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The sympathetic nervous system: malignancy, disease, and novel functions

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The sympathetic division of the autonomic nervous system comprises cell types as diverse as sympathetic neurons, which control internal organs and blood vessels via noradrenergic nerve terminals, the endocrine, catecholamine-releasing chromaffin cells, and the chemoreceptive type I cells of the carotid body, which serve as peripheral oxygen sensors. While sympathetic neurons form the para- and prevertebral ganglia, chromaffin cells are situated in the adrenal medulla or in paraganglia located in close proximity to large blood vessels, along sympathetic nerves innervating pelvic organs and within sympathetic ganglia during development. Paraganglionic cells also include the type I cells of the carotid body, which originate from sympathetic progenitors of the superior cervical ganglion during embryonic development (Kameda 2014).

The fascination of these so-called sympathoadrenal (SA) cells lies not only in their multiple vital functions but also in their intriguing pathobiology with a variety of benign and malignant tumors originating from them. Neuroblastoma is the most common extra-cranial solid tumor of childhood. This neoplasm is associated with the developing sympathetic nervous system or the developing adrenal medulla. In contrast, pheochromocytoma and paraganglioma usually occur in adults. By definition, pheochromocytomas originate from the adrenal medulla, while paragangliomas are derived from extra-adrenal chromaffin tissue or chemoreceptive paraganglia (Lloyd et al. 2017).

How the distinct individual properties and functions of SA cells are related to their susceptibility to tumor formation and their tumor biology is one of the central points addressed in this special issue. Neuroblastoma, as a tumor derived from developing tissue, has been in particular associated with dysfunction or mutations of molecules that regulate SA cell development (Chan et al. 2018), including MYCN, ALK (Janoueix-Lerosey et al. 2018), and PHOX2B (Trochet et al. 2004; Mossé et al. 2004). Some pheochromocytomas and paragangliomas have been intimately linked to hypoxic pathway genes, including hypoxia-inducible transcription factors (HIFs) and the von Hippel-Lindau tumor suppressor (VHL) (Fliedner et al. 2018; Kluckova and Tennant 2018). This may at least in part be related to the fact that oxygen sensing is an integral part of the normal physiology of type I carotid body cells as well as early chromaffin cells (Nurse et al. 2018). Hypoxic pathway genes are important for the proper function of oxygen sensing and the plasticity of these cells (Prabhakar and Semenza 2016; Nurse et al. 2018). These molecules, in particular HIF-2α, in addition have putative functions during the early development of SA cells (Brown et al. 2009), linking them also to neuroblastoma (Påhlman and Mohlin 2018). The recent discovery that sympathetic neurons and adrenal chromaffin cells are derived from separate progenitors with distinct molecular fingerprints (Furlan et al. 2017), contradicting

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the original concept of a common SA progenitor, will be of high value for the understanding of the pathogenesis of neuroblastoma as well as pheochromocytoma. It also raises new questions as to the cellular origin of neuroblastoma (see Chan et al. (2018) and the "Neuroblastoma" section) and to the developmental origin of type I cells of the carotid body.

This special issue covers the pathobiology of SA cells, with a focus on tumor biology. Reviews range from very basic to clinically orientated research aiming to understand the pathogenesis of these diseases, to identify appropriate diagnosis and therapy strategies, and to determine the individual prognosis. In an effort to link pathobiology to regular cell function, this special issue also deals with selected aspects of the normal biology of SA cells. It includes a review providing a comprehensive picture on the development of the diverse types of SA cells (Chan et al. 2018), a section covering neuroblastoma from development and animal models up to its clinical features, a part on pheochromocytomas and paragangliomas, and a section on the role and pathology of oxygen sensing pathways in the carotid body and chromaffin cells. This special issue aims to bridge the gap between fundamental and clinical science, to identify similarities and differences between the distinct types of tumors originating from SA cells, and to foster new ideas for the benefit of both fundamental science and patients.

Neuroblastoma

In this part of the special issue, current knowledge on neuroblastoma is delineated from clinical presentation, genetic predisposition, tumor development, and animal models to major tumor driver genes, intratumor heterogeneity, and relapse.

