A bittersweet symphony

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Abstract

This issue analyzes new work expanding the range of how genetic dysregulation of succinate dehydrogenase subunit (SDHx) genes can cause cancer syndromes with a prominent endocrine component, in this case Carney triad, which is characterized by gastrointestinal stromal tumors, paraganglioma, and pulmonary chondromas.

Key Words

- ▶ gene regulation
- ▶ Carney triad

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Introduction

Carney triad was originally recognized following the painstaking clinicopathological work of Dr J Aidan Carney at the Mayo Clinic between 1975 and 1977 (Carney *et al.* 1977). The initial triad consisted of gastric gastrointestinal stromal tumor (GIST; at the time gastric 'leiomyosarcoma'), paraganglioma, and pulmonary chondroma. While it is traditional to use the term GIST in describing the gastric lesions in Carney triad, they are probably better described as 'gastric stromal sarcoma'. Subsequently, other features – some of which were present in the early cases – have been added to the original triad, including adrenal cortical adenoma and esophageal leiomyoma; as noted by Dr Carney himself, what started as a triad may actually be a pentad (Carney 2009). Carney triad has long been a puzzling entity, having all of the features of a genetic disease, including multiple rare tumor types occurring metachronously or synchronously in different organs and having particularly aggressive features, including young age at onset. This profile in Carney triad is accompanied by a remarkably strong female predisposition. However, unlike the overlapping autosomal dominant Carney–Stratakis syndrome (CSS) of paraganglioma and GIST (Carney & Stratakis 2002, McWhinney *et al.* 2007, Pasini *et al.* 2008), Carney triad is not a familial condition. This has led to the question being raised on more than one occasion: is Carney triad a genetic disorder? (Matyakhina *et al.* 2007, Stratakis & Carney 2009).

Succinate dehydrogenase gene dysregulation is instrumental in orchestrating various endocrine cancer syndromes

The past few decades have observed remarkable successes in determining the causes of paragangliomas and GIST, two of the component tumors of the Carney triad (Neumann & Eng 2009, Barnett *et al.* 2013, Vicha *et al.* 2014). The number of mutated genes associated with sporadic and familial types of paraganglioma/pheochromocytoma has grown rapidly, involving processes that generally cluster as hypoxic/oxidative and kinase-dependent pathways (Gimenez-Roqueplo *et al.* 2012, Vicha *et al.* 2014). Core to our understanding of the pathogenesis of many of these tumors is the role of dysregulation of succinate dehydrogenase (SDH) subunit genes, *SDHA* through *SDHD* (collectively *SDHx*). Mutations of these genes occurring at the germline and somatic levels interfere with mRNA expression and protein function and have profound effects on tumoral biochemical pathways, thereby enhancing neoplastic growth. However, genetic modulation of *SDHx* function is not limited to a paraganglioma–pheochromocytoma phenotype alone. CSS is also caused by *SDHx* mutations, while mutations in these genes are also an important cause of sporadic GIST (McWhinney *et al.* 2007, Pasini *et al.* 2008, Janeway *et al.* 2011). Given that *SDHx* mutation syndromes also encompass a group of mitochondrial/neuromuscular

disorders (Alston *et al.* 2012), it is clear that the clinical phenotypes associated with *SDHx* mutations are remarkably varied.

Mutation, however, is only one of a number of mechanisms by which alteration in gene function may occur in cancers. For instance, epigenetic alterations are well established as playing an important role in both syndromic and sporadic endocrine cancers (Rodriguez-Rodero *et al.* 2014). This issue of *Endocrine-Related Cancer* reports results of a new study that demonstrates that specific and preferential hypermethylation of one SDH subunit gene (*SDHC*) is a crucial pathophysiological event in Carney triad (Haller *et al.* 2014). This study led by groups at the Eunice Kennedy Shriver National Institute of Child Health and Human Development and the Friedrich-Alexander University Erlangen-Nuremberg, Germany, demonstrates that hypermethylation of the *SDHC* promoter region is an abnormality that is specific to Carney triad tumors when compared with similar tumors that occurred outside of the setting of Carney triad. This epigenetic mechanism of *SDHC* silencing as a tumorigenic mechanism was further underlined by the demonstration of a marked reduction in *SDHC* mRNA in Carney triad tumors, decreased SDHC protein on Western blots of tumors, and a reduced functional activity of complex II that was attributable to SDHC deficiency. Crucially, this molecular genetic signature of a specific *SDHC* abnormality in Carney triad tumors differentiated them markedly from similar tumors such as sporadic GIST due to *KIT* mutations or familial paraganglioma syndrome type 1. This finding answers a long-standing conundrum about the etiology of Carney triad (yes, it is an (epi)genetic disorder!). It also expands significantly the variety of mechanisms by which *SDHx* gene dysregulation can orchestrate various distinct and overlapping neoplasia syndromes.

