Molecular Pathology of Gastric Carcinoma

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Abstract

Gastric carcinoma (GC) is a biologically heterogeneous disease involving numerous genetic and epigenetic alterations. A very small proportion of GCs can be caused by a specific germ-line mutation of the E-cadherin gene (CDH1). Sporadic GC is developed through multistep processes that begin with Helicobacter pylori-induced atrophic gastritis. Epstein-Barr virus is another infectious cause of GC, and the above two infection-associated GCs are characterized by global CpG island methylation in the promoter region of cancer-related genes. Mutations of tumor protein p53 (TP53) and β-catenin (CTNNB1) genes occur early in the development of GC and contribute to gastric carcinogenesis. Furthermore, significant numbers of GCs show loss of Runx3 due to hemizygous deletion and hypermethylation of the promoter region. Aberrant Cdx2 expression has been shown in preneoplastic lesions as well as GC. However, it remains unclear whether Cdx2 plays an oncogenic role in gastric carcinogenesis. GC with microsatellite instability is also a well-defined subset exhibiting distinctive clinicopathologic features.

Introduction

Gastric carcinoma (GC) is one of the most common malignant tumors and the second most common cause of cancer death worldwide although there has been a steady decline in the incidence and mortality risk of GC over several decades in most countries [1]. There are large geographical variations, and the incidence and death rate of GC are more than twice as high in Asian/Pacific Islanders as in Whites [2]. Despite the tremendous improvements in diagnosis and treatment technologies, the prognosis of advanced GC remains poor and the survival of affected patients is less than 40% even after a curative surgical resection and adjuvant therapy [3].

GC is considered as a multifactorial disease since many inherited and environmental factors play a role in gastric carcinogenesis, including the host’s genetic background, infectious agents and dietary habits. Chronic atrophic gastritis has been known to be the most important and the greatest risk factor for GC. Helicobacter pylori infection, classified by the WHO as a class I carcinogen since 1994 [4], is the most common cause of chronic gastritis and therefore has been extensively studied.
GC is a heterogeneous disorder that can be divided into at least two main histological types based on the Lauren classification: intestinal and diffuse type. These two types have distinct morphological, clinical and epidemiological features. Intestinal-type GC is more frequently observed in older patients and follows multifocal atrophic gastritis, which is accompanied by intestinal metaplasia or dysplasia. On the other hand, diffuse GC occurs more commonly in young patients and its links with atrophic gastritis or intestinal metaplasia are poor, or do not exist. These differences might represent a different molecular mechanism of tumor development and progression. For example, microsatellite instability and promoter hypermethylation are more commonly found in intestinal-type than in diffuse-type GC while specific genes, such as CDH1, are more often hypermethylated in diffuse-type GC [5]. GC can be also classified into four phenotypes according to mucin (MUC1, MUC2 and CD10) expression: G-type (gastric or foveolar phenotype), I-type (intestinal phenotype), GI-type (intestinal and gastric mixed phenotype) and N-type (neither gastric nor intestinal phenotype) GC. Some distinct genetic changes in GC have been revealed to be associated with the mucin phenotypic expression. For instance, TP53 mutations are more common in I-type than in G-type GC, whereas microsatellite instability is more frequent in G-type than in I-type GC. In addition, some specific epigenetic alterations are also known to be involved in distinct mucin expression of GC. Indeed, methylation of hMLH1 occurred more frequently in MUC2-negative GC, whereas MGMT was more frequently methylated in MUC2-positive GC than in MUC2-negative GC [6].

With the advances in the molecular genetics of cancer, it has been revealed that carcinogenesis is a multistep process involving alterations of multiple genes, such as point mutation, recombination, amplification, and/or deletion. Recent evidence has established that epigenetic modifications play a crucial role in the carcinogenesis as well. Especially transcriptional silencing by promoter hypermethylation is now considered as an important mechanism of functional loss of tumor suppressor genes. Despite the increasing body of knowledge about molecular mechanisms of gastric carcinogenesis, the overall view on the molecular pathology of GC remains fragmentary. In this review, we describe the major molecular events and epigenetic changes involved in gastric carcinogenesis and progression.

