OPINION

CpG island methylator phenotype in cancer

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Abstract | DNA hypermethylation in CpG-rich promoters is now recognized as a common feature of human neoplasia. However, the pathophysiology of hypermethylation (why, when, where) remains obscure. Cancers can be classified according to their degree of methylation, and those cancers with high degrees of methylation (the CpG island methylator phenotype, or CIMP) represent a clinically and aetiologically distinct group that is characterized by 'epigenetic instability'. Furthermore, CIMP-associated cancers seem to have a distinct epidemiology, a distinct histology, distinct precursor lesions and distinct molecular features.

DNA methylation is a biochemical modification that, in human cells, primarily affects cytosines when they are part of the symmetrical dinucleotide CpG[1,2]. Cytosine methylation has long been a challenging scientific puzzle. Although it is nearly ubiquitous among multicellular organisms, two important biological models — *Saccharomyces cerevisiae* and adult *Drosophila melanogaster* — have undetectable levels of DNA methylation. In mammals, DNA methylation is essential for normal development[3], but its evolutionary *raison d'être* remains controversial. A commonly held hypothesis is that DNA methylation originally evolved to silence repetitive elements[4], and that this silencing property has also been put to use in other situations where transcriptional silencing is required, such as imprinting (a process whereby one of the two alleles of a gene are permanently inactivated, depending on which parent that allele was inherited from) and X-chromosome inactivation. Genetic anomalies in DNA methylation (or associated mechanisms) leads to various developmental diseases in humans[5].

Methylation in development and cancer

Research into DNA methylation has been progressing at a furious pace, despite uncertainty about its origin and physiological function. This much is known: most CpG sites have been lost from mammalian genomes during evolution, but about 1% of human DNA consists of short areas where CpG sites have escaped depletion[1,2]. Most of the remaining CpG sites are normally methylated in adult cells. About half of all genes have a CpG island in their promoter region, and this gene configuration is what has recently attracted the most attention. Most promoter CpG islands are normally unmethylated, regardless of the expression state of the associated gene. However, in silenced areas, such as the inactive X-chromosome in females and the silenced allele of imprinted genes, promoter-associated CpG islands are generally methylated, and this methylation is essential for maintaining the silenced state. Mechanisms regulating the establishment of methylation remain poorly understood, but the consequences of CpG island methylation are becoming increasingly clear. Methylation triggers the binding of methylated DNA-specific binding proteins to CpG sites, attracting histone-modifying enzymes that, in turn, focally establish a silenced chromatin state (FIG. 1).

Consistent with a resurgence of interest in the idea that cancer is a disease of faulty development, there has been a revival of interest in the epigenetic processes involved in neoplastic development and progression[5,6]. Epigenetic information, after all, is essential for development, and it is clear that cancer is ultimately a disease of aberrant gene expression. An epigenetic contribution to cancer is no longer in doubt. Early experiments showing that the reprogramming of epigenetic information after fertilization reverses the malignant phenotype[7] have been confirmed using modern technology such as nuclear transfer[8,9]. The potential reversibility of epigenetic changes through pharmacological manipulation makes this area acutely important in cancer management[5,10], and a specific DNA methylation inhibitor (5-azacytidine) has now been approved for use as an anti-cancer agent in the USA[11].

DNA methylation is central to the aberrant epigenetics of cancer. As described in extensive, recent reviews, cancer cells often have both a loss of global methylation and a gain of methylation at the promoters of selected CpG islands, resulting in the silencing of hundreds of genes per cancer cell, including tumour-suppressor genes[6,12]. Indeed, DNA methylation is proving to be a useful marker of disease risk, activity and prognosis in various malignancies[13,14]. However, a careful evaluation of the causes of aberrant methylation in cancer has been somewhat overlooked. There is a modest increase in DNA-methyltransferase activity in cancer[15], and immunohistochemistry studies have confirmed overexpression of DNA (cytosine-5)-methyltransferase 1 (DNMT1) protein in primary cancer cells[16], albeit with significant heterogeneity. Some of this overexpression might be related to the regulation of DNA-methyltransferases by the cell cycle[17], and RNA levels of *DNMT1* or *DNMT3* have largely not proven useful in explaining aberrant patterns of DNA methylation in cancer[18].

