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Factors Affecting the Nuclear Localization of β-Catenin in Normal and Malignant Tissue

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ABSTRACT

The canonical Wnt signaling pathway has been the focus of intensive research because of its frequent dysregulation in human cancers. Much of this has been directed towards the aberrant expression and/or activity of the central mediator of this pathway, β-catenin. In particular, the nuclear localization of β-catenin and subsequent inappropriate activation of TCF/LEF-mediated transcription appears to be an important process in both the establishment and maintenance of cancer stem cells. Despite this, the exact mechanisms controlling β-catenin nuclear localization in both normal and malignant cells are poorly understood. This prospect article brings together the many mechanisms previously reported to regulate the nuclear localization of β-catenin and how they are relevant to cancer. J. Cell. Biochem. 115: 1351–1361, 2014.
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β-Catenin is a member of the armadillo family of proteins and has dual functionality. At the cell membrane it serves as an important junctional component by linking the classical cadherins to the actin cytoskeleton through binding of α-catenin; alternatively β-catenin has also been well characterized as a central mediator of canonical Wnt signaling [Clevers and Nusse, 2012]. In the absence of an external Wnt ligand this pathway is held constitutively inactive through continual degradation of non-junctional β-catenin. Degradation is mediated by a destruction complex consisting of CK-1, GSK-3β, Axin and APC. CK-1 and GSK-3β phosphorylate β-catenin on serine (S33/37/45) and threonine (T41) residues, creating recognition sites for β-TrCP, ultimately targeting β-catenin for proteasome-mediated degradation [Aberle et al., 1997].

Activation of the canonical pathway occurs upon binding of a Wnt ligand to a transmembrane receptor complex consisting of the Frizzled and LRP family of receptors. Activation of this complex causes the entire destruction complex to bind phosphorylated LRP5/6, where β-catenin is still phosphorylated, but can no longer be ubiquitinated by β-TrCP [Li et al., 2012a]. The saturated destruction complex fails to bind newly synthesized β-catenin leading to its cytosolic accumulation and eventual translocation to the nucleus through a largely undefined mechanism (discussed below). Here, β-catenin co-activates Wnt target genes such as *MYC* [He et al., 1998] and *CCND1* (*Cyclin D1*; [Shtutman et al., 1999]) through binding of the transcription factors TCF/LEF, in a complex also requiring BCL-9 and pygopus [Kramps et al., 2002]. Activation of this pathway is critical for the development and maintenance of normal stem cells and is therefore frequently implicated during malignant transformation.

REGULATION OF NUCLEAR β-CATENIN LOCALIZATION IN NORMAL AND MALIGNANT CELLS

The standard Wnt signaling model described above predicts that stabilization of β-catenin automatically results in its translocation to the nucleus where it participates in transcriptional activation; however observations in a number of contexts show that an additional level of regulation exists in which nuclear accumulation of β-catenin is controlled independently of its stabilization. Such regulation seems to be particularly important for normal stem/progenitor cells. An important study from the Staal group has previously demonstrated that the precise dose of Wnt signaling (as measured in Axin2/conductin$^{LacZ/+}$ Wnt-reporter mice) is critical for

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correct hematopoietic stem/progenitor cell (HSPC) function and lineage-fate decisions [Luis et al., 2011]. Very low doses of Wnt signaling (and presumably nuclear β-catenin) were required to sustain normal hematopoiesis, whilst very high Wnt signaling levels impaired HSPC self renewal and differentiation. Further studies from embryonic and skin stem cells have similarly demonstrated the importance of Wnt signaling dose for normal development thus reaffirming the importance of regulating the correct level of nuclear β-catenin [Gaspar and Fodde, 2004; Silva-Vargas et al., 2005].

There is evidence that this control of nuclear entry can be lost in malignant cells. β-Catenin is deregulated in acute myeloid leukemia (AML) where its overexpression confers an adverse prognosis for the patient [Mikesch et al., 2007]. Recent data from our lab has revealed a differential capacity between normal HSPC and leukemic cells in the ability to control nuclear β-catenin levels. Normal human HSPC strongly resist the nuclear localization of β-catenin whereas a high frequency of leukemic cell lines freely translocate cytosolic β-catenin (and γ-catenin) in response to stabilized cytosolic levels. This in turn may explain the high frequency of primary AML blasts showing nuclear β-catenin [Morgan et al., 2012] (Fig. 1). These data suggest the existence of another rate-limiting step in Wnt signaling which tightly controls nuclear localization of β-catenin in normal hematopoietic cells but is dysregulated during leukemogenesis.

Such observations are not restricted to hematopoietic tissue. A study by Phelps and colleagues using both human cell lines and a zebrafish model of colon adenoma progression showed that initially APC loss was insufficient to stimulate nuclear β-catenin translocation. Nuclear accumulation of β-catenin and subsequent adenoma progression only presented upon the onset of secondary oncogenic *K-Ras* mutations in a RAF1/RAC1 dependent mechanism [Phelps et al., 2009]. This suggests that β-catenin stabilization through APC loss alone is insufficient for β-catenin nuclear translocation and further dyregulation is required. Similar observations have been made in primary human colon tissue. For example, Kobayashi et al. [2000] found that cytoplasmic β-catenin expression was significantly higher in adenoma and carcinoma tissue compared with normal mucosa, but was not correlated with nuclear levels. Furthermore, nuclear β-catenin was observed only in advanced carcinoma tissue and not in any adenomas of familial adenomatous polyposis coli (FAP) or sporadic origin. Anderson et al. [2002] showed that, similar to normal tissue, nuclear β-catenin was absent from over 90% of polyps taken from FAP patients but present in around 50% of carcinoma cases. Many other studies have indicated an under-represented level of nuclear β-catenin relative to cytoplasmic/membrane level in colorectal tissue [Inomata et al., 1996; Brabletz et al., 1998; Brabletz et al., 2000; Brabletz et al., 2001; Blaker et al., 2003]. Abraham et al. [2001] have demonstrated a similar scenario in gastric cancer. Activating