**E-Cadherin Gene (CDH1)**

E-cadherin is a cell adhesion protein that is expressed at the adherence junctions of epithelial tissue and required for development, cell differentiation and maintenance of epithelial architecture. Downregulation of E-cadherin is commonly observed in many sporadic tumors, and its loss during tumor progression has led to a concept of E-cadherin acting as a suppressor of tumor invasion and metastasis [7]. On the other hand, germ-line mutations in the CDH1 gene result in a cancer syndrome, a hereditary diffuse gastric cancer (HDGC), which comprises approximately 1–3% of all GCs [8]. Until now, more than 100 different germ-line mutations of CDH1 have been described in HDGC families [9]. Carriers of CDH1 germ-line mutations have a 70% lifetime risk of advanced diffuse gastric cancer [9] and tend to present with several hundred microscopic foci of stage T1a intramucosal signet ring cell carcinoma, which are termed ‘early HDGC’ (eHDGC) [10]. The tumor cells in multifocal eHDGC show very low E-cadherin expression, implying that the wild-type CDH1 allele is also suppressed or lost in tumor cells. Studies have revealed that the second hit is caused by promoter hypermethylation of CDH1 in at least 50% of cases [11]. The methylation patterns observed in eHDGCs are monoallelic and specific for each focus, indicating that each eHDGC has an independent monoclonal origin and that the epigenetic silencing of CDH1 by promoter methylation is an early event in tumor development [12]. Remarkably, promoter methylation has also been identified as a major mechanism underlying E-cadherin downregulation in sporadic diffuse gastric cancers [13]. Of interest, eHDGCs have unique features, i.e. they are hypoproliferative and lack Wnt pathway activation [11]. Lineage-labeling experiments with gastric differentiation markers have demonstrated that eHDGCs develop from the mucous neck region where gastric stem cells or progenitor cells exist [12]. Thus, it is tempting to hypothesize that perturbed stem cell division contributes to malignant transformation initiated by loss of adhesion and polarity [11]. Together, these findings suggest that E-cadherin deficiency can initiate diffuse GC via distinct pathways that do not require any growth advantages.

**H. pylori Infection**

Chronic atrophic gastritis and intestinal metaplasia are histological precursors of intestinal-type GC. Multistep gastric carcinogenesis includes a sequence of events
that begins with H. pylori-induced superficial gastritis, progressing towards chronic atrophic gastritis, intestinal metaplasia, dysplasia and finally GC [14]. The causal relationship between H. pylori and chronic gastritis has been confirmed by the results of interventional studies showing that bacterial eradication leads to the regression of precursor lesions [15]. It is well known that persistent inflammation induces increased tissue turnover, which predisposes to an excessive rate of proliferation, and in many cases results in more frequent mitotic errors and an increased rate of genetic mutation [8]. In addition, chronic inflammation causes genetic instability through the generation of reactive oxygen species and reactive nitrogen species which can directly damage the genomic and mitochondrial DNA [16].

It is now clear that bacterial virulence factors are important determinants that have been implicated in the development of GC. The most studied virulence factor of H. pylori is cagA protein. Infection with a cagA-positive H. pylori strain in comparison with a cagA-negative strain increases the risk for development of GC [17]. Once translocated into host cells, cagA induces a growth factor-like response in gastric epithelial cells by forming a physical complex with the Src homology 2 domain (SH2)-containing tyrosine phosphatase (SHP-2) in a phosphorylation-dependent manner [18]. Notably, it has also been reported that cagA-positive H. pylori infection of the gastric epithelium triggered aberrant expression of activation-induced cytidine deaminase (AICDA), a gene originally linked to immunoglobulin class switching and B lymphocyte hypermutation, which resulted in the accumulation of nucleotide alterations in the TP53 tumor suppressor genes [19]. Therefore, it was hypothesized that aberrant AICDA expression caused by H. pylori infection might be a mechanism of mutation accumulation in the gastric mucosa during H. pylori-associated gastric carcinogenesis. Another virulence gene involved in gastric carcinogenesis is the vacuolating cytotoxin (vacA), which induces gastric epithelial cell apoptosis and also suppresses local immune response by interfering with T cell activation [20].