Neuroblastoma patients are stratified into various categories including low-risk patients with survival rates greater than 90% and aggressive, high-risk neuroblastoma with current overall survival rates below 50%. Risk stratification is important to avoid overtreatment of low-risk patients resulting in late complications and to select and improve targeted therapies for high-risk patients. Clinical presentation, diagnosis, risk stratification according to the INRG staging, and therapy are reviewed by Tolbert and Matthay (2018). Notably, targeted small molecule inhibitors and immunotherapy are presently used for relapsed and refractory patients but may be included in the personalized initial therapy of newly diagnosed patients.

Current research aims to understand the mechanisms that underlie tumor initiation, growth, and relapse using different animal models where tumors can be induced by the expression of single or combinations of potential tumor driver genes identified in high-throughput genetic analysis. However, as about two thirds of neuroblastoma tumors are devoid of obvious deleterious mutations, the search also continues for other mechanisms of tumor initiation, e.g., perturbations of epigenetic mechanisms controlling sympathoadrenal development that may lead to genomic instabilities and subsequently to chromosomal alterations. Thus, the detailed description of the normal development of sympathoadrenal cells by Chan et al. (2018) provides an essential reference point for studies on aberrant development during neuroblastoma development. The classical scheme of normal sympathoadrenal development has been recently changed by single-cell RNAseq and cell lineage analysis demonstrating that sympathetic neurons and chromaffin cells have a different cellular origin, deriving directly from neural crest cells and Schwann cell progenitor cells (SCPs), respectively (Furlan et al. 2017). These findings implicate SCPs, besides neural crest progenitor cells and sympathetic neuroblasts, as potential cell of origin for neuroblastoma.

Neuroblastoma induction by transgenic expression of a tumor driver gene in noradrenergic cells was first shown in the mouse for MYCN, followed by activated Alk and Lin28B (reviewed by Tsubota and Kadomatsu (2018)). As their expression was controlled by the Th or Dbh promotor, noradrenergic neuroblasts were thus identified as cell of origin, at least in these models. However, as the MYCN-transfected neural crest cell line JoMa-1 and MYCN-transfected primary neural crest cells generate neuroblastoma upon subcutaneous injection in mice, neural crest cells may also represent cells of origin for human neuroblastoma (discussed by Janoueix-Lerosey et al. (2018) and Tsubota and Kadomatsu (2018)). Both mouse and zebrafish have been used to confirm the neuroblastoma driver function of gene amplifications (MYCN) or somatic mutations (ALK^{F1174L}) and to define molecular mechanisms underlying tumor development. This included evidence for cooperative interactions between MYCN and ALK^{F1174L} or MYCN and PTPN11 resulting in strongly increased tumor penetrance. Tumor models were also used for pharmacological screens leading to the identification of novel drugs (reviewed by Casey and Stewart (2018)). As pointed out by Casey and Stewart (2018), the zebrafish model may be superior to that of the mouse for studying TERT- and ATRX-dependent mechanisms of telomere lengthening that are potential drivers in a large proportion of human high-risk neuroblastoma (Peifer et al. 2015; Valentijn et al. 2015). Mouse telomeres are much longer and Tert mutants in mice are viable in contrast to mutations affecting telomere maintenance in human and zebrafish. Thus, although key oncogenic pathways are conserved between human, mouse, and zebrafish, the specific advantages and disadvantages of the models (e.g., the gene duplications in zebrafish) should be taken into consideration depending on the question analyzed.

Whereas transgenic animal models have been essential for the analysis of specific oncogene functions, patient-derived xenografts (PDX) display features that are very close to human neuroblastoma with respect to molecular and cellular characteristics, including invasive behaviors. Braekeveldt and Bexell (2018) discuss methodology, advantages, and limitations of neuroblastoma PDX, highlighting also the usefulness of PDX in the search for novel treatments of relapsed neuroblastoma.