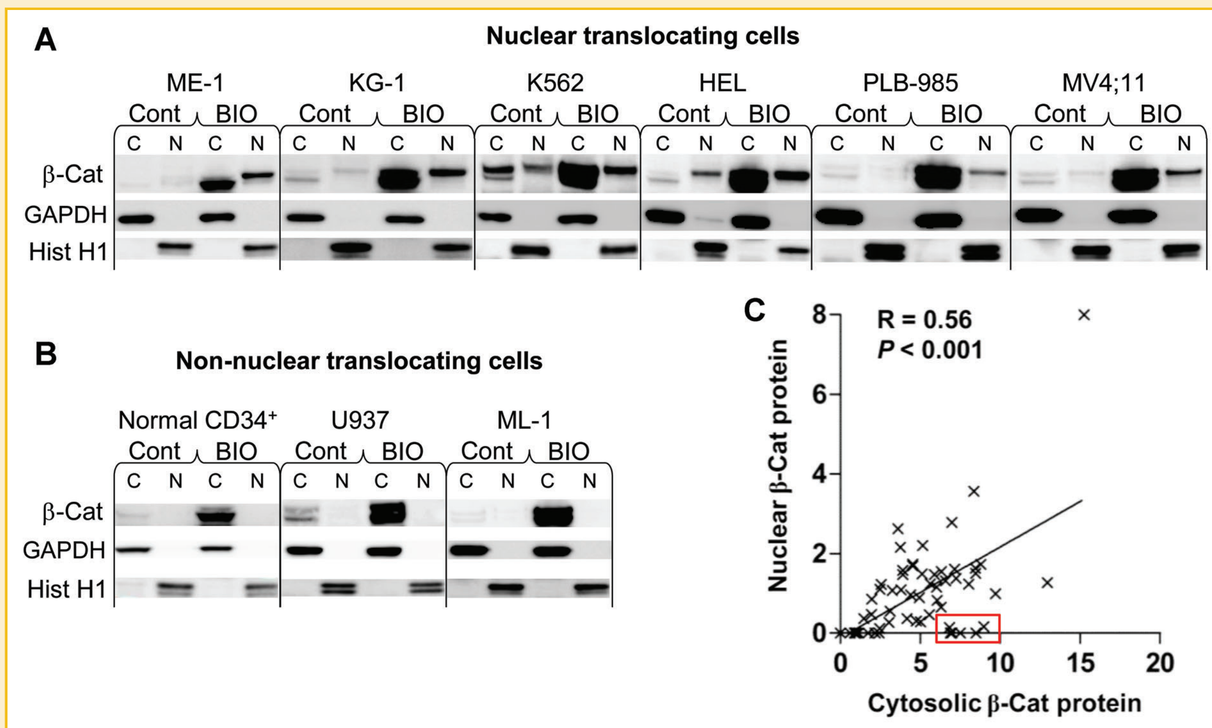

Fig. 1. The selective nuclear translocation of β-catenin in normal and leukemic hematopoietic cells. The localization of β-catenin is shown in leukemic cell lines and normal $CD34^+$ HSPC in response to the Wnt agonist, 6-bromoindirubin-3′-oxime (BIO) highlighting, (A) nuclear translocating and (B) non-nuclear translocating hematopoietic cells. C: The levels of cytosolic and nuclear β-catenin are summarized for primary AML patient blasts showing that most patients appear to freely translocate β-catenin, while a subset (red box) appear to retain the phenotype of normal cells (cytosolic β-catenin but little or no detectable nuclear β-catenin). Adapted from Morgan et al. [2012].

β-catenin mutations were found in 91% of fundic gland polyps, yet none of these samples demonstrated nuclear β-catenin accumulation despite diffuse cytoplasmic and membranous staining. Finally, in a mouse model of liver cancer, Anna et al. [2000] demonstrated that despite the presence of β-catenin mutations in all anthraquinone or oxazepam-induced liver tumors, nuclear β-catenin was only present in the advanced hepatoblastomas, and absent from normal liver, hepatocellular adenomas and carcinomas from the same sections. Taken together, the above studies suggest strict mechanisms exist that control nuclear β-catenin localization regardless of mutational status and cytosolic β-catenin level.

MECHANISMS REGULATING NUCLEAR LOCALIZATION OF β-CATENIN

Proteins less than 30 kDa are readily diffusible through the nuclear pores, however proteins larger than 50 kDa (like β-catenin; 92 kDa) require active transport via the nuclear pore complex (NPC) machinery [Gasiorowski and Dean, 2003]. Given that the β-catenin polypeptide lacks any recognizable nuclear localization or nuclear export signal (NLS/NES) sequences [Fagotto et al., 1998] mechanisms must exist to assist β-catenin in these processes. Below we describe the main mechanisms (summarized in Table I and Fig. 2) implicated to date and consider their relevance to carcinogenesis.

DIRECT CLASSICAL IMPORT

For NLS-containing proteins nuclear entry will invariably be facilitated through the classical import pathway. This pathway is efficient and capable of nuclear transport rates of between 100–1,000 imports per nuclear pore complex (NPC) per minute [Ribbeck and Gorlich, 2001]. NLS-containing proteins form an import complex in the cytoplasm with the NLS receptor which comprises two proteins: importin-α and importin-β. The importin-α:β heterodimer subsequently docks the complex and facilitates movement through the NPC by binding nucleoporins. On the nuclear side, RanGTP binds importin-β causing dissociation from importin-α, and release of the protein cargo, in an energy-dependent process. Both importins are consequently recycled to the cytosol to initiate further such interactions [Stewart, 2007]. Despite the lack of any NLS on β-catenin elements of this import machinery may still be used by β-catenin. Studies by Fagotto et al. [1998] in rat liver cells showed that β-catenin could enter the nucleus through direct interaction with the nuclear envelope in an energy-dependent process. Similarly, a study utilizing a cell-free system produced demonstrated β-catenin entering the nucleus via a Ran-independent mechanism [Yokoya et al., 1999]. Both of these studies concluded that the central armadillo domains of β-catenin are sufficiently homologous to the tandem repeating motifs (called HEAT motifs) of the importin-β central domain to permit direct interaction with the nuclear pore proteins. This hypothesis is supported by a study in *Xenopus* embryos which showed that the β-catenin armadillo domain alone was sufficient for nuclear localization [Funayama et al., 1995]. Further studies identified the precise armadillo repeat regions (R) that most efficiently facilitate β-catenin's nuclear import (R10-12 mainly, and R3-8) through direct interaction with specific nucleoporins [Sharma et al., 2012]. However, these findings have been contradicted by Suh and Gumbiner [2003] who used soluble β-catenin present in the cytosol of *Xenopus* eggs to demonstrate it cannot form direct interactions with nucleoporins. Furthermore this process is constitutive and so inconsistent with the regulated entry of β-catenin discussed above. It could be that direct membrane interaction is still subject to regulatory controls since Fagotto et al., acknowledged inhibitory activities for β-catenin import in the cytosol, and the inner nuclear membrane protein Emerin has been shown to limit flux of β-catenin into the nucleus [Markiewicz et al., 2006]. Alternatively, constitutively imported β-catenin may be rapidly exported (see "Nuclear export" below). Either way, the potential direct interaction of β-catenin with the NPC warrants further investigation since multiple components of the classical import pathway are dysfunctional in human cancer [Kau et al., 2004].

CYTOSOL-NUCLEAR CHAPERONES

The absence of an NLS motif on β-catenin means its transit through the NPC may require the assistance of an NLS-containing chaperone protein. A wide range of cytosol-nucleus shuttle proteins from multiple contexts have been proposed for β-catenin including Smad 3/4 (MSCs; [Jian et al., 2006], chondrocytes; [Li et al., 2006; Zhang et al., 2010], FoxM1 (glioma cell lines; [Zhang et al., 2011]), IRS-1 (mouse embryonic fibroblasts; [Chen et al., 2005]), MUC-1 (pancreatic tumor cell lines; [Wen et al., 2003], mammary gland; [Li et al., 2011]), BCL-9 (various; [Brembeck et al., 2004]; HEK293T cells; [Krieghoff et al., 2006]) and the androgen receptor (prostate cancer cell lines; [Mulholland et al., 2002; Pawlowski et al., 2002]). Some of these attract particular interest given their relevance to malignancy. FoxM1 is a NLS-containing transcription factor that shuttles between the cytoplasm and nucleus [Ma et al., 2005] and is critical for normal development [Ye et al., 1997]. Its overexpression has been reported in many human tumors [Pilarsky et al., 2004] and in a recent report, FoxM1 was found to directly associate with β-catenin and facilitate its nuclear translocation and transcriptional activity in glioma cells [Zhang et al., 2011]. Interestingly, the incidence of APC loss or sporadic stabilizing *CTNNB1* mutations is low in glioma [Paraf et al., 1997] and furthermore, FoxM1 appeared to translocate β-catenin irrespective of total β-catenin level [Zhang et al., 2011]. Given, the frequency of FoxM1 dysregulation in human cancer it would be interesting to examine the interaction of FoxM1 with β-catenin in other tissues and how this differs between normal development and cancer. MUC-1 is a large transmembrane glycoprotein typically expressed on the surface of secretory epithelial cells and has been implicated in most types of human carcinoma [Kufe, 2009]. The polypeptide is actually comprised of two subunits which remain together following post-translational processing and membrane transport, but are subsequently cleaved. The function of the extracellular domain has been well defined, however the role of the short cytoplasmic tail of MUC-1 (MUC-1 CT) is poorly understood. Most studies indicate a signaling function and MUC-1 CT expression has been shown to correlate with β-catenin expression in cancers [Baldus et al., 2004; Udhayakumar et al., 2007]. Further studies have demonstrated that MUC-1 CT associates with β-catenin, assists its transit into the nucleus, and