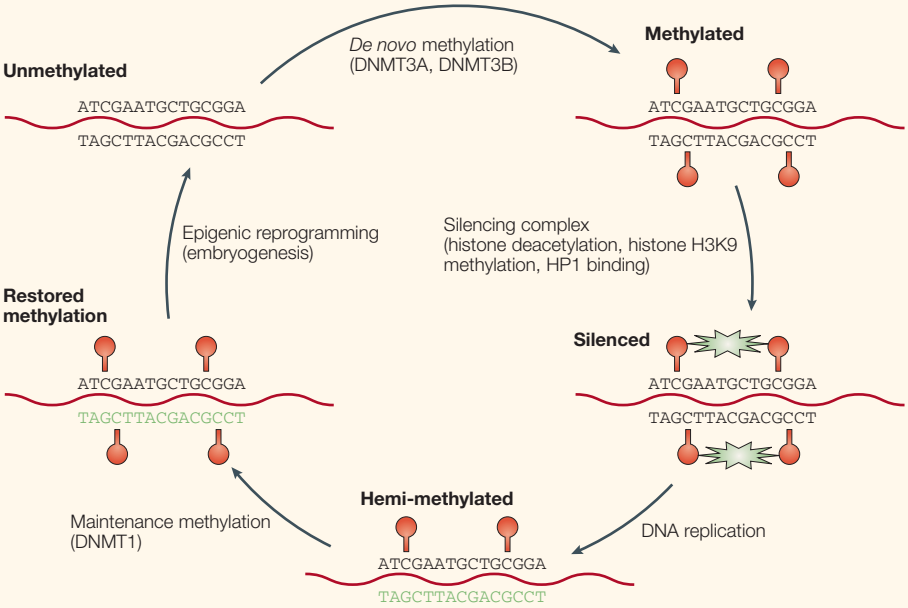

Figure 1 | **DNA methylation and gene silencing.** In early embryogenesis, DNA is largely devoid of methylation (top left). Post implantation, *de novo* methylation begins (red circles), mediated primarily by DNA (cytosine-5-)-methyltransferase-3α (DNMT3A) and -3β (DNMT3B) (top). When methylation affects CpG islands, methyl-binding proteins trigger a silencing cascade (activity illustrated by green stars) whereby histone H3K9 is sequentially deacetylated and then methylated, allowing heterochromatin protein 1 (HP1) to bind; eventually resulting in closed chromatin (bottom right). After DNA replication, newly synthesized DNA (in green) is unmethylated. However, DNMT1 rapidly scans DNA and deposits methyl groups on newly synthesized DNA, opposite methyl groups present on the old DNA strand. This results in faithful replication of methylation patterns (bottom left) and the maintenance of silencing. Adult patterns of methylation are erased by epigenetic reprogramming in early embryogenesis (top left).

This remains an area of research interest. Associations between methylation and exposure to carcinogens such as viruses[19], smoking[20] and radiation[21] have been observed but have not led to mechanistic insights into the process. The central conundrum in the field remains unresolved: is aberrant DNA methylation a rare, random process that is selected for in neoplastic cells, or does it result from specific defects in the methylation process? This question is similar to one that has divided the field of genetic changes in cancer for years[22]: do mutations arise stochastically and are selected for, or does cancer development require processes that accelerate the rate at which genetic defects are acquired? The discovery of multiple mutator phenotypes in cancer, mostly as a result of defects in DNA repair, has resolved the latter question, but the issue of epigenetic changes (and DNA methylation in particular) remains largely controversial. Multiple concordant methylation events — the CpG island methylator phenotype (CIMP) — in a subset of colon cancers provides evidence for a process akin to the mutator phenotype that affects CpG island methylation instead (BOX 1).

Discovery of CIMP

Aberrant CpG island methylation events in cancer were first described almost two decades ago[23], but most of the early studies in the field focused on the examination of isolated genes. A few years ago, as more and more genes affected by the process were identified, puzzling data emerged indicating that in some tumours, groups of genes had consistently increased methylation. This was demonstrated statistically by showing that methylation of two separate genes was correlated in a given tumour type. This was most apparent in colon cancer, where it was shown that sporadic tumours with microsatellite instability (MSI) had increased frequencies of promoter methylation affecting multiple genes such as *CDKN2A*, which encodes the protein INK4A, and *THBS1* (thrombosponsin 1)[24]. Remarkably, parallel studies established that these MSI-positive tumours also had hypermethylation and silencing of the mismatch repair gene *MLH1*[25]. The suggestion that hypermethylation leads to MSI (through the silencing of *MLH1*) was supported by the observation that the inhibition of methylation reversed the mismatch repair defect in a colon cancer cell line[26]. Subsequent studies have added several loci to the list of genes that are preferentially hypermethylated in sporadic MSI-positive cases, including *HPP1* (hyperplastic polyposis gene 1, also known as *TMEFF2*)[27] and *CDKN2A*, which encodes ARF[28] and other proteins. Interestingly, these genes were not as frequently methylated in colon tumours from individuals with inherited MSI caused by germline mutations in mismatch repair genes[29], demonstrating that mismatch repair defects *per se* do not accelerate aberrant methylation.

