REVIEW

Role of bone morphogenetic proteins in adrenal physiology and disease

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Abstract

Bone morphogenetic proteins (BMPs) are members of the transforming growth factor- β superfamily of ligands that impact on a multitude of biological processes including cell type specification, differentiation and organogenesis. Furthermore, a large body of evidence points towards important BMP-dependent mechanisms in tumorigenesis. In accordance with their diverse actions, BMPs have been demonstrated to serve as auto-, para- and endocrine modulators also in a number of hormonal systems. In this review, we highlight novel aspects of BMP-dependent regulatory networks that pertain to adrenal physiology and disease, which have been uncovered during recent years. These aspects include the role of BMP-dependent mechanism during adrenal development, modulating effects on catecholamine synthesis and steroidogenesis and dysregulation of BMP signalling in adrenal tumorigenesis. Furthermore, we summarize potential therapeutic approaches that are based on reconstitution of BMP signalling in adrenocortical tumour cells.

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Introduction

Bone morphogenetic proteins (BMPs) are cytokines belonging to the transforming growth factor- β (TGF- β) superfamily, which have been originally identified by their capacity to induce endochondral bone formation (Hogan 1996). On an evolutionary scale, BMP signalling is highly conserved over at least 700 million years (Blitz & Cho 2009). Accordingly, BMP-dependent functions are involved in a multitude of biological processes. Among their many functional properties, BMPs control cell type specification, differentiation and organogenesis (von Bubnoff & Cho 2001). Furthermore, they have been demonstrated to act as auto- or paracrine modifiers of tumour growth and to have functional impact in a number of cancer entities (Langenfeld *et al.* 2003, Hsu *et al.* 2005).

BMPs are synthesized as large precursor proteins and are proteolytically cleaved upon dimerization to yield active proteins. These mature dimers initiate signalling by binding to transmembrane serine/threonine kinase receptors. Notably, BMPs display different binding affinities to individual BMP and activin receptors (ActRs). BMP-6, for example, primarily binds to ActRI (Alk 2), whereas BMP-2 and BMP-5 mainly signal through BMPRIa (Alk 3) and BMPRIb (Alk 6; Liu *et al.* 1995, Yamashita *et al.* 1995). Upon ligand

binding, type II receptor transphosphorylates type I receptor, which in turn phosphorylates receptor-regulated SMAD proteins. These receptor SMADs (i.e. SMAD1, SMAD5 and SMAD8 for BMP signalling) heterodimerize with the common partner SMAD4 and translocate into the nucleus, where they activate or repress BMP target genes (e.g. inhibitor of DNA-binding proteins, ID proteins) depending on the recruitment of further co-factors (von Bubnoff & Cho 2001, Chen *et al.* 2004). In contrast, TGF- β signalling is primarily transduced by receptor SMADs SMAD2 and SMAD3, which are activated after TGF- β binding and receptor complex formation (TGF- β R2 with TGF- β R1 (Alk 5), Alk 1 or Alk 2).

BMPs are under tight regulation through proteins with antagonistic properties including extracellular noggin (Zimmerman *et al.* 1996) and follistatin (Iemura *et al.* 1998, Phillips & de Kretser 1998) and intracellular inhibitory SMADs among others (Hayashi *et al.* 1997, Imamura *et al.* 1997, Nakao *et al.* 1997). Besides this canonical pathway, BMPs have been reported to modulate also a number of other cascades, including the mitogen-activated protein kinase–extracellular signal-regulated kinase (MAPK–ERK) pathway, AKT–phosphatidylinositol 3-kinase (PI3K) pathway, β -catenin/WNT pathway (Ghosh-Choudhury *et al.* 2002, He *et al.* 2004, Nohe *et al.* 2004) and the stress activated protein kinase–cJUN N-terminal kinase (SAPK/JNK) pathway

(Goto *et al.* 2009). This crosstalk can partially explain the observed complexity, diversity and flexibility of BMP-induced cellular responses (Guo & Wang 2009).

In accordance with their diverse actions, BMPs have been demonstrated to serve as auto-, para- and endocrine modulators also in a number of hormonal systems. In this review, we will specifically highlight novel aspects of BMP-dependent regulatory networks that pertain to adrenal physiology and disease, which have been uncovered during recent years.