Host genetic factors have also been postulated to be involved in gastric carcinogenesis because only a small percentage (3%) of infected individuals ultimately develops GC. The genetic polymorphism of interleukin-1β, a proinflammatory cytokine gene, was studied in GC patients. While some studies demonstrated that the most proinflammatory genotypes of interleukin-1β are positively associated with GC [21]; other reports do not support these results, which might be explained by the population-specific cancer risks [22]. A polymorphism of interleukin 1 receptor antagonist (IL1RN), IL10 and TNF genes was also identified to be associated with increased risk for GC, which makes it possible to define a specific genetic profile associated with the highest risk for GC [23]. Furthermore, when combined with bacterial genotyping, the host’s genetic profile would provide a clinical tool to identify patients at high risk for GC [24].

Epstein-Barr Virus

Epstein-Barr virus (EBV) is a ubiquitous human herpesvirus that has well-established associations with a variety of malignant neoplasms, such as Burkitt’s lymphoma, extranodal NK/T cell lymphoma, nasopharyngeal carcinoma and GC. EBV was therefore classed as a group I carcinogen by the IARC and WHO [25]. EBV-associated GC is the result of monoclonal proliferation of EBV-infected epithelial cells and accounts for about 5% of GC [26]. EBV stably maintains its latent infection in carcinoma cells and expresses viral latent genes which belong to the latency I program: EBV-determined nuclear antigen 1 (EBNA1), EBV-encoded small RNA (EBER), latent membrane protein 2A (LMP2A) and BamHI-A rightward transcripts (BARTs) [27]. EBV-associated GC is a specific clinicopathologic subset with characteristics of younger age, male predominance, proximal location, lower rate of lymph node involvement, marked lymphocytic infiltration, and lace pattern within the mucosa [26]. EBV-positive GC also showed a distinct protein expression profile featured by frequent loss of p16 (CDKN2A), smad4, Fhit, and CD82 (KAI-1) compared to EBV-negative carcinomas, but they retained the expression of adenomatous polyposis coli (APC), deleted in colorectal cancer (DCC), and DNA repair proteins [28].

Most of the EBV-positive GCs exhibit the high CpG island methylator phenotype, thus global CpG island methylation in the promoter region of cancer-related genes is now considered as the most characteristic abnormality in EBV-associated GC [29]. Although the exact mechanism of global hypermethylation by EBV infection remains to be elucidated, viral LMP2A was demonstrated to be responsible for aberrant hypermethylation by activation of host DNA methyltransferase 1, which causes Pten loss through CpG island methylation of the PTEN promoter in EBV-associated GC [30]. LMP2A also up-regulates Birc5 (survivin) expression through the activation of NF-κB [31] and activates extracellular signal-regulated kinases (ERK/MAPK1) [32]. Furthermore, LMP2A inhibits TGF-β-induced apoptosis in a GC cell line.
line through activation of the Ras/PI3K/Akt pathway [33]. These findings suggest that LMP2A is an oncogenic molecule that provides a selective advantage for EBV-infected epithelial cells, which may eventually lead to development of EBV-associated GC.

**Tumor Protein p53 (TP53)**

Tumor protein p53 gene (TP53) is the most commonly mutated gene in human tumors; it acts as a tumor suppressor gene that induces cell cycle arrest and apoptosis. Approximately 50% of all cancers involve missense mutations of one p53 allele coupled with a deletion of the second allele, which lead to complete loss of p53 function of one p53 allele coupled with a deletion of the second allele that induces cell cycle arrest and apoptosis. Apoptosis is a process by which DNA is fragmented and caspases are activated in response to a variety of stimuli, including DNA damage. The p53 protein is a transcription factor that induces the expression of genes that promote cell cycle arrest and apoptosis, thereby preventing the propagation of damaged cells. Mutations in p53 can lead to uncontrolled cell proliferation and the development of cancer.