While the findings described herein by Haller *et al.* (2014) are the first to definitively link *SDHC* genetic dysregulation to tumorigenesis in Carney triad, earlier work from the Stratakis group had suggested that *SDHC* might have a role to play. Matyakhina *et al.* (2007) demonstrated that among 41 tumors in 37 cases of Carney triad, the most frequent recurrent genetic abnormality was a large-scale copy number loss that included the locus for SDHC (chromosome 1q21–q23.3). In addition to the epigenetic mechanism for the etiology of Carney triad through hypermethylation and silencing of *SDHC*, Haller *et al.* (2014) noted a definite – albeit less clear-cut – hypermethylation signal at the *SDHB* promoter. This epigenetic silencing profile of *SDHB*, along with *SDHB* copy number changes and loss of SDHB staining in Carney triad tumors implies that SDHx-related abnormalities might be somewhat more

generalized than *SDHC* alone (Matyakhina *et al.* 2007, Gaal *et al.* 2011, Haller *et al.* 2014). Study of more Carney triad tumors will help to define the relative variability or specificity of hypermethylation patterns in *SDHC* and *SDHB*. The etiology of this seemingly specific *SDHC*-focused epigenetic profile in Carney triad remains to be explained. This finding stands in sharp contrast to the more general hypermethylation profile in tumors from patients with germline *SDHx* gene mutations (Killian *et al.* 2013, Letouze *et al.* 2013).

The finding of a specific *SDHC* hypermethylation profile of tumors in Carney triad raises the possibility of using this epigenetic signature to study the nature of Carney triad itself. Carney triad is a disease characterized by incomplete disease penetrance. Patients usually present with only one of the component tumors, and a further third have two of the three tumors; presentation with all three together is very rare (Carney 2009). As only a quarter of patients will eventually develop all three tumors during their lifetimes, 75% of cases will have a gastric stromal sarcoma plus a pulmonary chondroma or a paraganglioma (Carney 2009). The presence of a specific *SDHC* hypermethylation signature in affected tumors could add a new method for firming up a suspected diagnosis of Carney triad in patients who have undergone surgical biopsy or resection. This molecular genetic signature could also assist in determining whether apparently sporadic GIST or paragangliomas might represent emerging Carney triad or CSS cases. This type of information could be useful clinically where family medical history is not known or is incomplete. Collection of clinical, pathological, and genetic information regarding patients and their tumors has already shown itself to be of help in studying the role of *SDHx* mutations in the etiology of paraganglioma syndromes, and differentiating among various etiologies of GISTs (van Nederveen *et al.* 2009, Gaal *et al.* 2011); we may need to incorporate tumoral epigenetic signatures into the pathology repertoire.

Using hypermethylation signatures, it could also be possible to explore the other potential disease associations with Carney triad. For example, the so-called fourth component of Carney triad, adrenal cortical adenomas, occurs in 19% of cases; most of them are unilateral and asymptomatic, with about half of them being detected by imaging (Carney *et al.* 2013). These tumors can be managed conservatively, but it would be intriguing to study existing excised adrenal tumors from Carney triad cases to examine if they too demonstrate hypermethylation of *SDHC*. The potential involvement of other rare associations such as esophageal leiomyoma and esophageal carcinoid in the Carney triad could be similarly explored. Finally, the

reason for the remarkable female predominance of Carney triad will need to be assessed against the light of this new epigenetic disease mechanism.

Summary

Taken together, the range of syndromic conditions and sporadic tumors associated with abnormal *SDHx* gene regulation is increasingly varied. It is instructive, perhaps, to recognize that the mechanism by which a specific gene, such as *SDHC*, is dysregulated can lead to very important differences in disease characteristics at presentation and through the lifetime of the patient. The current study on Carney triad underlines the need to examine all of the different possible mechanisms of gene regulation when pursuing the etiology of unsolved endocrine tumor syndromes.

Declaration of interest

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