BMPs in adrenal development

BMPs have been demonstrated to be involved in a variety of key developmental processes such as dorsal–ventral axis specification, epithelial–mesenchymal interactions and apoptosis (Liu & Niswander 2005). However, as most genetically modified animal models with disrupted BMP signalling are prone to early intrauterine death (Mishina *et al.* 1995, Huber 2006), *in vivo* studies detailing BMP-dependent mechanisms required for adrenal gland development are sparse. Indirect evidence for the importance of BMP signalling in adrenal development has been gathered in sheep of the Booroola phenotype. In these animals, which are mainly defined by an increased rate of ovulation and an increased litter size, a spontaneously occurring mutation in *BMPR1B* had been identified (Mulsant *et al.* 2001). Interestingly, this mutation which results in a single amino acid substitution of the *BMPR1B* and reduction of BMP-4-induced signalling in granulosa cells (Fabre *et al.* 2003) is associated with smaller adrenal size in affected young animals, whereas the weight of heart, liver, lungs and kidneys was found to be unchanged (Souza & Baird 2004).

In addition to these gross anatomical observations, more refined mechanistic experiments have provided support for BMP-dependent regulation of adrenal development, specifically of adrenomedullary cell specification from the neural crest. The neural crest itself emerges from the dorsal surface of the neural tube and generates a population of multipotent progenitor cells, which have the capacity to give rise to both sympathetic neurons and chromaffin cells located in the adrenal medulla (Huber 2006). Chromaffin cell differentiation is believed to involve the inhibition of terminal neuronal differentiation with downregulation of neurofilament expression and lack of neurite growth (Michelsohn & Anderson 1992). Tissues surrounding neural crest cells and sympathoadrenal progenitor cells during their migration and at their final locations are considered to be important for the induction of a sympathetic neuronal or chromaffin cell phenotype. In this context, it is widely assumed that BMPs from the wall of the dorsal aorta in concert

with cell-specific transcription factors are required for the specification of sympathoadrenal cells (Schneider *et al.* 1999). This hypothesis is based on an increasing body of evidence gathered in *in vitro* and *in vivo* models: BMP-2, BMP-4 and BMP-7 have been demonstrated to increase the number of catecholaminergic and neuronal cells in culture of avian neural crest cells, and overexpression of BMP-4 in chicken embryo is sufficient to induce cells ectopically expressing sympathoadrenal-specific markers (Reissmann *et al.* 1996). Similarly, introduction of a constitutively active type I BMP receptor in neural crest cells is sufficient to induce the development of adrenergic cells (Varley *et al.* 1998). However, BMP-2 and BMP-4 can also induce apoptosis in cultured sympathoadrenal progenitor cells, which are counteracted by addition of fibroblast growth factor and nerve growth factor. It has been proposed that exposure of progenitor cells to pro-apoptotic factors such as BMP-2/BMP-4 make them transiently dependent on other target-derived factors for their survival and development (Song *et al.* 1998). These apparently contradictory functional properties of BMP-2/BMP-4 could, thus, present an important mechanism at an earlier time point during adrenomedullary development to pre-sort a heterogeneous cell population for a subsequent more effective specification.

In addition to the expression of BMPs in the dorsal aorta and their ability to influence progenitor cell development, members of the BMP family have been discovered to be expressed also in the adrenal cortex. In accordance with the role of BMPs during early development to differentiate progenitor cells towards an adrenergic cell type, BMP-4 originating from adrenocortical cells is able to induce tyrosine hydroxylase (TH) expression in avian TH-negative sympathoadrenal progenitor cells (Huber *et al.* 2008). Accordingly, noggin, an extracellular acting BMP antagonist, could be demonstrated to prevent the increase of TH-positive cells in adrenal explants without affecting cell proliferation. Overall, these data highlight the importance of timely and spatially fine-tuned distribution of BMPs for the generation of precursor cells, which are required for the proper development of chromaffin cells within the adrenal gland (Fig. 1A). Furthermore, these observations do not apply for all TGF- β family members in general. TGF- β (isoforms 2 and 3), for example, is synthesized by cells in the wall of the dorsal aorta as well as at later stages in both cortical and migrating chromaffin cells within the developing adrenal gland. However, the *in vivo* effects of TGF- β on chromaffin cells are limited to their inhibiting action on cell proliferation in contrast to that of chromaffin cell phenotype determination as observed for BMPs (Combs *et al.* 2000, Huber *et al.* 2002).

