EGFR-targeted therapy. Int. J. Cancer 133, 1259–1265. https://doi.org/10.1002/ijc.28106.
van den Broek, E., den Uil, S.H., Coupé, V.M.H., Delis-van Diemen, P.M., Bolijn, A.S., Bril, H., Stockmann, H.B.A.C., van Grieken, N.C.T., Meijer, G.A., Fijneman, R.J.A., 2018. MACROD2 expression predicts response to 5-FU-based chemotherapy in stage III colon cancer. Oncotarget 9, 29445–29452. https://doi.org/10.18632/oncotarget.25655.
van den Broek, E., Dijkstra, M.J.J., Krijgsman, O., Sie, D., Haan, J.C., Traets, J.J.H., van de Wiel, M.A., Nagtegaal, I.D., Punt, C.J.A., Carvalho, B., Ylstra, B., Abeln, S., Meijer, G.A., Fijneman, R.J.A., 2015. High prevalence and clinical relevance of genes affected by chromosomal breaks in colorectal cancer. PLoS One 10, e0138141. https://doi.org/10.1371/journal.pone.0138141.
van Dijk, E., Biesma, H.D., Cordes, M., Smeets, D., Neerincx, M., Das, S., Eijk, P.P., Murphy, V., Barat, A., Bacon, O., Prehn, J.H.M., Betge, J., Gaiser, T., Fender, B., Meijer, G.A., McNamara, D.A., Klinger, R., Koopman, M., Ebert, M.P.A., Kay, E.W., Hennessey, B.T., Verheul, H.M.W., Gallagher, W.M., O'Connor, D.P., Punt, C.J.A., Loupakis, F., Lambrechts, D., Byrne, A.T., van Grieken, N.C.T., Ylstra, B., 2018. Loss of chromosome 18q11.2-q12.1 is predictive for survival in patients with metastatic colorectal cancer treated with bevacizumab. J. Clin. Oncol. 36, 2052–2060. https://doi.org/10.1200/JCO.2017.77.1782.
Varma, S., Pommier, Y., Sunshine, M., Weinstein, J.N., Reinhold, W.C., 2014. High resolution copy number variation data in the NCI-60 cancer cell lines from whole genome microarrays accessible through CellMiner. PLoS One 9, e92047. https://doi.org/10.1371/journal.pone.0092047.
Voorham, Q.J.M., Carvalho, B., Spiertz, A.J., van Grieken, N.C.T., Mongera, S., Rondagh, E.J.A., van de Wiel, M.A., Jordanova, E.S., Ylstra, B., Kliment, M., Grabsch, H., Rembacken, B.J., Arai, T., de Bruine, A.P., Sanduleanu, S., Quirke, P., Mulder, C.J.J., van Engeland, M., Meijer, G.A., 2012. Chromosome 5q loss in colorectal flat adenomas. Clin. Cancer Res. 18, 4560–4569. https://doi.org/10.1158/1078-0432.CCR-11-2385.
Wagner, A.K., Kadri, N., Snäll, J., Brodin, P., Gilfillan, S., Colonna, M., Bernhardt, G., Höglund, P., Kärre, K., Chambers, B.J., 2017. Expression of CD226 is associated to but not required for NK cell education. Nat. Commun. 8, 15627. https://doi.org/10.1038/ncomms15627.
Wangsa, D., Braun, R., Stuelten, C.H., Brown, M., Bauer, K.M., Emons, G., Weston, L.A., Hu, Y., Yang, H.H., Vila-Casadesús, M., Lee, M.P., Brauer, P., Warner, L., Upender, M., Hummon, A.B., Camps, J., Ried, T., 2019. Induced chromosomal aneuploidy results in global and consistent deregulation of the transcriptome of cancer cells. Neoplasia 21, 721–729. https://doi.org/10.1016/j.neo.2019.04.009.
Watson, R.G., Muhale, F., Thorne, L.B., Yu, J., O'Neil, B.H., Hoskins, J.M., Meyers, M.O., Deal, A.M., Ibrahim, J.G., Hudson, M.L., Walko, C.M., McLeod, H.L., Auman, J.T., 2010. Amplification of thymidylate synthetase in metastatic colorectal cancer patients pretreated with 5-fluorouracil-based chemotherapy. Eur. J. Cancer 46, 3358–3364. https://doi.org/10.1016/j.ejca.2010.07.011.
Williams, B.R., Prabhu, V.R., Hunter, K.E., Glazier, C.M., Whittaker, C.A., Housman, D.E., Amon, A., 2008. Aneuploidy affects proliferation and spontaneous immortalization in mammalian cells. Science 80 (322), 703–709. https://doi.org/10.1126/science.1160058.
Winawer, S.J., Zauber, A.G., Ho, M.N., O'Brien, M.J., Gottlieb, L.S., Sternberg, S.S., Waye, J.D., Schapiro, M., Bond, J.H., Panish, J.F., Ackroyd, F., Shike, M., Kurtz, R.C., Hornsby-Lewis, L., Gerdes, H., Stewart, E.T., 1993. Prevention of colorectal cancer by colonoscopic polypectomy. N. Engl. J. Med. 329, 1977–1981. https://doi.org/10.1056/NEJM199312303292701.
Wolf, M., Mousses, S., Hautaniemi, S., Karhu, R., Huusko, P., Allinen, M., Elkahloun, A., Monni, O., Chen, Y., Kallioniemi, A., Kallioniemi, O.-P., 2004. High-resolution analysis of gene copy number alterations in human prostate cancer using CGH on cDNA microarrays: impact of copy number on gene expression. Neoplasia 6, 240–247. https://doi.org/10.1593/neo.3439.
Xie, C., Tammi, M.T., 2009. CNV-seq, a new method to detect copy number variation using high-throughput sequencing. BMC Bioinf. 10, 80. https://doi.org/10.1186/1471-2105-10-80.
Xie, T., Cho, Y.B., Wang, K., Huang, D., Hong, H.K., Choi, Y.-L., Ko, Y.H., Nam, D.-H., Jin, J., Yang, H., Fernandez, J., Deng, S., Rejto, P.A., Lee, W.Y., Mao, M., 2014. Patterns of somatic alterations between matched primary and metastatic colorectal tumors characterized by whole-genome sequencing. Genomics 104, 234–241. https://doi.org/10.1016/j.ygeno.2014.07.012.
Xie, T., D'Ario, G., Lamb, J.R., Martin, E., Wang, K., Tejpar, S., Delorenzi, M., Bosman, F.T., Roth, A.D., Yan, P., Bougel, S., Di Narzo, A.F., Popovici, V., Budinská, E., Mao, M., Weinrich, S.L., Rejto, P.A., Hodgson, J.G., 2012. A comprehensive characterization of genome-wide copy number aberrations in colorectal cancer reveals novel oncogenes and patterns of alterations. PLoS One 7, e42001. https://doi.org/10.1371/journal.pone.0042001.
Yaeger, R., Yao, Z., Hyman, D.M., Hechtman, J.F., Vakiani, E., Zhao, H., Su, W., Wang, L., Joelson, A., Cercek, A., Baselga, J., de Stanchina, E., Saltz, L., Berger, M.F., Solit, D.B., Rosen, N., 2017. Mechanisms of acquired resistance to BRAF V600E inhibition in colon cancers converge on RAF dimerization and are sensitive to its inhibition. Cancer Res. 77, 6513–6523. https://doi.org/10.1158/0008-5472.CAN-17-0768.
Yan, D., Yi, Song, Chiu, W.C., Qin, L.G., Kin, W.H., Hung, C.T.K., Linxiao, H., Wai, C.K., Yi, Sui, Tao, Y., Tao, T., 2017. Integrated analysis of chromosome copy number variation and gene expression in cervical carcinoma. Oncotarget 8. https://doi.org/10.18632/oncotarget.22403.
Ye, K., Schulz, M.H., Long, Q., Apweiler, R., Ning, Z., 2009. Pindel: a pattern growth