The incidence of TP53 mutations in invasive GC ranges from 0 to 76.9%, and the mutational spectrum of TP53 in GC is very wide [36]. There are several sites where mutation occurs more commonly than in others; they include codons 175, 248, 273, 282, 245, and 213. Of interest is the fact that they are all CpG sites. Transition of G:C to A:T at CpG sites is the most common type of mutation irrespective of the histologic type of GC [36]. More than one mutation can be present in a single tumor [37], and there can be heterogeneity of the p53 mutational status within a given tumor [38]. Young patients have a lower incidence of TP53 mutations than older ones, and advanced GC tends to have a higher incidence of mutations. Furthermore, TP53 mutations occur much more commonly in tumors arising in the cardia than in tumors in the antrum, and they are more common in metastatic than in primary GC [36].

It appears that TP53 alterations occur early in the development of GC because they are frequently found in precancerous lesions. For example, point mutations of TP53 have been demonstrated in 52% of H. pylori-associated gastritis even though they were located in non-hot spot codons [39]. Indeed, it has recently been revealed that aberrant AICDA expression induced by H. pylori infection can cause TP53 mutations. Helicobacter infection in TP53 knockout mice resulted in the development of dysplastic lesions, whereas these infections in normal mice failed to produce any pre-neoplastic changes, which showed that H. pylori infection and p53 may act in a synergistic fashion in gastric carcinogenesis [36]. TP53 mutations were also found in 37.5% of intestinal metaplasia and 58% of the dysplastic lesions [40]. Interestingly, while silent mutations tend to be observed in adenomas with mild or moderate degrees of dysplasia, missense mutations were found in adenomas with high-grade dysplasia, suggesting that the presence of missense mutations of TP53 in adenomas might be a key indicator of malignant transformation [41].

**β-Catenin Gene (CTNNB1)**

The Wnt signaling pathway, initially considered to play a crucial role in embryonic development, is also known to play an important role in cancer development, and β-catenin, functioning in cadherin-based epithelial cell adhesion, is a key regulator of Wnt signaling. Elevation of cytoplasmic β-catenin level can occur by the binding with Wnt ligands or by mutations in APC, AXIN1 or CTNNB1. Interestingly, mutation of CTNNB1, which encodes β-catenin, seems to be exclusive to the mutations that inactivate Apc protein. Oncogenic mutations involving the amino-terminal region of β-catenin make it refractory to regulation by Apc [42]. Accumulation of β-catenin in the cytoplasm results in its binding to members of the Tcf/Lef family of transcription factors and its nuclear translocation, where the Tcf/β-catenin complex activates target genes such as MYC and cyclin D1 gene (CCND1) [42]. Indeed, the vast majority of colorectal tumors contain APC mutations, but the overall frequency of CTNNB1 mutations is lower [42]. On the other hand, a significant percentage of gastric tumors has either CTNNB1 or APC mutations. The incidence of mutations in intestinal- versus diffuse-type GC remains unclear. Park et al. [43] reported no mutations in diffuse-type GC, but found that 27% of intestinal-type tumors carried a mutation. Whereas, Clements et al. [44] found that 26% of tumors with β-catenin nuclear staining contained CTNNB1 mutations, but did not identify any difference between diffuse- and intestinal-type GC. Therefore, further studies are necessary to reveal the exact contribution of Wnt pathway activation in gastric carcinogenesis. Selective targeting of gastric cancer cells with the activated β-catenin pathway showed a synergistic effect with chemotherapy, which could be used in practically all transformed cells with an active β-catenin/Tcf [45].