A more extensive study of this phenomenon was made possible by the development of methods to identify CpG islands that are differentially methylated in specific situations[30]. Using 33 such loci to profile the methylation status of normal colonic mucosa compared with colorectal cancer, loci could be divided into two groups[31]. Most loci (26 out of 33 (79%)) are methylated in normal colonic mucosa as a function of age and undergo more extensive methylation in cancer. Therefore, the dominant 'cause' of methylation in cancer is actually methylation in normal tissues that arises as a function of age. A discussion of age-related methylation is beyond the scope of this article, but it is important to note that this finding has now been reproduced in several studies and is probably a universal phenomenon in epithelial malignancies[32,33]. However, when age-related methylation was filtered out, it became apparent that methylation of the remaining seven loci, which tended to be limited to neoplastic tissues, was clustered in a specific subset of cases[31]. This phenomenon, descriptively termed CIMP, defined a group of cancers with a 3–5-fold elevated frequency of aberrant gene methylation. Confirmation that this group is distinct came when separate genes were examined. Methylation of each of the genes encoding INK4A, MLH1 and THBS1 was almost exclusively limited to the CIMP-positive subset. In fact, 70–80% of sporadic MSI-positive colon cancers could be attributed to CIMP and associated MLH1 methylation. These data divided sporadic colorectal cancers into four distinct groups: $CIMP^{+}MSI^{+}$, $CIMP^{+}MSI^{-}$, $CIMP^{-}MSI^{+}$ and $CIMP^{-}MSI^{-}$. The distinct nature of these four groups was confirmed by widely divergent mutation rates in the tumour suppressor gene *TP53* and the oncogenes *KRAS* and *BRAF*[34,35]. A stark conclusion of this work was that, at the molecular level, sporadic colon cancer is in fact a collection of (at least) four different diseases.

The findings that led to the definition of CIMP have now been reproduced in several (although not all) studies. Concordant methylation of multiple genes and/or

Box 1 | **Definition of the CpG island methylator phenotype**

An important hurdle in the field will be to achieve a consensus definition for the CpG island methylator phenotype (CIMP). This is no trivial issue, given the variety of methods available for studying DNA methylation, each of which might give a slightly different definition[70]. The choice of genes and the minimal number of genes examined is also essential. In all studies of CIMP so far (positive or negative), each group has used different methods and different genes, which can only contribute to the confusion. Moreover, the choice of genes is also tissue-type dependent, and a definition for colon cancer might not be applicable to other cancers.

In order to move towards a consensus, two issues are crucial: first, a quantitative method is needed for studying methylation. CIMP seems to affect both the frequency and the extent of methylation[45], and the latter is missed if one uses a sensitive but non-quantitative method. Second, in order to select genes to define the phenotype, genes with high levels of methylation in normal tissues must be avoided. This is fairly straightforward in the colon, but is problematic in other diseases such as breast cancer, in which the relevant normal cells (breast epithelium) are a small fraction of the tissue that can be obtained at surgical resection or biopsies.

It is obvious that large studies are needed (large referring to both the number of genes and the number of cases) before this issue is resolved. An unbiased approach using quantitative analysis of a large number of genes in both normal and neoplastic tissues could definitively help resolve whether CIMP exists and what are the best markers to define it. In colon tumours, appropriate tests to confirm the presence of CIMP should include objective factors unequivocally linked to CIMP, such as histology, as well as mutations in *KRAS* and *BRAF*. This endeavour will require an exploration of statistical tools to analyse the data and it could initially follow the methods developed for analysis of microarray data[71]. One possible approach would be to perform a supervised data analysis using CIMP-associated endpoints (for example, the mismatch repair gene *MLH1*-associated microsatellite instability in sporadic colorectal cancer, or poorly differentiated mucinous sporadic colorectal cancers with mutations in *KRAS* or *BRAF*), then identify a minimal set of markers and confirm these in a separate group of tumours. This should be compared with an unsupervised clustering approach. The results of such large studies might then guide appropriate statistical treatment of methylation data sets in other tumours, where CIMP-associated endpoints are not yet clearly defined.