TABLE I. Summary of the Mechanisms Regulating Nuclear Localization of β-Catenin

Name of factor(s)	System studied	Mode of action	Relevance to cancer	Reference
Direct interaction of β-catenin with nuclear membrane components				
Nucleoporins	Rat liver cells, *Xenopus* embryo, murine cells	β-Catenin interacts with nucleoporins via central armadillo repeats (R10-12 and R3-8) to enter/exit the nucleus. β-Catenin can also freely shuttle out of the nucleus in a CRM1 independent manner	β-Catenin expression and localization is aberrant in many cancer types including colon cancer, breast cancer, ovarian cancer and leukemia	Fagotto et al. [1998], Funayama et al. [1995], Kau et al. [2004], Mikesch et al. [2007], Sharma et al. [2012], Simon et al. [2005], Wiechens and Fagotto [2001], Yokoya et al. [1999] Contradictions:Suh and Gumbiner [2003]
Emerin	Human fibroblast cells	Limits the flux of β-catenin into the nucleus	Unknown	Markiewicz et al. [2006]
CRM1 dependent import and nuclear retention				
Smad 3/4	Adult human MSC and chondrocytes	Binds to β-catenin and shuttles it into the nucleus	Smad 3/4 have been implicated in many types of cancer including pancreatic, colorectal and breast	Jian et al. [2006], Li et al. [2006], Zhang et al. [2010]
FoxM1	Human glioma cell lines	Binds β-catenin and shuttles it into the nucleus	FoxM1 overexpression is observed in many solid tumors including gliomas and gastric cancer. It is reported to directly bind β-catenin facilitating its nuclear translocation in glioma cells	Pilarsky et al. [2004], Zhang et al. [2011]
IRS-1	Mouse embryonic fibroblasts	IGF-1 phosphorylated IRS-1 shuttles β-catenin into the nucleus	Overexpression of IRS-1 has been observed in a broad range of solid tumors and levels of IRS-1 have been linked with β-catenin signalling in ovarian cancer	Chen et al. [2005]
MUC-1	Pancreatic tumor cell lines, mammary gland	Binds to β-catenin and promotes nuclear translocation	MUC-1 has been implicated in many human cancers and MUC-1 CT expression correlates with β-catenin expression in colorectal and gastric cancers	Baldus et al. [2004], Kufe [2009], Li et al. [2011], Udhayakumar et al. [2007], Wen et al. [2003]
BCL-9	Various cells	Can bind in complex with Pygo to promote β-catenin nuclear localization and retention	BCL-9 overexpression is observed in solid tumors. Rearrangements involving BCL-9 are associated with hematological malignancies	Townsley et al. [2004]
Androgen receptor	Prostate cancer cell lines	Promotes β-catenin nuclear entry	Increased androgen receptor signalling is a linked to human cancer, e.g., breast cancer	Mulholland et al. [2002], Pawlowski et al. [2002]
LEF1	MDCK cells, Neuro2A cells (mouse), *Xenopus* embryos, SW480 cells (colon cancer), NIH3T3 cells	LEF1 binds to β-catenin via its N-terminus and promotes nuclear β-catenin localization and retention.		
		Contradictions:mutated β-catenin which is unable to bind to LEF1 can still enter the nucleus	LEF1 is dysregulated in colon cancer and leukemia	Behrens et al. [1996], Fu et al. [2014], Henderson et al. [2002], Huber et al. [1996], Jamieson et al. [2011], Orsulic and Peifer [1996], Petropoulos et al. [2008], Prieve and Waterman [1999], Simcha et al. [1998], Tandon et al. [2011]

Table 1. (*Continued*)

Name of factor(s)	System studied	Mode of action	Relevance to cancer	Reference
TCF	*Xenopus*, HEK293T cells	XTCF-3 (*Xenopus* TCF1 homologue) binds directly to β-catenin and promotes its nuclear translocation. TCF4 overexpression shifts β-catenin localization to the nucleus. In addition, TCF4 is proposed to act by retaining β-catenin in the nucleus	TCF4 has a role in retention of β-catenin in leukemia patients exhibiting abnormal tyrosine kinase activity, e.g., BCR-ABL. TCF4 mediated β-catenin nuclear retention may also contribute to oncogenesis in colorectal cancer	Coluccia et al. [2007], Krieghoff et al. [2006], Molenaar et al. [1996], Morin et al. [1997], Korinek et al. [1997], van de Wetering et al. [2002]
CRM1 dependent export and cytoplasmic/membrane retention				
APC	NIH3T3 cells, HCT116 cells, HEK293T cells, SW480 cells, MCF-7 cells, hematopoietic cells	Direct binding to β-catenin and shuttling of β-catenin into the cytoplasm	Loss of APC activity can influence the progression of colorectal cancer. Reduced APC gene expression as a result of epigenetic mechanisms is observed in breast cancer	Coluccia et al. [2007], Henderson [2000], Krieghoff et al. [2006], Neufeld et al. [2000a,b], Seo and Jho [2007]
Axin	*Xenopus* oocytes, HEK293 cells, COS7 *Drosophila* S2 cells	Axin has the capacity to shuttle β-catenin out the nucleus	Mutations in Axin have been identified in colorectal cancer	Cong and Varmus [2004], Krieghoff et al. [2006], Wiechens et al. [2004]
α-Catenin	HCT116 cells	α-Catenin contains two NES sites and has LMB (Leptomycin B) sensitive β-catenin export activity. Adherens junction components such as α-catenin and cadherins also act as retention factors by localizing β-catenin to the cell membrane	In tumors phosphorylation of β-catenin Y residues disrupt its interaction with α-catenin and cadherins, thereby releasing β-catenin into the cytoplasm. A loss or reduction in α-catenin expression has been implicated in solid tumor progression and metastasis. The loss of α-catenin may also play a role in myeloid leukemias	Giannini et al. [2004], Harris and Peifer [2005], Nelson and Nusse [2004]
CRM1 independent export				
Ran BP-3	*Xenopus* embryo, HeLa nuclear extract	Shuttles active β-catenin from the nucleus to the cytoplasm in an APC and CRM1 independent manner	Unknown	Hendriksen et al. [2005]

CRM1, chromosome maintenance region 1, Smad3/4, mothers against decapentaplegic-related homolog 3/4; FoxM1, forkhead box M1; IRS-1, insulin receptor substrate 1; IGF-1, insulin-like growth factor 1; MUC-1, mucin 1, cell surface associated; BCL-9, B-cell CLL/lymphoma 9; Pygo, pygopus; LEF-1, lymphoid enhancer-binding factor 1; TCF, T-cell factor; APC, adenomatous polyposis coli; BCR-ABL, breakpoint cluster region-C-abl oncogene 1, non-receptor tyrosine kinase (fusion protein); RanBP-3, Ran-binding protein 3.

increases the expression of Wnt target genes [Huang et al., 2003; Wen et al., 2003; Li et al., 2011]. MUC-1 would represent an attractive candidate for further study as a broad regulator of nuclear β-catenin localization given its expression in multiple tissue types [Kufe, 2009]. Perhaps the most interesting of all the proposed cytosol-nuclear chaperones are the TCF/LEF transcription factors which are downstream effectors of the Wnt pathway itself. This raises the intriguing possibility of such molecules providing a "piggy back" for catenins *en route* to transducing a Wnt signal. Behrens et al. [1996] was the first to show that LEF-1 could form a complex with β-catenin through its N-terminus, and that exogenous LEF-1 expression caused the nuclear translocation of β-catenin in both Neuro2A and MDCK cells. Huber and colleagues further confirmed the association of LEF-1 and β-catenin in Neuro2A and SW480 cells, and demonstrated nuclear translocation of β-catenin upon exogenous LEF-1 expression in *Xenopus* embryos [Huber et al., 1996]. Simcha et al. [1998] have since also shown exogenous LEF-1 to translocate endogenous β-catenin to the nucleus of MDCK cells, although others have highlighted that LEF-1 may not be essential for β-catenin import, since mutated β-catenin (unable to bind LEF-1) still enters the nucleus [Orsulic and Peifer, 1996; Prieve and Waterman, 1999]. A few reports have also implicated other TCF family members in regulating β-catenin distribution. XTCF-3, a *Xenopus* TCF-1 homolog, could directly interact with β-catenin in a yeast model, and microinjection of XTCF-3 into Xenopus embryos resulted in re-distribution of β-catenin from the membrane/cytosol to the nucleus [Molenaar et al., 1996]. Finally, a study in HEK293T cells demonstrated the ability of exogenous TCF-4 (also known as TCF7L2) to shift cytoplasmic β-catenin to the nucleus [Krieghoff et al., 2006], although the authors suggest this may arise through increased nuclear retention (discussed further in the section below) rather than cytosol-nuclear shuttling.