Figure 1 Roles of BMP during adrenal development (A) and endocrine function during adulthood (B). (A) Sympathoadrenal progenitor cells undergo differentiation to adrenomedullary chromaffin cells induced by BMP-2/BMP-4/BMP-7 and secreted by the dorsal aorta and adrenal cortex respectively. (B) Summary of various interrelations of BMPs with adrenocortical steroidogenesis and adrenomedullary catecholamine biosynthesis (for details see text). (MC2-R, melanocortin 2 receptor/ACTH receptor; Ang II, angiotensin II; AT1-R, angiotensin II type 1 receptor; —■, inhibition; —►, stimulation).

BMP-dependent mechanisms in adrenal physiology

Adrenal medulla

In addition to the developmental context, BMPs secreted locally by the adrenal cortex are good candidates that could also modify the cellular fate of tissue progenitor cells in the adult adrenal medulla. Furthermore, direct functional effects of BMPs on adrenomedullary hormone secretion have been postulated. Owing to the close anatomic connection, functional interdependence between the adrenal cortex and the adrenal medulla had been suspected already in the 1970s (Wurtman *et al.* 1972). Glucocorticoids secreted by the adrenal cortex were found to be transported mainly through a corticomedullary portal system into the medulla to stimulate the synthesis of catecholamine enzymes and, thus, maintain the phenotype of chromaffin cells (Shepherd & Holzwarth 2001). Interestingly, BMPs are thought to influence catecholamine production of adrenomedullary cells via an indirect mechanism through interfering with steroid hormone-dependent signalling pathways (Kano *et al.* 2005, Goto *et al.* 2009). In rat adrenomedullary PC-12 cells, it has been demonstrated that BMPs can inhibit dopamine expression and reduced DOPA decarboxylase mRNA expression in a dose-dependent manner, thereby acting antagonistic to glucocorticoids. Conversely, glucocorticoids could be shown to inhibit BMPRII, Alk 2 as well as Alk 3 expression to maintain catecholamine production, providing a complex but finely regulated system for

glucocorticoid-dependent catecholamine synthesis (Kano *et al.* 2005). Similarly to glucocorticoids, BMPs have the potential to influence aldosterone-dependent dopamine production in PC-12 cells. Aldosterone is capable of functioning through classic genomic mechanisms via stimulation of the mineralocorticoid receptor or to elicit non-genomic signals via the regulation of various second-messenger pathways (Funder 2005). In this context, BMP-4 has been found to enhance aldosterone-induced non-genomic actions on TH and dopamine synthesis via the RHO/SAPK/JNK pathway, but was not able to stimulate production in the absence of aldosterone (Goto *et al.* 2009).

Overall, these findings demonstrate a functional interrelationship between adrenocortical cells that produce and secrete both steroid and BMP hormones, and adrenomedullary cells, thereby modulating and fine-tuning adrenal catecholamine output (Fig. 1B).

Adrenal cortex

In addition to their modulating effects on steroid hormone-dependent mechanisms, BMPs have also been demonstrated to directly impact on steroidogenesis. Aldosterone synthesis and secretion, which is primarily stimulated by angiotensin II (Ang II), potassium, and – within short-term regulation – ACTH, have been recognized to be further modulated through an increasing number of growth factors and cytokines (Aguilera 1993, Ehrhart-Bornstein *et al.* 1998). One of the rate-limiting steps for aldosterone production is the conversion of deoxycorticosterone to aldosterone, which is catalyzed by aldosterone synthase.

Promoter activation of *CYP11B2*, which encodes aldosterone synthase, can be induced within minutes upon specific stimulation (Spyroglou *et al.* 2009). ACTH mainly signals through the cAMP/protein kinase A pathway, whereas Ang II binds to the Ang II type 1 receptor (AT1-R), which is a G_q-protein-coupled receptor and activates inositol trisphosphate/Ca²⁺, the diacylglycerine (DAG)/protein kinase C (PKC) pathway and finally the MAPK–ERK pathway for signalling (Holland *et al.* 1995, Foster *et al.* 1997). *In vitro* experiments in NCIh295R adrenocortical tumour cells have revealed BMP-6-induced and SMAD1/SMAD5/SMAD8-mediated augmentation of aldosterone secretion through a crosstalk with Ang II-dependent pathways (Suzuki *et al.* 2004, Inagaki *et al.* 2006). On the contrary, potassium-induced aldosterone production was not found to be influenced by BMP-6. These results were endorsed by experiments following inhibition of endogenous BMP-6 through neutralizing antibodies, which reduced Ang II-induced aldosterone secretion but not potassium-induced aldosterone secretion. These results indicate the possibility that endogenous BMP-6 produced by adrenocortical cells could play an important autocrine role in modulating the steroidogenic actions of Ang II.