Until such experiments are completed, our laboratory has been defining CIMP in colon cancers by quantitatively studying a reduced set of genes , namely *MINT1*, *MINT2*, *MINT31*, *CDKN2A* and *MLH1*.

clustering reminiscent of CIMP has been confirmed in colorectal cancer[36–38] and has also been observed in glioblastomas[39], gastric cancer[40,41], liver cancer[19], pancreatic cancer[42], oesophageal cancer[14], ovarian cancer[43], acute lymphocytic leukaemia[44] and acute myelogenous leukaemia[45]. A few studies, however, have not found evidence of concordant methylation in colon cancers or other diseases[18,46]. A common feature of these latter studies is the use of methylation specific PCR (MSP) — a very sensitive, non-quantitative technique to measure methylation — and/or the inclusion of all genes in the analyses without attempting to filter out age-related methylation. Indeed, a recent study in colorectal cancer[47] concluded that methylation is primarily a characteristic of ageing and that, using unselected genes, CIMP cannot be identified in this disease, exactly as predicted by the original observations[31]. In fact, re-examination of data in that paper does reveal clustering of methylation of specific genes (such as *CDKN2A*, encoding varients INK4A and ARF) in the subset of cases that has *MLH1* methylation-based MSI, indicating that CIMP might be discernible with appropriate statistical analysis. Every research group that has quantitatively studied the methylation status of the genes originally used to define CIMP has confirmed that there is concordant methylation in colon, gastric, pancreatic and ovarian cancer.

The origin of DNA methylation in cancer

CIMP has fundamental implications for our understanding of the origin of tumour-suppressor gene methylation in cancer. First and foremost, the fact that different cancers have significantly different rates of tumour-suppressor gene inactivation through DNA methylation-associated processes implies that methylation is not random. In turn, this means that aberrant DNA methylation has a cause that should be searched for and identified. Teleologically, it also implies that this process is selected for during neoplastic transformation and, ultimately, it provides strong evidence that epigenetic tumour-suppressor gene inactivation is a cause of cancer development. This issue cannot be stressed strongly enough. Early developments in the field were met with substantial skepticism, with many investigators arguing that DNA methylation simply results from neoplastic transformation[48]. Indeed, it is likely that much aberrant DNA methylation is a by-product of the neoplastic process, perhaps an extension of the age-related methylation process. Nevertheless, the presence of CIMP shows that all methylation events are not equal, and that DNA methylation does not necessarily follow malignant transformation. Rather, the subset of tumours with intense methylation is probably formed, in part, through changes in cell physiology resulting from epigenetic silencing.

CIMP cancers — unique diseases

The molecular identification of CIMP led to a reappraisal of its clinical and pathological features, and, in many respects, established it as defining a unique subset of colorectal cancers[31,34,36,38]. CIMP-positive cancers tend to occur in older patients, are overrepresented in proximal tumours of the colon, and occur more often in women. Genetically, CIMP positive cases are also distinct, with a paucity of p53 mutations and a remarkably high rate of mutations in *KRAS* or *BRAF*[34,35], such that nearly every CIMP-positive tumour has evidence of activation of the RAS oncogenic pathway. CIMP-positive cases are also distinct pathologically. Initially, this uniqueness was thought to be related to the strong link between MSI and CIMP. However, CIMP is also associated with distinct features in cases without MSI[36], and recent studies of stage IV colorectal cancer (in which MSI is rare) identified unique histological features of CIMP-positive cases including poor differentiation and unusual gland architecture (REF. 36; L. Chirieac, personal communication). CIMP-positive cases also show unique clinical attributes. MSI-positive cases tend to have a good prognosis[49], whereas $CIMP^{+}MSI^{-}$ cases have a particularly poor outcome (REF. 50; L. Shen, personal communication). Most remarkably, CIMP is central to recent findings that indicate an alternative precursor lesion to colorectal cancer, a concept that has profound clinical implications[51]. A hitherto overlooked hyperplastic polyp might well be the precursor to serrated adenomas which, in turn, give rise to MSI-positive cancers. In fact, serrated adenomas seem to be a methylation-based disease, showing a high incidence of CIMP, as well as *KRAS* and *BRAF* mutations[35,52–54] (BOX 2).