**Runt-Related Transcription Factor 3 (RUNX3)**

Runx3 binds DNA with the core-binding factor β-subunit (CBFB), and activates or represses the principal regulators of growth, survival and differentiation pathways. RUNX3 is now accepted as a tumor suppressor gene in a wide range of invasive and preinvasive epithelial and...
mesenchymal tumors [46]. Its suppressive activity was first reported in gastric epithelial cells of RUNX3 knockout mice in 2002 [47]. Gastric mucosa of RUNX3-null mice exhibits enhanced proliferation, suppressed apoptosis and reduced sensitivity to transforming growth factor-β (TGF-β). About 45–60% of human GCs showed loss of Runx3 expression due to hemizygous deletion and hypermethylation of the promoter region. H. pylori infection as well as precursor lesions such as intestinal metaplasia and gastric adenoma also showed RUNX3 hypermethylation, indicating a role for RUNX3 in gastric carcinogenesis [47, 48]. RUNX3 is a downstream effector of the TGF-β signaling pathway. In response to TGF-β, Runx3 inhibits gastric epithelial proliferation by inducing the CDKN1A (p21) gene, which indicates that the tumor suppressor activity of Runx3 is at least partially associated with its ability to induce p21 expression [49]. Furthermore, Runx3 also upregulates the expression of pro-apoptotic gene BCL2L11 (Bim) in gastric cancer cells treated with TGF-β. Bcl2l11 was downregulated in the gastric epithelium of RUNX3 knockout mice, and the BCL2L11 promoter contained conserved Runx3 binding elements. Together, these findings suggested the critical role of Runx3 in transcriptional upregulation of Bcl2l11 in TGF-β-induced apoptosis [50]. In addition to antiproliferative and apoptotic effects contributing to gastric carcinogenesis, Runx3 affected the progression and metastasis of GCs as well. For example, restoration of Runx3 strongly inhibited peritoneal metastases of GC in an animal model [51], and inhibited the expression of vascular endothelial growth factor A (VEGFA) and suppressed the angiogenesis, growth, and metastasis of GCs [52].

**Caudal Type Homeobox 2 (CDX2)**

CDX2 is a gene for an intestine-specific transcription factor to direct intestinal development, differentiation and maintenance of the intestinal phenotype. Cdx2 not only stimulates proliferation and differentiation of intestinal epithelial cells by transcriptional activation of intestine-specific genes such as mucin precursor 2 (MUC2), sucrase-isomaltase (SI), and carbonic anhydrase 1 (CA1) but also inhibits growth through activation of p21 (CDKN1A), a cyclin-dependent kinase inhibitor [53, 54]. Previous observations suggested a tumor-suppressive role for Cdx2 in colon carcinogenesis. Whereas Cdx2 is normally expressed in intestinal mucosa, gastric epithelial cells do not express Cdx2. In contrast, gastric mucosa undergoing intestinal metaplasia consistently shows ectopic Cdx2 expression, which suggested that Cdx2 plays a role in the development of intestinal metaplasia and subsequent gastric carcinogenesis [55]. Indeed, it has been demonstrated that gastric Cdx2 expression alone was sufficient to induce intestinal metaplasia in mice [56], and long-standing intestinal metaplasia induces invasive GC in CDX2 transgenic mice [57]. In addition, Cdx2 expression was increased in high-grade dysplasia and intestinal-type gastric adenocarcinomas compared to low-grade dysplasia [58]. Taken together, these findings suggest that Cdx2 expression in GC may contribute to the progression of gastric carcinogenesis and that its activation may represent an early event. However, some contradictory studies have shown that Cdx2 expression was progressively reduced in gastric dysplasia and cancer [59, 60], and Cdx2-positive GCs had a significantly better outcome than Cdx2-negative GCs [61]. Furthermore, overexpression of Cdx2 significantly inhibited cell growth and reduced the motility and invasion of cancer cells in vitro, which supported the notion that Cdx2 plays a similar tumor-suppressive role in GC as in colorectal carcinoma [62]. Although the differences in cutoff values to define Cdx2 positivity or the lack of subtyping criteria for gastric epithelial dysplasia may in part explain these conflicting results [60], further studies are required to clarify the exact role of Cdx2 in gastric carcinogenesis.