Following these findings, it had been suspected that the phenomenon called ‘aldosterone breakthrough’, which is defined as a state of sustained aldosterone synthesis from the adrenal cortex during long-term treatment with angiotensin converting enzyme (ACE) inhibitors and/or AT1-R blockers (Staessen *et al.* 1981), could also be mediated by BMP-6. In accordance with this hypothesis, Otani *et al.* (2008) found that expression of BMP-6 and its receptors was decreased by long-term Ang II exposure but restored by co-treatment with AT1-R blockers. Furthermore, neutralization of Alk 2 and BMP-6 function attenuated the escaping effect from aldosterone suppression by AT1-R blockers. Given the stimulatory effects of BMP-6 on Ang II-induced aldosterone secretion, it is well conceivable that this auto/paracrine mechanism could play an important role in fine-tuning of aldosterone release also in this clinically relevant context.

In contrast to BMP-6, we could recently demonstrate that both BMP-2 and BMP-5 are able to overall suppress forskolin-induced steroidogenesis in NCIh295R cells (Johnsen *et al.* 2009), similar to what had been reported earlier for TGF-β1 (Liakos *et al.* 2003). Specifically, secretion of aldosterone, cortisol, and dehydroepiandrosterone-sulphate, was reduced by BMP-2 and BMP-5 in a dose-dependent manner. In line with this finding, expression levels of several steroidogenic enzymes, catalyzing the different steps of hormone production, as well as the ACTH receptor (melanocortin 2 receptor, MC2-R), were also inhibited by BMP-2 and BMP-5 treatment in a dose- and time-dependent

manner (Johnsen *et al.* 2009). We further investigated whether BMP-induced decrease in steroidogenesis would be dependent from the transcription factor SF-1, which has been demonstrated to regulate the expression of key determinants of gonadal and adrenal steroidogenesis (Sarkar *et al.* 2000). Detailed promoter analysis of the *MC2-R*, utilizing 5′ deletion constructs as well as those harbouring point mutation in SF-1-binding sites, revealed an SF-1-independent mechanism of BMP inhibition on the *MC2-R* promoter. Instead, two consensus sites for E-box elements were identified in the last 64 bp fragment of the *MC2-R* promoter, which could potentially be targeted by transcription factors of the basic helix-loop-helix (bHLH) family. Interestingly, a subgroup of this family, the ID proteins, function as negative regulators of the bHLH proteins and are targets of BMP-mediated gene transcription. Thus, it is tempting to speculate that the observed BMP-induced inhibition of *MC2-R* expression could be mediated by upregulation of ID protein levels. Only recently, in accordance with this concept, studies have reported the involvement of bHLH factors in glucocorticoid secretion by their interaction with the steroid acute regulatory protein (*StAR*) promoter (Son *et al.* 2008, Ratajczak *et al.* 2009) and the regulation of tissue-specific expression of the *MC2-R* promoter by E-Box elements (Blondet *et al.* 2004). Similarly, TGF-β has been recognized to inhibit adrenal steroidogenesis through downregulation of *StAR* (Brand *et al.* 1998). In addition, TGF-β-dependent inhibition of *CYP11B1* and *CYP11B2* promoter activity could be demonstrated (Liakos *et al.* 2003). Interestingly, while downregulation of *StAR* was found to be a SMAD dependent but SF-1-independent mechanism, TGF-β-mediated influence on *CYP11B1/CYP11B2* promoter displayed a SMAD-independent mode of action (Liakos *et al.* 2003).

In summary, BMPs are important modulators of both catecholamine synthesis and steroidogenesis. They own the potential to govern hormone production as well as enzyme and receptor expression and, moreover, are able to modulate steroidogenic actions by their interactions with involved downstream pathways. The final functional impact, however, depends on the individual member of the BMP family and the particular underlying environmental and cellular conditions, such as presence of participating co-factors and the type and development stage of target cells (Fig. 1B).