The recent discovery that neuroblastoma cell lines display either a noradrenergic, a neural crest-like/mesenchymal, or a mixed identity and that most primary tumors are composed of a mixture of noradrenergic and neural crest-like/mesenchymal neuroblastoma cells (Boeva et al. 2017; van Groningen et al. 2017) raises the question whether neural crest-like/mesenchymal cells may represent neuroblastoma cancer stem cells. Putative neuroblastoma cancer stem cells have been identified based on marker gene expression, increased ability for tumor formation, chemoresistance, and formation of metastases (reviewed by Tomolonis et al. (2018)). Of particular interest are highly tumorigenic CD114-positive cells that express neural crest cell and EMT markers and are present as a minor population in all neuroblastoma cell lines and primary neuroblastoma samples analyzed. However, additional in vivo analysis by serial re-implantation experiments is required to see whether these are transformed stem cells that self-renew, differentiate into diverse progenies, and drive continuous tumor growth (Kreso and Dick 2014). Intratumoral heterogeneity has also been inferred from the sequential acquisition of somatic mutations during tumor progression and is supported by the finding that pre-existing clones of cells with activated RAS/MAPK and MYCN expand in relapsed neuroblastoma (Schulte et al. 2018). As emphasized by these authors, the existence of diverse populations of cancer cell clones that respond differentially to therapy represents a major challenge for treatment design in personalized treatment. Påhlman and Mohlin (2018) point out that hypoxia-inducible factor (HIF)-2α, which is transiently expressed in sympathetic neuroblasts during their early normal development, is a marker of a certain population of "stem cell-like" neuroblastoma cells in the perivascular niche and associated with aggressive disease. They suggest that HIF-2 represents a possible therapeutic target for the treatment of neuroblastoma proposing a strategy that involves the induction of differentiation of immature HIF-2α-positive tumor cells by inhibiting HIF-2.

Spontaneous regression is observed in many types of tumors but it is most prevalent in neuroblastoma. The fourfold increase in neuroblastoma prevalence observed in mass screenings implicates that 75% of tumors detected will regress subsequently. Interestingly, spontaneously regressing tumors are characterized by specific molecular traits, which correlated with good prognosis and allowed the classification as MS tumors (4S in the old nomenclature). As detailed by Brodeur (2018), high-level expression of TrkA, low telomerase, and epigenetic and immunological properties of 4S tumors indicate potential mechanisms involved in tumor regression and how to enhance this process in patients.

Genetic determinants that predispose to neuroblastoma have been first identified in familial forms of the disease with mutations in ALK observed in 75% and PHOX2B mutations in about 15% of cases. In contrast, very few recurrent somatic mutations are present in sporadic neuroblastoma, in contrast to many adult cancers with disease-defining somatic mutations driving tumor development. However, many common susceptibility alleles were discovered by genome-wide association studies that are associated with neuroblastoma development and progression. The enrichment of some of these variants in low- or high-risk tumors implicates specific effects of defined genotypes on neuroblastoma phenotypes. As reviewed by Ritenour et al. (2018), these studies identified genes with oncogenic or tumor suppressive roles, including CASC15, NBAT-1, BARD1, LMO1, HACE1, and LIN28B. Interestingly, a number of rare germline variants (e.g., TP53, BARD1, BRCA1, BRCA2) that may have a higher effect size were recently discovered by extensive whole genome and exon sequencing of neuroblastoma patients. The identification of CASC15 and NBAT-1 long non-coding RNAs in neuroblastoma development illustrates the importance of epigenetic regulation in neuroblastoma development. As detailed in this issue by Durinck and Speleman (2018), epigenetic mechanisms generally play important roles in cancer, with about 20% of mutations in pediatric cancers observed in epigenetic regulators involved in DNA methylation, histone modification, and chromatin remodeling. Characteristic traits of the neuroblastoma epigenome are linked to specific neuroblastoma subtypes, allowing improved tumor diagnosis and providing novel approaches for therapeutic intervention.

After its discovery as predominant driver of familial neuroblastoma, ALK turned out to be also the most frequently somatically mutated gene in sporadic neuroblastoma. As tyrosine kinase receptor, it represented a promising therapeutic target and small molecule inhibitors were rapidly translated to clinical trials. The present state of our understanding of ALK function in normal development of sympathoadrenal cells as well as in neuroblastoma initiation, synergistic interactions between activated ALK and MYCN, and current progress in approaches to pharmacologically interfere with tumor growth is covered by Janoueix-Lerosey et al. (2018).