NUCLEAR EXPORT

In addition to nuclear import mechanisms, nuclear export systems may be just as pivotal in controlling the nuclear localization of β-catenin. Nuclear export of a protein typically occurs through chromosome maintenance region 1 (CRM1; also referred to as exportin1 or Xpo1) which binds proteins bearing a leucine-rich NES and transports them through the nuclear pore in an energy-dependent process [Hutten and Kehlenbach, 2007]. The small GTPase Ran, in its GTP-bound form, is responsible for loading transport cargo onto the CRM1 receptor, and remains with the complex throughout its cytosolic transit before being recycled back into the nucleus. β-Catenin lacks a canonical NES but nuclear export could instead be facilitated through a NES-containing chaperone. Interestingly, some of the best characterized nuclear-cytosol chaperones are negative regulators of Wnt signaling. Both Neufeld et al. [2000a] and Henderson [2000] identified two active NES on the APC amino terminus which were found to be essential for its CRM1-dependent nuclear export. Henderson further noted the co-localization of APC with β-catenin in colon cancer cell lines and showed that site-directed mutagenesis of the NES, or leptomycin B (LMB; CRM1 inhibitor) treatment, abrogated its shuttling capacity leading to nuclear β-catenin accumulation in NIH3T3 and HCT116 cells. Further, re-introduction of wild type APC into SW480 cells (which harbor a truncated APC mutant) returned β-catenin to the cytoplasm for degradation. Neufeld et al. [2000b] demonstrated very similar findings and confirmed the physical association of APC with β-catenin in the nuclei of HCT116 and MCF-7 cells. Axin also shuttles between the nucleus and cytoplasm of *Xenopus* oocytes and HEK293 cells in a CRM1-dependent manner although the corresponding effects on β-catenin localization in this study were inconsistent [Wiechens et al., 2004]. Cong and Varmus [2004] further confirmed Axin as a CRM1-dependent nuclear-cytoplasmic shuttling protein in 293, COS7 and *Drosophila* S2 cells, and characterized the NLS/NES necessary for this activity. They showed that transfection of Axin into 293 cells shifted β-catenin from nucleus to cytoplasm, and that this process could be inhibited through mutation of the NLS in Axin. In this context α-catenin may also be of interest since it contains two NES and has LMB-sensitive β-catenin export activity in HCT116 cells [Giannini et al., 2004]. Mutation of these sites leads to loss of β-catenin binding and a reduced capacity to repress β-catenin/TCF-dependent transcription. Other CRM1-dependent nuclear β-catenin export chaperones have been described including chibby [Li et al., 2008, 2010], menin [Cao et al., 2009], LZTS2 [Thyssen et al., 2006], PAK4 [Li et al., 2012b] and Kank [Wang et al., 2006]. However, it seems unlikely that CRM1/Ran-GTP mediated export could alone explain the mislocalization of β-catenin in cancer since most studies report over activity of this system with the general approach being to inhibit the complex [Nguyen et al., 2012].

CRM1-independent mechanisms of nuclear β-catenin export have also been described. In vitro experiments from the Henderson lab using digitonin-semi-permeabilized SW480 cells showed β-catenin nuclear export continued despite the presence of $APC^{mut/mut}$, LMB treatment and the absence of exogenous cytosolic factors [Eleftheriou et al., 2001]. The Fagotto group has also demonstrated β-catenin can freely shuttle between nucleus and cytoplasm in a CRM1 and Ran-GTP independent manner [Wiechens and Fagotto, 2001]. The authors propose β-catenin is able to mediate this through non-canonical export sequences which exist in the N and C-terminals of the molecule (C-terminal sequence being most efficient). These sequences share no homology with typical NES or molecules associated with nuclear export, leading the authors to conclude that specific uncharacterized interactions are responsible. They further propose that such a scenario make it likely β-catenin's localization is governed more by retention as discussed further in the next section. Hendriksen et al. [2005] used *Xenopus* embryos and HeLa nuclear extract to identify Ran binding protein 3 (RanBP-3) as a novel nuclear interaction partner of β-catenin. RanBP-3 could antagonize β-catenin/TCF-4 mediated transcription in HEK293T cells by re-localizing the active form from nucleus to cytoplasm in a CRM1 and APC-independent mechanism. Finally, as already mentioned above (see "Direct classical import") CRM1-independent nuclear export has been demonstrated by direct interaction of β-catenin itself with the nuclear membrane. Sharma et al. [2012] demonstrated that the armadillo repeat sequences R10-12 of β-catenin were the most efficient at mediating nuclear export of the molecule through direct interaction with specific transport-active nucleoporins. Together these data suggest that β-catenin can utilize more than one mechanism of nuclear exit depending on the context.

NUCLEAR RETENTION

The finding that β-catenin can interact directly with the nuclear membrane and direct its own nuclear import/export raises the possibility that β-catenin localization is governed by the relative abundance of compartmental β-catenin binding partners. A number of *Drosophila* studies have identified the nuclear protein Pygopus (Pygo) as an essential component for Wnt signaling [Belenkaya et al., 2002; Kramps et al., 2002 ; Thompson, 2004]. Kramps et al. showed that nuclear Pygo could bind Armadillo (homolog of human β-catenin) by using legless (Lgs; homolog of human BCL-9) as an adaptor, and that this trimolecular complex could simultaneously bind dTCF/Pangolin (homolog of human TCF4) to activate Wnt target gene transcription. A further study from the Bienz group confirmed Pygo was constitutively localized to the nucleus of *Drosophila* embryos and was required to capture and maintain Armadillo levels during wingless signaling through Lgs binding [Townsley et al., 2004]. In the same study co-transfection of Pygo and BCL-9 into SW480 cells increased the levels of endogenous nuclear β-catenin and TCF-mediated transcription, showing this mechanism is also relevant for human cells. Interestingly, co-transfection of Pygo and BCL-9 into 293T or HCT-116 failed to increase nuclear β-catenin or TCF/LEF transcription suggesting that other context specific mechanisms are required to nuclear localize β-catenin before Pygo/BCL-9 can affect their retention properties. The observation that β-catenin immunoprecipitates in a trimolecular nuclear complex with Pygo and BCL-9, but that a quadruple complex of β-catenin/Pygo/BCL-9/TCF4 is barely detectable [Thompson, 2004] suggests although Pygo/BCL-9 may perform the initial nuclear