**Microsatellite Instability**

Microsatellites are repeating sequences of 1–6 base pairs of DNA throughout the genome, and microsatellite instability is defined as length changes of these microsatellites caused by impairment of the DNA mismatch repair system. Microsatellite instability has been found in many sporadic carcinomas as well as in hereditary nonpolyposis colorectal cancer, a syndrome in which germ-line mutations of the DNA mismatch repair genes, MSH2 or MLH1, are present [63]. Whereas microsatellite instability in patients with hereditary nonpolyposis colorectal cancer mainly results from germ-line mutations of one or several DNA mismatch repair genes, somatic mutations of MSH2 and MLH1 are rare in sporadic carcinomas even in GC with microsatellite instability [64]. Many studies suggest that hypermethylation in the promoter CpG island of MLH1 gene is responsible for protein downregulation and subsequent microsatellite instability in sporadic GC [65, 66]. Microsatellite instability has been observed in GC where it ranged from 9.5 to 44%, depending on the group of cases and the number of markers examined [67–71].
Notably, GC associated with a gastric adenoma showed a higher frequency of the microsatellite instability phenotype compared to those carcinomas without an adenoma [72]. GC with microsatellite instability is a well-defined subset of GCs exhibiting distinctive clinicopathologic features, such as antral location, intestinal type, expanding growth pattern, lower prevalence of lymph node metastases, and improved survival. They display frequent frameshift mutations of TGF-β type II receptor (TGFBRII) (90.3%), BAX (61.3%), hMSH3 (38.7%), and E2F4 (61.3%) genes [70], which have microsatellite sequences in the coding region. These mutational events occur in one or two alleles of these genes, and accumulation of mutation may also extend to genes involved in genome integrity including the mismatch repair genes, suggesting that these mutations result in malignant transformation [73].

**ERBB2 Gene**

Erbb2 alteration plays an important role in the development and progression of many epithelial tumors. ERBB2 amplification is particularly frequent, especially in breast cancer, and treatment with trastuzumab, a monoclonal antibody to Erbb2 protein, has been shown to be highly effective in ERBB2-amplified breast cancer [74]. ERBB2 gene amplification and overexpression of Erbb2 protein in GC were also found in a large number of studies. However, the results are inconsistent and ranged from 4 to 16% when performed by FISH [75–77]. Interestingly, the amplification of ERBB2 is more common in intestinal-type GC than in diffuse-type GC [75]. In some studies, GC with ERBB2 amplification was significantly associated with poor outcome [77, 78]. On the other hand, there are also several studies documenting that amplification was unrelated to prognosis [75, 76]. Combined treatment with Erbb2-targeting agents and chemotherapeutic agents for gastric cancer with Erbb2 overexpression exhibited a synergistic antitumor effect in vitro [79]. And there are case reports stating that treatment with trastuzumab showed dramatic antitumor effects in patients with metastatic GCs [80, 81]. Although it remains controversial whether ERBB2 amplification in gastric cancer is homogenous in a tumor or within primary tumors and its metastases [75, 82, 83], trastuzumab may be an attractive option in ERBB2-amplified gastric cancers. Indeed, preliminary results of phase III clinical trials showed the positive effect on overall survival of trastuzumab in combination with chemotherapy compared with chemotherapy alone in patients with Erbb2-positive advanced GC (ToGA, protocol BO18255). In addition, new therapeutic agents with multiple targets that include blocking the Erbb2 pathway are emerging [84].