Pheochromocytoma and Paraganglioma

In contrast to neuroblastomas, which typically arise in infancy or very early childhood and result from somatic mutations of developmentally regulated genes, the pathogenesis of pheochromocytoma and paraganglioma is more complex. At least 40% are hereditary, and the causative mutations are largely distinct from those associated with neuroblastoma. Although a few pheochromocytomas and paragangliomas, particularly hereditary cases, present in young children, most occur in adults. There are also marked genotype-phenotype correlations that segregate the tumors into distinct "clusters" differing in predominant tumor location, catecholamine production, propensity for metastasis, and development of multiple versus solitary tumors.

Most basic research on pheochromocytomas and paragangliomas begins in the context of genotype-phenotype clusters (reviewed by Fishbein and Wilkerson (2018)). Two major clusters were initially described through transcriptional profiling of hereditary tumors (Dahia et al. 2005). "Cluster 1," characterized by hypoxic signaling pathways, initially consisted of tumors with mutations of *VHL* and of genes encoding subunits of succinate dehydrogenase, collectively referred to as *SDH*x genes. "Cluster 2" is characterized by kinase signaling pathways and initially consisted of tumors with mutations of *RET* and *NF1*. Somatic mutations of some of the same genes, especially *NF1*, *RET*, and *VHL* but not *SDH*x genes, are now frequently recognized in sporadic as well as in hereditary pheochromocytomas and paragangliomas. *NF1* mutations, the most common, are reported in 20–30% of sporadic cases. The clusters have also been expanded by discovery of new hereditary susceptibility genes and new somatic mutations not usually associated with hereditary susceptibility. The latter include occasional mutations of *HRAS* and *ATRX*. Tumors with germline or somatic mutations are assigned to clusters according to the overall match of their transcription signatures. However, these signatures are not always equivalent to those of VHL, *NF1*, or *RET*, and sub-clustering has been proposed. Mutation of *TMEM127*, which encodes a protein involved in intracellular protein trafficking, is included in cluster 2 but is not associated with RAS activation. *MAX*, which encodes a protein that heterodimerizes with and antagonizes transcriptional effects of MYC protein, is also in cluster 2. Within cluster 1, there are marked differences between tumors harboring *VHL* mutations and those with mutations of genes encoding subunits of succinate dehydrogenase (Fliedner et al. 2018). The former usually involve the adrenal and are benign, while the latter are usually extra-adrenal, often are multiple, and often metastasize, especially those tumors with *SDHB* mutations. A new group of aggressive tumors characterized by Wnt signaling pathways was described in The Cancer Genome Atlas (TCGA) publication in 2017 (Fishbein et al. 2017).

The relationships between genotype, tumor location, and catecholamine production in pheochromocytomas and paragangliomas pose conundrums from a developmental perspective. In normal SA tissues, epinephrine is produced almost exclusively in the adrenal gland, while other paraganglia produce norepinephrine and/or dopamine. This largely holds true for the corresponding tumors, except that adrenal tumors with *VHL* or *SDHx* mutations are usually noradrenergic. In addition, quantitative differences in production of catecholamines and dopamine are associated with different genotypes. Possible explanations for these associations are analyzed by Fliedner et al. (2018) who suggest that different innate sensitivities of developing chromaffin cell populations to hypoxia might exacerbate vulnerability to mutations that alter hypoxic signaling pathways and by Fishbein and Wilkerson (2018), who emphasize the primary influences of genotype.