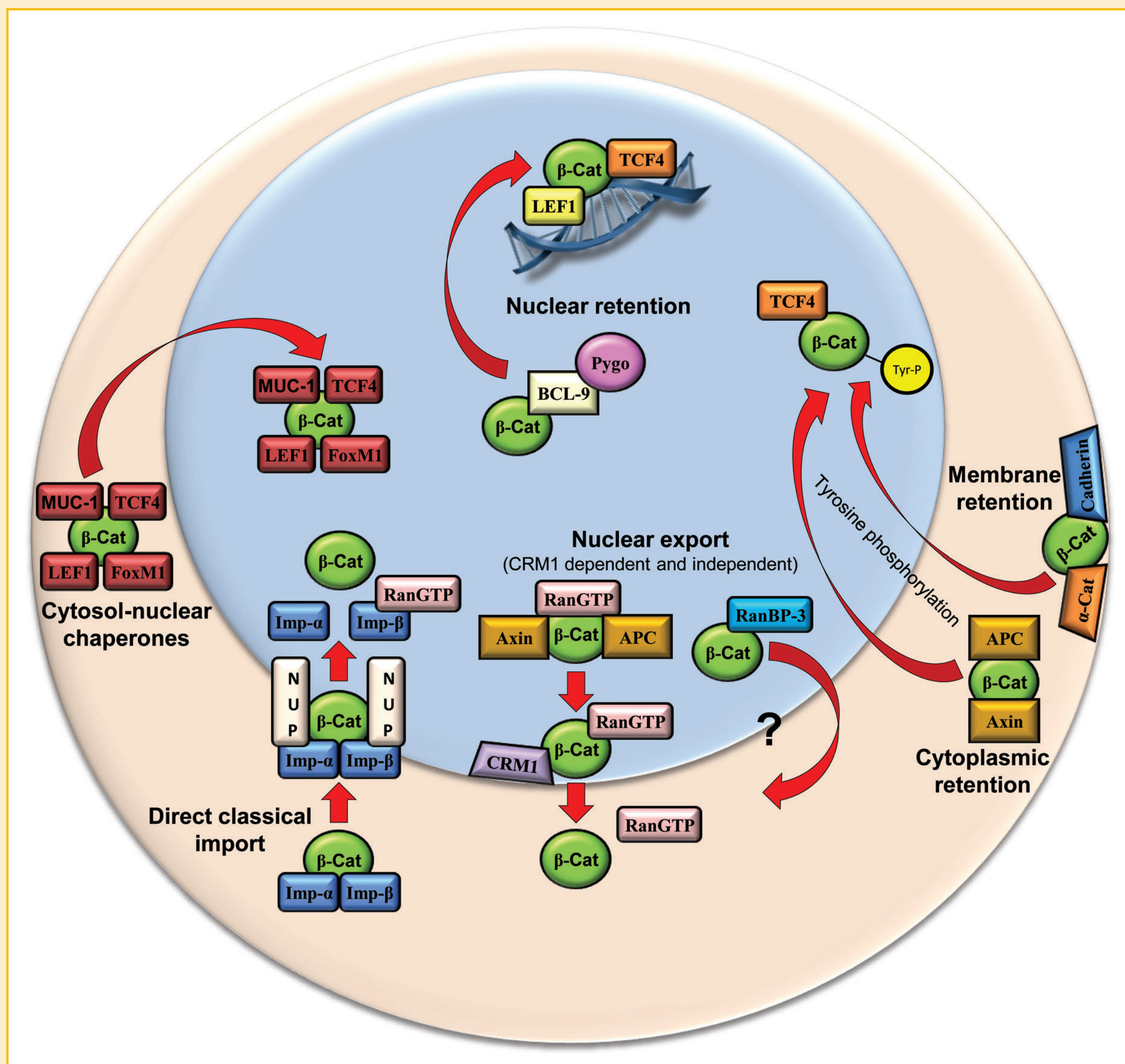

Fig. 2. Summary of the cellular mechanisms proposed to regulate nuclear localization of β-catenin. Schematic summarizing the main mechanisms documented to control β-catenin nuclear localization (for further details see text). NUP, Nucleoporin; Imp-α, Importin-α; Imp-β, Importin-β; Pygo, Pygopus; α-Cat, α-Catenin; β-Cat, β-Catenin; Tyr-P, Tyrosine phosphorylation.

β-catenin capture, it may be TCF4 which is the ultimate recipient and chief retainer. TCF4 was first shown to serve as a nuclear anchor for β-catenin in *Drosophila* where nuclear retention of Armadillo in Axin mutant embryos was dependent on the presence of dTCF/Pangolin [Tolwinski and Wieschaus, 2001]. Work in human cells suggests that whilst APC and Axin retain β-catenin in the cytoplasm (discussed further in section below), it is TCF4 which governs nuclear levels through retention [Krieghoff et al., 2006]. After co-transfection of both TCF4 and β-catenin into HEK293T cells, TCF4 shifted β-catenin to the nucleus, slowed its rate of nucleo-cytoplasmic shuttling, and limited the mobility of β-catenin within the nucleus, indicative of a retention role. The nuclear retention of β-catenin by TCF4 probably contributes in part to the oncogenic potential of this complex that is so fundamental to colorectal carcinogenesis [Korinek et al., 1997; Morin et al., 1997; van de Wetering et al., 2002]. However, whether TCF4 has a nuclear retentive effect on β-catenin in other cancer types, such as those of hematopoietic origin, is unclear and warrants further investigation. The Henderson group has also identified LEF1 as a nuclear retention factor for β-catenin through both overexpression in SW480 [Henderson et al., 2002] and silencing studies in NIH3T3 cells [Jamieson et al., 2011]. LEF1 represents an attractive candidate for retaining nuclear β-catenin in leukemia since its deregulation is frequently reported in hematological malignancy [Petropoulos et al., 2008; Tandon et al., 2011; Fu et al., 2014] and its expression (LEF1 mRNA and protein) is elevated in AML blasts whilst undetectable in normal $CD34^{+}$ HSPC [Simon et al., 2005]. More recently, β-catenin has been identified in a nuclear complex with YAP1 and TBX5 in multiple cancer cell lines and the activity of this complex was found to be essential for the transformation and survival of β-catenin-driven cancers [Rosenbluh et al., 2012]. However the ability of this complex to retain β-catenin in the nucleus and its relevance to normal stem cells was not examined in detail.

CYTOPLASMIC/MEMBRANE RETENTION

Just as nuclear retention factors can positively influence nuclear β-catenin level, cytoplasmic or membranous binding partners may be just as critical in negatively regulating the nuclear localization of β-catenin through sequestration and retention outside of the nucleus. Junctional components, such as cadherin and α-catenin, have frequently been shown (in an epithelial context) to negatively regulate nuclear β-catenin level by sequestering it to adhesion complexes at the cell membrane [Nelson and Nusse, 2004]. Such interactions are typically disturbed through tyrosine (Y) phosphorylation of β-catenin. Multiple epithelial studies have demonstrated Y-phosphorylation causes β-catenin release from structural membrane complexes and this is exploited by tumor cells to both weaken tissue integrity and increase the signaling capacity of β-catenin [Harris and Peifer, 2005]. Clearly, such a mechanism would not be relevant to all tissues since not all cells express junctional membrane components (e.g., hematopoietic cells) but cytosolic retaining factors may instead be more important. Krieghoff et al. [2006] have already shown that exogenous expression of APC or Axin can both enrich β-catenin in the cytoplasm and further reduce its cytosolic mobility in HEK293T cells. Seo and Jho [2007] have additionally demonstrated that a phosphorylated form of APC preferentially retains β-catenin in the cytoplasm. Such a scenario has been formally demonstrated in hematopoietic cells by Coluccia et al. [2007]. In this study, the oncogenic tyrosine kinase BCR-ABL was found to directly associate with β-catenin in CML cells, prevent its serine/threonine phosphorylation (required for degradation), and instead promote Y-phosphorylation of β-catenin on Y86 and Y654 residues. This led to preferential β-catenin binding with nuclear TCF4 over cytosolic Axin, whilst Y-dephosphorylated β-catenin was preferentially retained in the cytoplasm with Axin. Other tyrosine kinases such as KIT and FLT3 may act in a similar manner in hematopoietic cells [Kajiguchi et al., 2007, 2008]. Therefore, cytoplasmic/membrane retention may be a nuclear β-catenin controlling mechanism applicable to multiple tissues and its dysregulation, particularly by phosphorylation, warrants further investigation given the high frequency of tyrosine kinase abnormalities observed in cancer.