Role of BMPs in adrenal tumorigenesis

Pathophysiological relevance

A clear indication that BMP signalling pathways can contribute to carcinogenesis comes from genetic studies of familial cancer syndromes. For example, familial juvenile polyposis, an inherited hamartomatous

polyposis syndrome with a high risk for colon cancer, has been associated with mutations in *SMAD4* or *BMPRIA* (Howe *et al.* 1998, Zhou *et al.* 2001). Beyond these familial forms, aberrations in BMP signalling pathways occur in various sporadic human cancers including several endocrine tumour entities such as those deriving from the ovary (Theriault *et al.* 2007, Le Page *et al.* 2009), the pituitary (Paez-Pereda *et al.* 2003, Giacomini *et al.* 2007) and the thyroid (Franzen & Heldin 2001). However, the expression levels of individual BMPs not only vary among tumour tissues but each BMP also differs in its functional properties depending on the cell type and tumour stage (Langenfeld & Langenfeld 2004, Hsu *et al.* 2005, Langenfeld *et al.* 2006). In line with this notion, it has been shown that BMP-2 and BMP-3 are down-regulated in prostate (Horvath *et al.* 2004) and in colorectal cancer (Loh *et al.* 2008), whereas other members are up-regulated, such as BMP-4 in colorectal tumours or BMP-2 in non-small cell lung carcinomas (Langenfeld *et al.* 2003, Noshio *et al.* 2005). Moreover, while several *in vitro* studies indicate that BMP-2 acts as a growth suppressor in a variety of tumour cell lines (Ghosh-Choudhury *et al.* 2000, Mathura *et al.* 2000, Wong *et al.* 2003, Otani *et al.* 2007), there is also evidence that BMP-2 has mitogenic properties in specific lung cancer cell lines (Langenfeld *et al.* 2003) and can stimulate angiogenesis in developing tumours (Langenfeld & Langenfeld 2004).

In a recent study detailing expression analysis of members of the TGF- β superfamily in human adrenal tissues, we could demonstrate downregulation or loss of expression of BMP-2 and BMP-5 in adrenocortical carcinoma samples in comparison to normal adrenal glands. This lower expression, which was also evident in two adrenocortical cancer cell lines (NCIh295R and SW13), was accompanied by a reduced activity of downstream pSMAD1/pSMAD5/pSMAD8 and ID proteins. BMP-2/BMP-5 treatment of the adrenocortical tumour cell lines resulted in a dose- and time-dependent reduction of cellular proliferation and viability, demonstrating the general potential of BMPs to inhibit adrenocortical tumour growth (Johnsen *et al.* 2009). Treatment of adrenocortical tumour xenografts hosted in nude mice with rhBMP-5 further reinforces BMP-mediated growth decelerating properties. These BMP-induced effects on cell proliferation may be partially explained by their ability to modulate insulin-like growth factor (IGF)-mediated signalling. In fact, it has been shown that BMPs are able to counteract the PI3K/AKT pathway, which mediates IGF-dependent actions on proliferation, apoptosis and cell survival in the adrenal cortex (Doepfner *et al.* 2007, Giulia *et al.* 2008). Notably, IGF2 is often overexpressed in adrenocortical tumours and carcinomas and is regarded as one of the most important growth factors for adrenal

tumorigenesis (Gicquel *et al.* 1994, Giordano *et al.* 2003). For neural cell growth, an IGF/EGF-dependent activation of AKT/ERK kinases has been postulated, which inhibits BMP-induced SMAD signalling through phosphorylation of SMAD proteins within their linker regions, resulting in a diminished nuclear translocation of SMAD heterodimers (Pera *et al.* 2003). Whether this mechanism also holds true in the context of adrenal tumorigenesis is yet to be elucidated.

Potential therapeutic implications

The shortcoming in detailed knowledge of underlying molecular pathways involved in adrenal tumorigenesis contributes to the lack of target-specific and patient-tailored therapies, which are therefore associated with substantial toxic side effects (Allolio & Fassnacht 2006, Terzolo *et al.* 2007). In this context, the finding of abrogated BMP signalling in adrenocortical carcinomas

Figure 2 Strategies to enforce BMP expression in adrenocortical tumour cells. (A) Enhancement of endogenous BMP expression through promoter demethylation by 5-aza-2'-deoxycytidine (5-AZA), thereby inhibiting DNA methylation transferases (DNMTs). (B) Upregulation of BMP expression through PKC pathway activation by phorbol ester PMA. Retinoid acid (RA) treatment for direct BMP promoter activation and upregulation of GATA-6 transcription with secondary stimulation of BMP expression. (C) Direct gene delivery for BMP overexpression via liposomal or viral vectors. (TF, transcription factors; CH3, methylation groups; —■, inhibition; —►, stimulation).