approach to detect break points of large deletions and medium sized insertions from paired-end short reads. Bioinformatics 25, 2865–2871. https://doi.org/10.1093/bioinformatics/btp394.

Yokota, T., Ura, T., Shibata, N., Takahari, D., Shitara, K., Nomura, M., Kondo, C., Mizota, A., Utsunomiya, S., Muro, K., Yatabe, Y., 2011. BRAF mutation is a powerful prognostic factor in advanced and recurrent colorectal cancer. Br. J. Canc. 104, 856–862. https://doi.org/10.1038/bjc.2011.19.

Yoon, S., Xuan, Z., Makarov, V., Ye, K., Sebat, J., 2009. Sensitive and accurate detection of copy number variants using read depth of coverage. Genome Res. 19, 1586–1592. https://doi.org/10.1101/gr.092981.109.

Zare, F., Dow, M., Monteleone, N., Hosny, A., Nabavi, S., 2017. An evaluation of copy number variation detection tools for cancer using whole exome sequencing data. BMC Bioinf. 18, 286. https://doi.org/10.1186/s12859-017-1705-x.

Zhou, W., Goodman, S.N., Galizia, G., Lieto, E., Ferraraccio, F., Pignatelli, C., Purdie, C.A., Piris, J., Morris, R., Harrison, D.J., Paty, P.B., Culliford, A., Romans, K.E., Montgomery, E.A., Choti, M.A., Kinzler, K.W., Vogelstein, B., 2002. Counting alleles to predict recurrence of early-stage colorectal cancers. Lancet (London, England) 359, 219–225. https://doi.org/10.1016/S0140-6736(02)07448-2.