From a clinical perspective, tumors with *SDH*x mutations are perhaps of greatest interest because they have been estimated to comprise up to 80% of familial aggregates of pheochromocytomas and paragangliomas and almost 40% of tumors that metastasize (Pasini and Stratakis 2009). The dual metabolic functions of SDH as TCA cycle intermediate and as complex ll in the mitochondrial electron transport chain suggest several hypothetical metabolic drivers of tumor development and potential therapeutic targets (reviewed by Kluckova and Tennant (2018)). First, accumulation of succinate may inhibit 2-OG-dependent dioxygenases that use 2-OG as a co-substrate and generate succinate as a byproduct. These include prolyl hydroxylase domain (PHD) enzymes that hydroxylate hypoxia-inducible transcription factors (HIFs) in the presence of oxygen, thereby tagging them for ubiquitination and degradation, and DNA hydroxylases that demethylate genomic DNA. The result is that SDH-deficient tumors show both pseudohypoxic signaling mediated by downstream targets of HIFs, and a "hypermethylator" phenotype that may inactivate tumor suppressor genes by promoter methylation. A second, more controversial, contributor to the pathogenesis of SDH-deficient tumors is overproduction of reactive oxygen species (ROS), which is postulated to result from reverse electron transport from an over-reduced ubiquinone pool between complex I/II and complex III. While ROS have been proposed to function as hypoxic signaling molecules in these tumors, the evidence reviewed by Kluckova and Tennant is inconclusive. Further, although DNA damage by ROS is thought to drive genome instability and increase mutation load in other types of tumors, pheochromocytomas and paragangliomas are known to have an extraordinarily low mutation load (Fishbein and Wilkerson 2018).

A difficulty in studies of pheochromocytoma and paraganglioma is the paucity of experimental models (Lepoutre-Lussey et al. 2018). In contrast to neuroblastomas, for which there are multiple human cell lines and mouse models, there are no human pheochromocytoma or paraganglioma cell lines, and rodent models for these tumors exist only for susceptibility genes in cluster 2. The latter includes the PC12 rat pheochromocytoma cell line (*Max* mutation) (Greene and Tischler 1976), MPC mouse pheochromocytoma cell line (*Nf1* mutation) (Powers et al. 2000), and a transgenic MEN2B mouse characterized by pheochromocytomas and malformed adrenal glands fused with sympathetic ganglia (*Ret* mutation) (Smith-Hicks et al. 2000). The inability to develop human cell lines results at least in part from the intrinsically low growth rate of pheochromocytomas and paragangliomas, which can take years for a single doubling in vivo. However, the absence of rodent models for cluster 1 tumors, particularly those with *SDHB* mutations, is perplexing. Although several types of genetically engineered mice have been generated to address this problem, none have developed pheochromocytomas or paragangliomas. One impediment to success might be that loss of the wild-type *SDHB*

allele usually occurs as a large deletion in chromosome 1p that probably also includes tumor suppressor genes. Chromosome 1p deletions are also associated with neuroblastomas and several other types of tumor.

Although there is much still to be learned about the developmental basis for differences between pheochromocytomas and paragangliomas in different locations and with different genotypes, clinical medicine has effectively utilized existing knowledge of the physiological characteristics of these tumors to develop new modalities of imaging and treatment. Taïeb and Pacak (2018) review current molecular imaging and theranostic approaches, which are largely based on phenotype and genotype correlations. Molecular imaging, also known as functional imaging, serves multiple purposes. These include discrimination of pheochromocytoma or paraganglioma from other types of masses identified by anatomic imaging (e.g., CAT scan or MRI), screening for multiple tumors and metastases, and detection of molecular targets for therapy. The last of these, detection of therapeutic targets, is the basis for theranostic imaging, which utilizes radiolabeled tracer molecules to identify tumor characteristics such as transporters or receptors that can later be targeted using the same tracers tagged with more destructive radioisotopes.

Three main attributes of pheochromocytoma and paraganglioma influence the choice of molecular imaging modality: catecholamine metabolism, somatostatin receptor expression, and glucose metabolism. Imaging focused on catecholamine metabolism exploits norepinephrine transporter, vesicular monoamine transporters, or amino acid transporters to concentrate tracer into tumor cells. The most widely utilized tracer is metaiodobenzylguanidine (MIBG), which is taken up by the norepinephrine transporter. Imaging is usually performed with MIBG tagged with ^{123}I, predominantly a gamma ray emitter. For tumor ablation, the radionuclide is ^{131}I, a strong beta particle emitter. MIBG is optimal for imaging and treatment of relatively well-differentiated tumors that produce abundant catecholamines, best exemplified by pheochromocytomas. However, it has also been useful in management of patients with neuroblastomas, especially for imaging (Park et al. 2017; Pfluger and Piccardo 2017). Methods have been proposed to increase the therapeutic efficacy of ^{131}I-MIBG in poorly differentiated tumors by upregulating norepinephrine transporter, in some cases using agents that might mimic normal developmental regulation (Martiniova et al. 2011).