**Epigenetic Alterations**

Epigenetic mechanisms refer to DNA methylation and histone modifications that result in altered gene expression without changing the coding sequence of the gene. Growing evidence strongly supports that epigenetic dysregulation plays an essential role both independently and cooperatively in tumor initiation and progression. The best characterized mechanisms are transcriptional silencing events that are associated with DNA hypermethylation at promoter regions of genes that regulate important cell functions. Promoter methylation affects virtually all of the pathways in the cellular network, such as DNA repair, cell cycle and apoptosis. It is well established that numerous tumor suppressor genes can be silenced through promoter CpG island methylation during carcinogenesis, and aberrant DNA methylation is the most common molecular lesion of cancer cells. Neither gene mutations nor cytogenetic abnormalities are as common in human tumors as DNA methylation alterations [85]. Thus, aberrant CpG island methylation can be used as a biomarker of malignant cells and as a predictor of their prognosis. In particular, the reversibility of epigenetic changes has made them attractive targets for cancer treatment with modulators that demethylate DNA and inhibit histone deacetylases, leading to reactivation of silenced genes.

Gastric cancer is one of the tumors with a high frequency of aberrant methylation, and it frequently shows the CpG island methylator phenotype [86]. A large number of genes that are suppressed by CpG island hypermethylation have been reported in GC, involving tumor suppressor, cell cycle regulator, apoptosis, invasion-related and DNA mismatch repair genes [87]. In particular, a number of genes, such as CDKN2A (p16), CDK2AP2 (p14), CDH1 (E-cadherin), MGMT (O6-methylguanine DNA methyltransferase), RASSF1, RUNX3, and DLC1, were frequently hypermethylated in GC [87]. The list of epigenetically silenced genes in GC is expected to grow even more in the future.

*H. pylori* and EBV infection have been shown to be closely associated with various degrees of methylation of CpG islands, which could contribute to gastric carcinogenesis. Methylation of tumor suppressor or tumor-related genes in precancerous lesions has also been investigated to evaluate whether the methylation changes contribute to tu-
In summary, aberrant methylation was frequently detected in gastric intestinal metaplasia of both cancer and noncancer patients. The methylation frequency of several genes was higher in GC than in intestinal metaplasia [88]. One study with a large sample collection of chronic gastritis, intestinal metaplasia, gastric adenoma and GC demonstrated that aberrant methylation occurred in early stages and tended to accumulate along the multistep gastric carcinogenesis [89]. The above findings suggest that progressive epigenetic changes play an important role during the progression of a premalignant lesion to cancer.

Most epigenetic studies of GC have focused on aberrant methylation in a single or in several genes, but recently a genome-wide search has been tried to identify novel methylation-silenced genes in gastric cancer [90, 91]. After treatment with a demethylating and/or deacetylating agent of the GC cell line, upregulated genes were screened for epigenetically silenced genes using oligonucleotide microarrays. TFPI2 was found to be highly methylated (81%) in GC, and its methylation was a significant and independent prognostic indicator in GC [90]. Most recently, a genome-wide DNA methylation analysis using a methylation microarray has been applied to directly measure the methylation level of the CpG islands throughout the genome and it has been suggested that this high-throughput method would be greatly helpful to find the novel methylation markers [92].

Conclusion

Cancer is widely considered as a heterogeneous group of diseases with markedly different biological properties that are attributed to a series of clonally selected genetic and epigenetic alterations in tumor suppressor genes and oncogenes [93]. Thus, identifying the characteristics of individual cancers from the point of view of molecular pathology has a great potential for future diagnosis and targeted therapy of cancer. Multiple genetic and epigenetic changes in oncogenes and tumor suppressor genes, cell cycle regulators, cell adhesion molecules and DNA repair genes have been demonstrated to be implicated in gastric carcinogenesis. Based on this emerging understanding of the molecular pathways, several targeted therapies, such as small-molecular inhibitors and antiangiogenic agents, are currently being evaluated in GC treatment [94]. In addition, the analysis of hypermethylation of cell cycle genes or DNA repair genes such as CDKN2A or hMLH1 in nonneoplastic gastric mucosa could be used to predict the risk of malignancy. Even though the complex inherent molecular heterogeneity of GC still remains to be clarified, genome-wide analysis techniques will also help us understand the molecular features of GC, which would provide further novel opportunities in the treatment of GC.

References

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