(ACC) could provide a novel target for pharmacological interventions (Fig. 2).

Specifically, restoration of silenced BMP expression could be aimed for to inhibit adrenocortical tumour growth. While hypermethylation of BMP-2 promoter regions has been reported to be the cause of diminished BMP-2 expression in prostate and gastric tumours (Horvath *et al.* 2004, Wen *et al.* 2006), this could not be demonstrated in ACC. However, *in vitro* treatment of NCIh295R cells with the demethylation agent 5-aza-2'-deoxycytidin (5-AZA) was followed by a decrease in BMP-2 promoter methylation, which was further associated with re-expression of endogenous BMPs and activation of downstream IDs (Johnsen *et al.* 2009). Whether these effects are the direct consequence of BMP-2 promoter demethylation or that of treatment-induced global epigenetic modifications cannot be answered from these *in vitro* experiments. However, despite its unspecific mode of action, 5-AZA-dependent treatment protocols for myelodysplastic syndrome and chronic myelomonocytic leukaemia have been associated with a rather restricted profile of documented side effects (Jabbour *et al.* 2008). Whether demethylation-based therapies can alter the phenotype of adrenocortical cancer in a clinically meaningful manner remains to be determined in future studies in appropriate *in vivo* models.

Another interesting approach to reactivate BMP signalling in adrenocortical tumour tissues could rely on stimulation of BMP expression through transcription factors such as the GATA-6 or through pharmaceutical agents including 9-*cis*-retinoid acid (RA) and PKC activators (such as the phorbol ester PMA), which have been described to stimulate BMP expression in extra-adrenal tissues (Sakaue *et al.* 1996, Sugiura 1999, Helvering *et al.* 2000). In line with these reports, overexpression of GATA-6 in NCIh295R cells was able to reconstitute BMP-5 expression and induced BMP-2 promoter activity, and similar effects were observable upon treatment with 9-*cis*-RA and PMA (Johnsen *et al.* 2009). Retinoid acids are under investigation in clinical trials for the chemical prevention of breast, skin and ovarian cancers (Lippman *et al.* 1995, Lotan 1995, 1996), whereas bryostatin-1, a specific and potent PKC activator, has revealed anti-tumour effects in lung and prostate tumours (Mohammad *et al.* 2000, Gonelli *et al.* 2009). Thus, it is conceivable that similar approaches might have beneficial effects also in the treatment of ACC patients.

Gene delivery that results in reconstitution of BMP expression in target tissues has been described in a growing body of literature in the fields of orthopaedics, orthodontics and tissue engineering (Martinek *et al.* 2003, Gugala *et al.* 2007, Chen *et al.* 2008). Recently, retroviral vectors have been found to be an efficient tool forcing BMP-2 expression in chondrogenic cells and

primary articular chondrocytes (Vogt *et al.* 2008, 2009). Similar results have been obtained with adenoviral vectors yielding high transduction efficiency and long-term gene expression of BMP-2 in autologous bone marrow mesenchymal stem cells (Chen *et al.* 2008) and primary osteoblast cultures (Egermann *et al.* 2006). Notably, besides liver targeting, adenoviruses have been demonstrated to have a natural tropism for the adrenal gland, as shown after systemic administration in mice and in non-human primates (Barzon *et al.* 2004), and adenovirus-based approaches can achieve high levels of transduction efficacy and successful adenoviral gene transfer into adrenocortical cells (Tajima *et al.* 1999, Alesci *et al.* 2002, Wolkersdorfer *et al.* 2002, Matkovic *et al.* 2009). In general, efficacy and specificity of delivery tools to target tumour sites are improving. These approaches include copolymer-protected vectors (Schillinger *et al.* 2008), liposomal tools with tumour-targeting affinities (Templeton 2009) and viral vectors with enhanced anti-tumour properties (Alemany 2009). Thus, these findings point towards potential approaches to restore endogenous BMP levels in adrenocortical carcinoma cells for the modulation of their functional properties and malignant potential providing hope for a specific and potent therapy of ACC patients with reduced side effects in the future.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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