Somatostatin receptor imaging is best suited to paragangliomas, especially those with SDHx mutations and those in the head and neck. The latter are often TH-negative but strongly express somatostatin receptors. Since approximately 1994, tumors with somatostatin receptors have been imaged using the OctreoScan, a patented scintigraphic technique using the ligand ^{111}In-DPTA-D-Phe-1-octreotide and providing a two-dimensional image (Maxwell and Howe 2015). Over the past decade, new radioligands and three-dimensional detection technology such as positron emission tomography (PET)/computed tomography (CT) have greatly increased the sensitivity of this imaging modality. A current approach employs the positron emitter ^{68}Ga linked to 1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetic acid (DOTA) and any of several somatostatin analogs, e.g., ^{68}Ga-DOTATATE, ^{68}Ga-DOTATOC, and ^{68}Ga-DOTANOC. In these constructs, the DOTA compound is a chelator that links the radioisotope to the peptide ligand. For tumor ablation (peptide radioreceptor therapy, PRRT), ^{68}Ga is replaced with ^{177}Lu, a short-range beta emitter, or ^{90}Y, a long-range beta emitter.

Imaging focused on glucose metabolism is not specific for pheochromocytomas and paragangliomas and is utilized for many types of tumors. The most commonly used tracer is the positron emitter ^{18}F-fluorodeoxyglucose (^{18}F-FDG). As discussed by Taïeb and Pacak (2018), ^{18F}FDG PET has been particularly valuable in localizing *SDHx*-related tumors, especially those with metastases, but might soon be supplanted by somatostatin receptor imaging. The sensitivity of this modality for tumors with *SDHx* mutations might result in part from upregulation of glucose transporters, which are downstream targets of hypoxia-inducible transcription factors, in pseudohypoxic tumors (Baumann et al. 2007).

Oxygen sensing and its pathology in the carotid body and chromaffin cells

In contrast to the chromaffin cells of the adrenal medulla, the type I (or glomus) cells of the carotid body and similar structures located along the branches of the vagus nerve (e.g., aortic bodies) are negligible in their contribution to circulating catecholamines but rather serve a sensory function and utilize their transmitters to excite sensory nerve fibers and for paracrine regulatory mechanisms. The carotid body is the primary sensor of peripheral arterial pO_2 that drives enhanced ventilation in the setting of lowered oxygen tension without concomitant increase in pCO_2, as is in the case of high altitude. This oxygen sensor function was originally proposed by the Spanish histologist Fernando de Castro as early as 1928 and was confirmed soon after that by the Belgian physiologists Jean-Francois Heymans and Corneille Heymans (father and son), resulting in awarding the Nobel Prize in Physiology or Medicine to Corneille Heymans in 1938 (de Castro 2009). Even beyond the end of that century, however, the molecular mechanisms of oxygen sensing in the carotid body remained unclear. Many, often complimentary but also contradictory hypotheses, have been posed and tested, but consensus was not reached. Indeed, the Novartis Foundation Symposium *Signalling Pathways in Acute Oxygen Sensing*, held in London in 2005, was opened by the chair, Michael Duchen, by stating: “This field has been almost unique, it seems to me, in the level of disagreement

and failure to reach consensus among researchers." (Duchen 2006). On this background, it is a fundamental breakthrough to tackle the unique properties of the carotid body by an unbiased approach at expression level. Comparing the transcriptome of the carotid body (oxygen sensor) and the adult adrenal medulla (closely related but not a professional oxygen sensor) unexpectedly revealed a selective high expression of a member of the olfactory receptor family, olfr78, which proved to represent a G-protein-coupled short-chain fatty acid receptor monitoring lactate accumulation in the carotid body (Chang et al. 2015). In this issue, Zhou and Matsunami (2018) comprehensively summarize the most recent developments in this line of research, extending transcriptome analysis to single-cell level. This approach corroborated the finding of overrepresentation of olfr78, but also added new players into the game, i.e., the atypical subunits of the mitochondrial electron transport chain Ndufa4l2 and Cox4i2 (Zhou et al. 2016; Gao et al. 2017). Zhou and Matsunami (2018) also point towards the enormous potential of this approach, if put on a larger scale, in distinguishing novel cell types, an issue being debated for a long time (e.g., type A and type B glomus cells in the rat carotid body, McDonald and Mitchell (1975)), but likewise never reaching consensus.