CONCLUSIONS

It is now widely accepted that the inappropriate localization of β-catenin to the nucleus is a key oncogenic process. The basic understanding of Wnt signaling whereby β-catenin translocates to the nucleus simply as a result of cytosolic accumulation may be an over simplification in many contexts. In reality the systems regulating nuclear β-catenin entry are likely to be multi-factorial and highly context-dependent. However, this should not perturb efforts to uncover these mechanisms since they are likely to be of significant therapeutic interest in malignancy.

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REFERENCES

Aberle H, Bauer A, Stappert J, Kispert A, Kemler R. 1997. Beta-catenin is a target for the ubiquitin-proteasome pathway. EMBO J 16:3797–3804.

Abraham SC, Nobukawa B, Giardiello FM, Hamilton SR, Wu TT. 2001. Sporadic fundic gland polyps: Common gastric polyps arising through activating mutations in the beta-catenin gene. Am J Pathol 158:1005–1010.

Anderson CB, Neufeld KL, White RL. 2002. Subcellular distribution of Wnt pathway proteins in normal and neoplastic colon. Proc Natl Acad Sci U S A 99:8683–8688.

Anna CH, Sills RC, Foley JF, Stockton PS, Ton TV, Devereux TR. 2000. Beta-catenin mutations and protein accumulation in all hepatoblastomas examined from B6C3F1 mice treated with anthraquinone or oxazepam. Cancer Res 60:2864–2868.

Baldus SE, Monig SP, Huxel S, Landsberg S, Hanisch FG, Engelmann K, Schneider PM, Thiele J, Holscher AH, Dienes HP. 2004. MUC1 and nuclear beta-catenin are coexpressed at the invasion front of colorectal carcinomas and are both correlated with tumor prognosis. Clin Cancer Res 10:2790–2796.

Behrens J, von Kries JP, Kuhl M, Bruhn L, Wedlich D, Grosschedl R, Birchmeier W. 1996. Functional interaction of beta-catenin with the transcription factor LEF-1. Nature 382:638–642.

Belenkaya TY, Han C, Standley HJ, Lin X, Houston DW, Heasman J, Lin X. 2002. Pygopus encodes a nuclear protein essential for wingless/Wnt signaling. Development 129:4089–4101.

Blaker H, Scholten M, Sutter C, Otto HF, Penzel R. 2003. Somatic mutations in familial adenomatous polyps. Nuclear translocation of beta-catenin requires more than biallelic APC inactivation. Am J Clin Pathol 120:418–423.

Brabletz T, Jung A, Hermann K, Gunther K, Hohenberger W, Kirchner T. 1998. Nuclear overexpression of the oncoprotein beta-catenin in colorectal cancer is localized predominantly at the invasion front. Pathol Res Pract 194:701–704.

Brabletz T, Herrmann K, Jung A, Faller G, Kirchner T. 2000. Expression of nuclear beta-catenin and c-myc is correlated with tumor size but not with proliferative activity of colorectal adenomas. Am J Pathol 156:865–870.

Brabletz T, Jung A, Reu S, Porzner M, Hlubek F, Kunz-Schughart LA, Knuechel R, Kirchner T. 2001. Variable beta-catenin expression in colorectal cancers indicates tumor progression driven by the tumor environment. Proc Natl Acad Sci U S A 98:10356–10361.

Brembeck FH, Schwarz-Romond T, Bakkers J, Wilhelm S, Hammerschmidt M, Birchmeier W. 2004. Essential role of BCL9-2 in the switch between beta-catenin's adhesive and transcriptional functions. Genes Dev 18: 2225–2230.

Cao Y, Liu R, Jiang X, Lu J, Jiang J, Zhang C, Li X, Ning G. 2009. Nuclear-cytoplasmic shuttling of menin regulates nuclear translocation of {beta}-catenin. Mol Cell Biol 29:5477–5487.

Chen J, Wu A, Sun H, Drakas R, Garofalo C, Cascio S, Surmacz E, Baserga R. 2005. Functional significance of type 1 insulin-like growth factor-mediated nuclear translocation of the insulin receptor substrate-1 and beta-catenin. J Biol Chem 280:29912–29920.

Clevers H, Nusse R. 2012. Wnt/beta-catenin signaling and disease. Cell 149:1192–1205.

Coluccia AM, Vacca A, Dunach M, Mologni L, Redaelli S, Bustos VH, Benati D, Pinna LA, Gambacorti-Passerini C. 2007. Bcr-Abl stabilizes beta-catenin in chronic myeloid leukemia through its tyrosine phosphorylation. EMBO J 26:1456–1466.

Cong F, Varmus H. 2004. Nuclear-cytoplasmic shuttling of Axin regulates subcellular localization of beta-catenin. Proc Natl Acad Sci U S A 101:2882–2887.

Eleftheriou A, Yoshida M, Henderson BR. 2001. Nuclear export of human beta-catenin can occur independent of CRM1 and the adenomatous polyposis coli tumor suppressor. J Biol Chem 276:25883–25888.

Fagotto F, Gluck U, Gumbiner BM. 1998. Nuclear localization signal-independent and importin/karyopherin-independent nuclear import of beta-catenin. Curr Biol 8:181–190.

Fu Y, Zhu H, Wu W, Xu J, Chen T, Xu B, Qian S, Li J, Liu P. 2014. Clinical significance of lymphoid enhancer-binding factor 1 expression in acute myeloid leukemia. Leuk Lymphoma 55:371–377.

Funayama N, Fagotto F, McCrea P, Gumbiner BM. 1995. Embryonic axis induction by the armadillo repeat domain of beta-catenin: evidence for intracellular signaling. J Cell Biol 128:959–968.

Gasiorowski JZ, Dean DA. 2003. Mechanisms of nuclear transport and interventions. Adv Drug Deliv Rev 55:703–716.

Gaspar C, Fodde R. 2004. APC dosage effects in tumorigenesis and stem cell differentiation. Int J Dev Biol 48:377–386.

Giannini A, Mazor M, Orme M, Vivanco M, Waxman J, Kypta R. 2004. Nuclear export of alpha-catenin: Overlap between nuclear export signal sequences and the beta-catenin binding site. Exp Cell Res 295:150–160.

Harris TJ, Peifer M. 2005. Decisions, decisions: Beta-catenin chooses between adhesion and transcription. Trends Cell Biol 15:234–237.

He TC, Sparks AB, Rago C, Hermeking H, Zawel L, da Costa LT, Morin PJ, Vogelstein B, Kinzler KW. 1998. Identification of c-MYC as a target of the APC pathway. Science 281:1509–1512.

Henderson BR. 2000. Nuclear-cytoplasmic shuttling of APC regulates beta-catenin subcellular localization and turnover. Nat Cell Biol 2:653–660.

Henderson BR, Galea M, Schuechner S, Leung L. 2002. Lymphoid enhancer factor-1 blocks adenomatous polyposis coli-mediated nuclear export and degradation of beta-catenin. Regulation by histone deacetylase 1. J Biol Chem 277:24258–24264.

Hendriksen J, Fagotto F, van der Velde H, van Shie M, Noordermeer J, Fornerod M. 2005. RanBP3 enhances nuclear export of active (beta)-catenin independently of CRM1. J Cell Biol 171:785–797.

Huang L, Ren J, Chen D, Li Y, Kharbanda S, Kufe D. 2003. MUC1 cytoplasmic domain coactivates Wnt target gene transcription and confers transformation. Cancer Biol Ther 2:702–706.