Although they are most apparent in colon cancer, the unique features of CIMP extend to other malignancies. CIMP-positive

Box 2 | **The hyperplastic polyp/serrated adenoma pathway**

Colon cancer has served as a ubiquitous model for the development and progression of neoplasia, from its earliest to most advanced stages[72]. However, recent molecular and pathological findings have led to questions about key parts of that model and are poised to markedly alter our thinking on the prevention of colon cancer[73]. Classical models ascribe all colon tumours to the aberrant crypt focus and adenomatous polyp precursor lesions. Colon cancer prevention by colonoscopy has focused on the adenoma, and adenoma removal is in fact associated with a reduction in colon cancer incidence. Prevention trials have also commonly used the adenoma as a surrogate endpoint.

However, for several years, polypoid lesions other than adenomas have been known to occur in the colon. The hyperplastic polyp is an easily recognizable lesion, but it has been given little attention since early studies concluded that it was not a precursor to malignancy. Serrated adenomas (so-called because of their histological appearance) have also been recognized as a rare variant of adenomatous polyps, but have been given little separate attention. The recognition of microsatellite instability (MSI)-positive cancers as a pathologically distinct subset has led investigators to re-evaluate this issue. Histological similarities between lesions, co-occurrence of unusual tumours and compelling molecular data now identify some hyperplastic polyps as precursors to serrated adenomas that seem, in turn, to be precursors to MSI-positive cancers[51]. There are remarkable similarities between proximal hyperplastic polyps, serrated adenomas and CpG island methylator phenotype (CIMP)-positive cancers (a high degree of methylation of multiple genes[53], frequent mutations in *BRAF*[35,54], and shared histological features such as serration[51]) and a strong argument can now be made that the serrated pathway is largely a CIMP pathway, and that it could be a precursor of many CIMP-positive cancers.

Needless to say, this concept is important clinically. Some hyperplastic polyps (for example, proximal ones and large ones) can no longer be ignored as having no malignant potential. Pathologists are now recognizing serrated lesions at a higher frequency, and a suggestion has emerged that these lesions could be more aggressive in terms of neoplastic transformation. Prevention trials should examine these lesions, given that their molecular characteristics are different than adenomas, and prevention strategies might have to be reviewed. Finally, the identification of individuals with multiple hyperplastic polyposis syndromes[52] might eventually lead to a cause for these lesions that is distinct from classical colon cancer familial genes.

and CIMP-negative gastric cancers tend to have different locations[41]. CIMP-positive acute lymphoblastic leukaemias rarely show the presence of the Philadelphia chromosome[44]. In general, the simultaneous methylation of multiple genes, a hallmark of CIMP, is associated with a poor outcome in multiple malignancies, including head and neck, lung, prostate and oesophageal cancer and acute leukaemias[14]. It is likely that, just as expression profiling is defining pathologically and clinically distinct groups of cancers, methylation profiling will also achieve a significant reduction in the high degree of heterogeneity that is currently recognized in various malignancies.

Causes of CIMP

The causes of CIMP remain unknown. More generally, the pathogenesis of aberrant DNA methylation in ageing and cancer and the mechanisms by which genes are selected for methylation have not been elucidated. A series of arguments have to be considered before proposing a unifying theory of the origin of neoplastic methylation patterns. First, not all genes are created equal with regards to methylation in cancer. *MLH1* and *MSH2* (mutant S homologue 2) — two genes that are involved in mismatch repair — give rise to similar phenotypes (hereditary nonpolyposis colon cancer) when mutated in the familial setting, but, in terms of epigenetic inactivation, *MLH1* is a frequent target whereas *MSH2* is never methylated in cancer, despite having a sizeable CpG island[55]. Second, methylation might follow gene inactivation in some cases[56]. However, hypermethylation cannot be attributed simply to transcriptional inactivation, otherwise, genes showing tissue-specific expression in normal tissues should be hypermethylated in non-expressing tissues, and this, for the most part, does not occur[2]. Instead, data strongly indicate that gene inactivation might facilitate the establishment of DNA methylation and/or spreading through other mechanisms[57]. Third, selection clearly plays a part in the process, during which tumour-suppressor gene methylation is selected for[12] and oncogene methylation is selected against[58]. Fourth, CIMP implies that a catastrophic event can affect the methylation machinery, thereby raising methylation rates (affecting diverse genes) by several fold in selected cancers.