At the next higher structural organization level, Jose Lopez-Barneo steps in and brings together what expression of atypical mitochondrial electron transport chain subunits of complex IV might mean for production of reactive oxygen species (ROS) further upstream in the transport chain, and how this might finally translate into activation of sensory nerve fibers through what he calls "chemosensory synapses" (López-Barneo 2018). This review broadens the view upon the carotid body in that not only excitation of sensory nerve fibers and respiratory reflexes are considered as responses to hypoxia, but also signaling through "chemo-proliferative synapses" to adjacent stem cells which represent a population of the glial-like type II (or sustentacular) cells that partially enwrap type I cells. In chronic hypoxemia, these cells expand, giving rise to both type I and type II cells and resulting in gross hypertrophy of the carotid body that can even be measured by weighing of carotid bodies obtained at autopsy (Smith et al. 1982). Lopez-Barneo's group was the first to discover this stem cell niche in the carotid body (Pardal et al. 2007), which nicely correlates with the just recently identified origin of adrenal chromaffin cells from Schwann cell progenitors (Furlan et al. 2017).

This proliferative response seen in chronic hypoxia is not so evident when there is chronic, repeated exposure to acute episodes of hypoxia, a condition termed "intermittent hypoxia" (Pawar et al. 2008; Welch et al. 2017). Patients experience intermittent hypoxia in sleep apnea, a disease characterized by periodic cessation of breathing during sleep. These patients exhibit heightened cardiovascular sympathetic activity and hypertension, and activation of the carotid body chemoreflex is considered as the major driver of these autonomic abnormalities (Prabhakar 2016). In this issue, Nanduri Prabhakar focusses upon the initial mechanisms within the carotid body leading to this abnormal activation (Prabhakar et al. 2018). As in acute oxygen sensing, ROS are involved, but they are not exclusively derived from the mitochondrial respiratory chain and are not the only gaseous messengers operating in this scenario. Activities of pro-oxidant enzymes are upregulated through HIF-dependent pathways and anti-oxidant enzymes are downregulated. The in-depth review covers the complex interplay between elevated ROS and the gaseous messenger carbon monoxide (CO) and hydrogen sulfide (H_2S) (Prabhakar et al. 2018).

Like the type I cells of the carotid body adrenal chromaffin cells can physiologically respond to acute hypoxia, but primarily during early development. As reviewed by Nurse et al. (2018), hypoxia triggers catecholamine release in perinatal chromaffin cells, when the innervation of the adrenal medulla is not yet functional. Adrenal chromaffin cells downregulate their sensitivity to acute hypoxia in parallel to splanchnic nerve innervation, but they can gradually regain it after denervation. Nurse et al. (2018) discuss the mechanism of hypoxia-triggered catecholamine release by chromaffin cells, its regulation by splanchnic nerve innervation, and the plasticity of chromaffin cells following chronic and intermittent hypoxia. While the precise mechanism of acute oxygen sensing is still unclear and controversially discussed, a recent study by Gao et al. (2017) provided novel insight into putative markers of oxygen sensitivity by comparing the transcriptome of SA cells with different sensitivities to oxygen (carotid body type I cells +++, adult chromaffin cells +, neurons of the superior cervical ganglion −). The authors highlight a gene expression signature putatively linked to oxygen sensing, which is shared by carotid body type I cells and chromaffin cells. It is in particular characterized by elevated levels of HIF-2α, pyruvate carboxylase, and the atypical mitochondrial electron transport subunits Ndufa4l2, Cox4i2, and Cox8b. From a pathobiological point of view, this characteristic molecular profile might be closely associated with the vulnerability of these cells with regard to the occurrence of cluster I (pseudohypoxic) pheochromocytoma/paraganglioma. The molecular players, mechanisms, and the plasticity of oxygen sensing are furthermore of high relevance in the context of clinical conditions such as obstructive lung disease, sleep apnea, and hypertension.

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