Huber O, Korn R, McLaughlin J, Ohsugi M, Herrmann BG, Kemler R. 1996. Nuclear localization of beta-catenin by interaction with transcription factor LEF-1. Mech Dev 59:3–10.

Hutten S, Kehlenbach RH. 2007. CRM1-mediated nuclear export: to the pore and beyond. Trends Cell Biol 17:193–201.

Inomata M, Ochiai A, Akimoto S, Kitano S, Hirohashi S. 1996. Alteration of beta-catenin expression in colonic epithelial cells of familial adenomatous polyposis patients. Cancer Res 56:2213–2217.

Jamieson C, Sharma M, Henderson BR. 2011. Regulation of beta-catenin nuclear dynamics by GSK-3beta involves a LEF-1 positive feedback loop. Traffic 12:983–999.

Jian H, Shen X, Liu I, Semenov M, He X, Wang XF. 2006. Smad3-dependent nuclear translocation of beta-catenin is required for TGF-beta1-induced proliferation of bone marrow-derived adult human mesenchymal stem cells. Genes Dev 20:666–674.

Kajiguchi T, Chung EJ, Lee S, Stine A, Kiyoi H, Naoe T, Levis MJ, Neckers L, Trepel JB. 2007. FLT3 regulates beta-catenin tyrosine phosphorylation, nuclear localization, and transcriptional activity in acute myeloid leukemia cells. Leukemia 21:2476–2484.

Kajiguchi T, Lee S, Lee MJ, Trepel JB, Neckers L. 2008. KIT regulates tyrosine phosphorylation and nuclear localization of beta-catenin in mast cell leukemia. Leuk Res 32:761–770.

Kau TR, Way JC, Silver PA. 2004. Nuclear transport and cancer: From mechanism to intervention. Nat Rev Cancer 4:106–117.

Kobayashi M, Honma T, Matsuda Y, Suzuki Y, Narisawa R, Ajioka Y, Asakura H. 2000. Nuclear translocation of beta-catenin in colorectal cancer. Br J Cancer 82:1689–1693.

Korinek V, Barker N, Morin PJ, van Wichen D, de Weger R, Kinzler KW, Vogelstein B, Clevers H. 1997. Constitutive transcriptional activation by a beta-catenin-Tcf complex in APC−/− colon carcinoma. Science 275:1784–1787.

Kramps T, Peter O, Brunner E, Nellen D, Froesch B, Chatterjee S, Murone M, Zullig S, Basler K. 2002. Wnt/wingless signaling requires BCL9/legless-mediated recruitment of pygopus to the nuclear beta-catenin-TCF complex. Cell 109:47–60.

Krieghoff E, Behrens J, Mayr B. 2006. Nucleo-cytoplasmic distribution of beta-catenin is regulated by retention. J Cell Sci 119:1453–1463.

Kufe DW. 2009. Mucins in cancer: Function, prognosis and therapy. Nat Rev Cancer 9:874–885.

Li TF, Chen D, Wu Q, Chen M, Sheu TJ, Schwarz EM, Drissi H, Zuscik M, O'Keefe RJ. 2006. Transforming growth factor-beta stimulates cyclin D1 expression through activation of beta-catenin signaling in chondrocytes. J Biol Chem 281:21296–21304.

Li FQ, Mofunanya A, Harris K, Takemaru K. 2008. Chibby cooperates with 14-3-3 to regulate beta-catenin subcellular distribution and signaling activity. J Cell Biol 181:1141–1154.

Li FQ, Mofunanya A, Fischer V, Hall J, Takemaru K. 2010. Nuclear-cytoplasmic shuttling of Chibby controls beta-catenin signaling. Mol Biol Cell 21:311–322.

Li Y, Yi H, Yao Y, Liao X, Xie Y, Yang J, Yan Z, Wang L, Lu S, Kuang Y, Gu M, Fei J, Wang Z, Huang L. 2011. The cytoplasmic domain of MUC1 induces hyperplasia in the mammary gland and correlates with nuclear accumulation of beta-catenin. PLoS ONE 6:e19102.

Li VS, Ng SS, Boersema PJ, Low TY, Karthaus WR, Gerlach JP, Mohammed S, Heck AJ, Maurice MM, Mahmoudi T, Clevers H. 2012a. Wnt signaling through inhibition of beta-catenin degradation in an intact Axin1 complex. Cell 149:1245–1256.

Li Y, Shao Y, Tong Y, Shen T, Zhang J, Li Y, Gu H, Li F. 2012b. Nucleo-cytoplasmic shuttling of PAK4 modulates beta-catenin intracellular translocation and signaling. Biochim Biophys Acta 1823:465–475.

Luis TC, Naber BA, Roozen PP, Brugman MH, de Haas EF, Ghazvini M, Fibbe WE, van Dongen JJ, Fodde R, Staal FJ. 2011. Canonical wnt signaling regulates hematopoiesis in a dosage-dependent fashion. Cell Stem Cell 9:345–356.

Ma RY, Tong TH, Cheung AM, Tsang AC, Leung WY, Yao KM. 2005. Raf/MEK/MAPK signaling stimulates the nuclear translocation and transactivating activity of FOXM1c. J Cell Sci 118:795–806.

Markiewicz E, Tilgner K, Barker N, van de Wetering M, Clevers H, Dorobek M, Hausmanowa-Petrusewicz I, Ramaekers FC, Broers JL, Blankesteijn WM, Salpingidou G, Wilson RG, Ellis JA, Hutchison CJ. 2006. The inner nuclear membrane protein emerin regulates beta-catenin activity by restricting its accumulation in the nucleus. EMBO J 25:3275–3285.

Mikesch JH, Steffen B, Berdel WE, Serve H, Muller-Tidow C. 2007. The emerging role of Wnt signaling in the pathogenesis of acute myeloid leukemia. Leukemia 21:1638–1647.

Molenaar M, van de Wetering M, Oosterwegel M, Peterson-Maduro J, Godsave S, Korinek V, Roose J, Destree O, Clevers H. 1996. XTcf-3 transcription factor mediates beta-catenin-induced axis formation in Xenopus embryos. Cell 86:391–399.

Morgan RG, Pearn L, Liddiard K, Pumford SL, Burnett AK, Tonks A, Darley RL. 2012. Gamma-Catenin is overexpressed in acute myeloid leukemia and promotes the stabilization and nuclear localization of beta-catenin. Leukemia 27:336–343.

Morin PJ, Sparks AB, Korinek V, Barker N, Clevers H, Vogelstein B, Kinzler KW. 1997. Activation of beta-catenin-Tcf signaling in colon cancer by mutations in beta-catenin or APC. Science 275:1787–1790.

Mulholland DJ, Cheng H, Reid K, Rennie PS, Nelson CC. 2002. The androgen receptor can promote beta-catenin nuclear translocation independently of adenomatous polyposis coli. J Biol Chem 277:17933–17943.

Nelson WJ, Nusse R. 2004. Convergence of Wnt, beta-catenin, and cadherin pathways. Science 303:1483–1487.

Neufeld KL, Nix DA, Bogerd H, Kang Y, Beckerle MC, Cullen BR, White RL. 2000a. Adenomatous polyposis coli protein contains two nuclear export signals and shuttles between the nucleus and cytoplasm. Proc Natl Acad Sci U S A 97:12085–12090.

Neufeld KL, Zhang F, Cullen BR, White RL. 2000b. APC-mediated down-regulation of beta-catenin activity involves nuclear sequestration and nuclear export. EMBO Rep 1:519–523.

Nguyen KT, Holloway MP, Altura RA. 2012. The CRM1 nuclear export protein in normal development and disease. Mol Int J Biochem Mol Biol 3:137–151.

Orsulic S, Peifer M. 1996. An in vivo structure-function study of armadillo, the beta-catenin homologue, reveals both separate and overlapping regions of the protein required for cell adhesion and for wingless signaling. J Cell Biol 134:1283–1300.