One model that accounts for the diverse observations discussed above has been suggested by studies on the formation of DNA methylation patterns during embryogenesis. This model[59], based on initial work by Turker and colleagues (reviewed in REF. 60), proposes the following: methylation initially arises in 'methylation centres', short sequences that attract DNA methyltransferases based on sequence features. Repetitive elements that can trigger recombination when they are expressed (for example, retrotransposons) form excellent candidates for such methylation centres, but whether they are the only ones involved remains to be seen. This concept solves one of the conundrums in the field – gene selectivity. Regardless of function, if a gene does not have a methylation centre close to the promoter, it will not be susceptible to DNA methylation in cancer even if it is transcriptionally silenced. Methylation centres are only one half of the equation. DNA methylation, once established, tends to spread in *cis*, and this spreading is crucial to eventual gene inactivation. CpG islands seem to have developed specific mechanisms to block such methylation spreading, and this barrier is key to maintaining the methylation-free state. Therefore, one view of DNA methylation in predisposed genes is that of a struggle between methylation-promoting events and methylation-protection events.

In light of this model, one can propose simple explanations for both age-related methylation and CIMP (FIG. 2). Initially, *de novo* methylation and spreading is a replication-dependent phenomenon[61], and is therefore crucially dependent on age. Ageing tissues simply extend the patterns of DNA methylation that were deposited during embryogenesis, and methylation slowly extends towards promoters, piling up at the borders of the islands where protection is presumed to be strong. For some genes, protection against the spreading of methylation is weak, either intrinsically or because protection is mediated by gene expression that can be reduced during ageing, and this will show up as age-related promoter methylation in normal and neoplastic tissues. For other genes, protection is strong, and these genes will only become hypermethylated if there is strong selection for methylation, or if the barrier is somehow lost, possibly by a genetic event. Given that there are probably a limited number of protective mechanisms, the inactivation of a single protecting gene might result in multiple genes being affected simultaneously, exactly as observed in CIMP. Of course, CIMP could also result

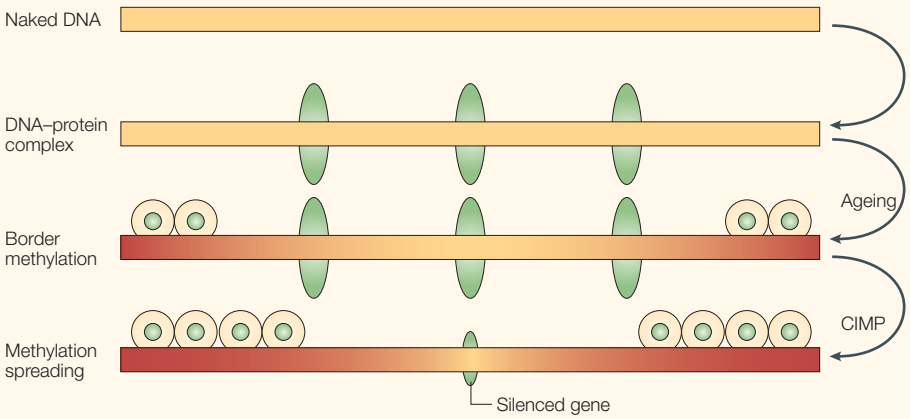

Figure 2 | **A model of hypermethylation in cancer.** Naked CpG island DNA (top) is unmethylated (yellow) and coated by proteins (green ovals) that protect against DNA methylation establishment and/or spreading. The nature of these proteins is unknown, but probably include transcription factors, co-activators or similar molecules. During repeated rounds of the stem-cell mobilization and replication that accompany ageing, DNA methyltransferases (circles) are recruited to the borders of some CpG islands, depositing methyl groups (red) and creating methylation pressure for these islands. The nature of this initial recruitment is unknown but is probably related to repetitive DNA sequences and/or retrotransposons. The balance of methylation pressure (circles) and methylation protection (ovals) is disrupted in the CpG island methylator phenotype (CIMP), resulting in the spread of methylation into the transcription start area and the triggering of the silencing cascade. As discussed in the text, the disruption of this balance will probably be achieved through the loss of protective proteins (as indicated in the bottom panel), which could occur by mutations that inactivate these proteins or the loss of expression by other mechanisms such as transcription factor loss or histone modifications. Theoretically, this balance could also be disrupted by overactive *de novo* methylation pressure (circles), for example by activating mutations in DNA methyltransferases.

from a markedly enhanced *de novo* methylation pressure, which could result from activating mutations in DNA methyltransferases or histone-modifying proteins. Finally, this model helps to explain why CIMP occurs more frequently in older individuals. Age-related methylation is required to predispose genes to DNA methylation-associated inactivation. Therefore, a genetic event causing CIMP might not affect young tissues, in which methylation is not yet built up at the island borders, ready to spread and inactivate the gene.

An alternative explanation for CIMP does not require specific genetic defects, but instead requires repeated exposures to 'epimutagens'. There is mounting evidence for an environmental influence on DNA methylation in both normal (ageing) tissues and cancer[32]. Indeed, there is an association between methylation and viral exposures in the liver[19] and in the stomach[62]. Diet might affect DNA methylation patterns[63], and it would be interesting to determine whether dietary or genetic factors influencing one-carbon metabolism (for example, folate) might affect CIMP. In addition, specific carcinogens seem to have particular preferences for inducing aberrant methylation in rodent models of cancer[21]. It is conceivable, therefore, that CIMP reflects chronic exposure to epimutagens that could then cause or accelerate cancer development through epigenetic pathways. Interestingly, a study of multiple polyps in individuals affected by hyperplastic polyposis showed that there was a high degree of concordance of methylation between the individual lesions[52], indicating a patient-specific predisposition to CIMP-positive tumours that could be related to either environment and lifestyle, or genetic predisposition. Finally, it is worth mentioning that a universal accelerator of DNA methylation is chronic inflammation, as indicated by studies in preneoplastic colon[64], oesophagus[65], liver[19] and lung[66]. Tumours that arise in the setting of chronic inflammation in the colon (for example, ulcerative colitis) are more likely to be CIMP-positive[67], again linking lifestyle and exposures to the phenotype.

Implications of CIMP

Perhaps the single most important implication of the CIMP concept is that methylation in cancer has a traceable cause. Therefore, a search for mutations in genes that are potentially involved in DNA methylation is relevant here, although genes that confer protection against methylation have so far not been identified. The observation of familial clustering of CIMP[68] or DNA methylation[69] might lead to the discovery of the gene(s) through classical genetic studies. It is likely that several multiple cancer familial syndromes will eventually be traced to abnormalities in genes that control DNA methylation. Therefore, linkage studies need to be directed by the molecular phenotype of the malignancies of interest, an approach that should increase the chances of correct gene identification by reducing heterogeneity in the tumours. If, however, CIMP can be traced unequivocally to environmental exposures, this should lead to a careful evaluation of the epimutagen concept. This issue has a substantial public health impact because the carcinogenic potential of various exposures is currently identified primarily through tests of mutagenicity. Such tests might underestimate the carcinogenic potential of exposures that lead to cancer primarily through epigenetic lesions.

The data reviewed above also argue strongly for methylation as an event that is selected for and is therefore physiologically relevant to the neoplastic process. There is still some controversy over the pathophysiological role of aberrant methylation in cancer[48]. Nevertheless, the presence of a hypermethylator phenotype provides the same degree of confidence in the role of methylation in cancer as mutator phenotypes do for genetic changes in cancer. In addition, these data indicate that a large number of CpG islands might be methylated simultaneously in some (but not all) cancers, which complicates the interpretation of the functional importance of methylation of individual genes, and the use of methylation as a screening tool for neoplasia. Importantly, the frequent association between CIMP and a poor prognosis also indicates a clinical implication. Ultimately, this should lead to a close evaluation of whether epigenetically acting drugs[10] are useful in this subset of cases, and whether epigenetic interventions might specifically prevent the emergence of CIMP-positive cancers.

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Competing interests statement
The author declares no competing financial interests.

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The following terms in this article are linked online to:

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colon cancer | gastric cancer | liver cancer | pancreatic cancer | oesophageal cancer | ovarian cancer | lymphocytic leukaemia | myelogenous leukaemia

FURTHER INFORMATION

MD Anderson Cancer Center resource on CpG island methylation:
http://www.mdanderson.org/departments/methylation