Paraf F, Jothy S, Van Meir EG. 1997. Brain tumor-polyposis syndrome: two genetic diseases?. J Clin Oncol 15:2744–2758.

Pawlowski JE, Ertel JR, Allen MP, Xu M, Butler C, Wilson EM, Wierman ME. 2002. Liganded androgen receptor interaction with beta-catenin: nuclear co-localization and modulation of transcriptional activity in neuronal cells. J Biol Chem 277:20702–20710.

Petropoulos K, Arseni N, Schessl C, Stadler CR, Rawat VP, Deshpande AJ, Heilmeier B, Hiddemann W, Quintanilla-Martinez L, Bohlander SK, Feuring-Buske M, Buske C. 2008. A novel role for Lef-1, a central transcription mediator of Wnt signaling, in leukemogenesis. J Exp Med 205:515–522.

Phelps RA, Chidester S, Dehghanizadeh S, Phelps J, Sandoval IT, Rai K, Broadbent T, Sarkar S, Burt RW, Jones DA. 2009. A two-step model for colon adenoma initiation and progression caused by APC loss. Cell 137:623–634.

Pilarsky C, Wenzig M, Specht T, Saeger HD, Grutzmann R. 2004. Identification and validation of commonly overexpressed genes in solid tumors by comparison of microarray data. Neoplasia 6:744–750.

Prieve MG, Waterman ML. 1999. Nuclear localization and formation of beta-catenin-lymphoid enhancer factor 1 complexes are not sufficient for activation of gene expression. Mol Cell Biol 19:4503–4515.

Ribbeck K, Gorlich D. 2001. Kinetic analysis of translocation through nuclear pore complexes. EMBO J 20:1320–1330.

Rosenbluh J, Nijhawan D, Cox AG, Li X, Neal JT, Schafer EJ, Zack TI, Wang X, Tsherniak A, Schinzel AC, Shao DD, Schumacher SE, Weir BA, Vazquez F, Cowley GS, Root DE, Mesirov JP, Beroukhim R, Kuo CJ, Goessling W, Hahn WC. 2012. Beta-Catenin-driven cancers require a YAP1 transcriptional complex for survival and tumorigenesis. Cell 151:1457–1473.

Seo E, Jho EH. 2007. Axin-independent phosphorylation of APC controls beta-catenin signaling via cytoplasmic retention of beta-catenin. Biochem Biophys Res Commun 357:81–86.

Sharma M, Jamieson C, Johnson M, Molloy MP, Henderson BR. 2012. Specific armadillo repeat sequences facilitate beta-catenin nuclear transport in live cells via direct binding to nucleoporins Nup62, Nup153, and RanBP2/Nup358. J Biol Chem 287:819–831.

Shtutman M, Zhurinsky J, Simcha I, Albanese C, D'Amico M, Pestell R, Ben-Ze'ev A. 1999. The cyclin D1 gene is a target of the beta-catenin/LEF-1 pathway. Proc Natl Acad Sci U S A 96:5522–5527.

Silva-Vargas V, Lo CC, Giangreco A, Ofstad T, Prowse DM, Braun KM, Watt FM. 2005. Beta-catenin and Hedgehog signal strength can specify number and location of hair follicles in adult epidermis without recruitment of bulge stem cells. Dev Cell 9:121–131.

Simcha I, Shtutman M, Salomon D, Zhurinsky J, Sadot E, Geiger B, Ben-Ze'ev A. 1998. Differential nuclear translocation and transactivation potential of beta-catenin and plakoglobin. J Cell Biol 141:1433–1448.

Simon M, Grandage VL, Linch DC, Khwaja A. 2005. Constitutive activation of the Wnt/beta-catenin signalling pathway in acute myeloid leukaemia. Oncogene 24:2410–2420.

Stewart M. 2007. Molecular mechanism of the nuclear protein import cycle. Nat Rev Mol Cell Biol 8:195–208.

Suh EK, Gumbiner BM. 2003. Translocation of beta-catenin into the nucleus independent of interactions with FG-rich nucleoporins. Exp Cell Res 290:447–456.

Tandon B, Peterson L, Gao J, Nelson B, Ma S, Rosen S, Chen YH. 2011. Nuclear overexpression of lymphoid-enhancer-binding factor 1 identifies chronic lymphocytic leukemia/small lymphocytic lymphoma in small B-cell lymphomas. Mod Pathol 24:1433–1443.

Thompson BJ. 2004. A complex of Armadillo, Legless, and Pygopus coactivates dTCF to activate wingless target genes. Curr Biol 14:458–466.

Thyssen G, Li TH, Lehmann L, Zhuo M, Sharma M, Sun Z. 2006. LZTS2 is a novel beta-catenin-interacting protein and regulates the nuclear export of beta-catenin. Mol Cell Biol 26:8857–8867.

Tolwinski NS, Wieschaus E. 2001. Armadillo nuclear import is regulated by cytoplasmic anchor Axin and nuclear anchor dTCF/Pan. Development 128:2107–2117.

Townsley FM, Thompson B, Bienz M. 2004. Pygopus residues required for its binding to Legless are critical for transcription and development. J Biol Chem 279:5177–5183.

Udhayakumar G, Jayanthi V, Devaraj N, Devaraj H. 2007. Interaction of MUC1 with beta-catenin modulates the Wnt target gene cyclinD1 in H. pylori-induced gastric cancer. Mol Carcinog 46: 807–817.

van de Wetering M, Sancho E, de Verweij CLW, Oving I, Hurlstone A, van der Horn K, Batlle E, Coudreuse D, Haramis AP, Tjon-Pon-Fong M, Moerer P, van den Born M, Soete G, Pals S, Eilers M, Medema R, Clevers H. 2002. The beta-catenin/TCF-4 complex imposes a crypt progenitor phenotype on colorectal cancer cells. Cell 111:241–250.

Wang Y, Kakinuma N, Zhu Y, Kiyama R. 2006. Nucleo-cytoplasmic shuttling of human Kank protein accompanies intracellular translocation of beta-catenin. J Cell Sci 119:4002–4010.

Wen Y, Caffrey TC, Wheelock MJ, Johnson KR, Hollingsworth MA. 2003. Nuclear association of the cytoplasmic tail of MUC1 and beta-catenin. J Biol Chem 278:38029–38039.

Wiechens N, Fagotto F. 2001. CRM1- and Ran-independent nuclear export of beta-catenin. Curr Biol 11:18–27.

Wiechens N, Heinle K, Englmeier L, Schohl A, Fagotto F. 2004. Nucleo-cytoplasmic shuttling of Axin, a negative regulator of the Wnt-beta-catenin Pathway. J Biol Chem 279:5263–5267.

Ye H, Kelly TF, Samadani U, Lim L, Rubio S, Overdier DG, Roebuck KA, Costa RH. 1997. Hepatocyte nuclear factor 3/fork head homolog 11 is expressed in proliferating epithelial and mesenchymal cells of embryonic and adult tissues. Mol Cell Biol 17:1626–1641.

Yokoya F, Imamoto N, Tachibana T, Yoneda Y. 1999. Beta-catenin can be transported into the nucleus in a Ran-unassisted manner. Mol Biol Cell 10:1119–1131.

Zhang M, Wang M, Tang X, Li TF, Zhang Y, Chen D. 2010. Smad3 prevents beta-catenin degradation and facilitates beta-catenin nuclear translocation in chondrocytes. J Biol Chem 285:8703–8710.

Zhang N, Wei P, Gong A, Chiu WT, Lee HT, Colman H, Huang H, Xue J, Liu M, Wang Y, Sawaya R, Xie K, Yung WK, Medema RH, He X, Huang S. 2011. FoxM1 promotes beta-catenin nuclear localization and controls Wnt target-gene expression and glioma tumorigenesis. Cancer Cell 20:427–442.