Cell. Mol. Life Sci. 66 (2009) 1850 – 1857
1420-682X/09/111850-8
DOI 10.1007/s00018-009-9153-z

Cellular and Molecular Life Sciences

CK2 and its role in Wnt and NF-κB signaling: Linking development and cancer

I. Dominguez, G. E. Sonenshein, and D. C. Seldin*

Section of Hematology-Oncology, Department of Medicine, Boston University School of Medicine and Boston Medical Center, 650 Albany Street, Boston, Massachusetts 02118 (USA), Fax: +1 617 630 7530,
e-mail: Dseldin@bu.edu

Online First 24 April 2009

Abstract. CK2 is a highly conserved tetrameric serine/threonine kinase present in all eukaryotic organisms. It is constitutively active, and appears to be regulated by level of expression and activity, and subcellular localization. In turn, it has been postulated to control the function of many proteins through changes in phosphorylation that affect protein stability, protein-protein interactions, and subcellular localization. Through these mechanisms, CK2 regulates many fundamental cellular properties. An enzyme that carries out such a master regulatory function is likely to be important in organismic development and in cancer. We have shown that overexpression of CK2 catalytic subunits is capable of promoting tumorigenesis, and that loss of CK2 catalytic subunits in development can be lethal. Through studies in cells, mice, and frogs, we and others have identified the Wnt and NF-κB pathways as two key signal transduction pathways that are regulated by CK2 activity, in embryonic development and in cancer. These results suggest that inhibiting CK2 could be useful in treating cancer, but dangerous to developing organisms. (Part of a Multi-author Review)

Keywords. Protein kinase CK2, casein kinase II, Wnt, NF-κB, carcinogenesis, epithelial-to-mesenchymal transition, development.

Linking development and cancer

The fastest-growing cells in the body are cells in the developing embryo and cancer cells in aggressive tumors. Embryonic development is a rigorously orchestrated process, in which stem cells self-renew and continuously generate progeny that follow a highly regulated process of differentiation to coordinately produce the functional organs of the newborn. The process depends upon cell-autonomous activities that are innate to each particular cell lineage, and is also controlled by cell-non-autonomous signals that are provided from neighboring cells or from molecules in the environment. Cancer in contrast is a process of dysregulated growth and differentiation, in which cells grow abnormally, fail to mature properly, and acquire new abilities to divide and invade, and to avoid apoptosis. These events lead to tumor formation and metastasis, and the eventual demise of the host. Although the result of cancer is diametrically opposed to that of development, in which new life is created, cancer cells have seized upon properties of developing cells to transform themselves. Cancer cells often hijack embryonic growth and differentiation pathways. From cancers, we can learn a great deal about normal growth control and about development: for example, programmed cell death or apoptosis is critical in embryonic development, but its mechanism was elucidated through the study of the failure of cancer cells to die. This review will focus on two specific signal transduction pathways, the Wnt pathway and the NF-κB pathway, that regulate key transcriptional events in development, play a role in some cancers, and can be modulated by CK2 activity.

* Corresponding author.

The Wnt Pathway

An example in which the complementary study of cancer and development has borne fruit has been the Wnt pathway. The name itself came from independent lines of research that converged on this one pathway. Harold Varmus and colleagues were examining genes activated by integration of mouse mammary tumor virus into the mouse genome, called ints. Roel Nusse and colleagues were studying the regulation of the development of the Drosophila wing; a mutant with abnormal wing development was termed wingless. When it was appreciated that int-1 and wingless were paralogs of the same secreted growth factor, their names were combined to form Wnt.

We now recognize that the Wnts are actually a large family of secreted cysteine-rich glycoproteins. Wnts act through two major signaling pathways. 'Canonical' Wnt signaling regulates cell proliferation, survival and fate determination, while 'non-canonical' signaling regulates tissue polarity and early morphogenetic movements [1]. The canonical pathway controls the intracellular levels of β-catenin, a protein independently involved in cell adhesion which serves as a co-transcriptional activator in Wnt signaling [1]. In resting cells, cytoplasmic β-catenin is targeted for proteosomal degradation by phosphorylation of its N-terminus by casein kinase 1 (CK1) and glycogen synthase kinase-3β (GSK3β) within a multi-protein 'destruction complex' where adenomatous polyposis coli (APC) and axin act as scaffolds. This destruction complex is inhibited by Wnt through dishevelled (Dvl), leading to stabilization and nuclear translocation of β-catenin (Fig. 1). Binding of β-catenin to members of the T cell-specific transcription factor/lymphoid enhancer-binding factor (TCF/LEF) family of DNA-binding proteins regulates transcription of genes that include *engrailed* and *siamois* in embryonic development, and *cyclin D* and *c-myc* in tumors [2]. Activation of canonical Wnt signaling in different model systems can be detected by regulation of a few well-characterized target genes, the transactivation of artificial TCF reporters, or an increase in nuclear β-catenin protein levels. Wnts are essential for embryonic development, and mutations have been linked to human developmental diseases such as tetra-amelia, a genetic disorder characterized by absence of all four limbs and other anomalies, and metabolic bone disease [3, 4].

CK2 was first linked to the Wnt pathway by the Nusse lab, who reported that CK2 is a Dvl kinase in *Drosophila* [5]. We and others have found this to be the case in mammalian cells as well [6]. One of the best-validated roles of Wnt signaling is in specification of the dorsal axis in developing *Xenopus laevis* embryos. The role of CK2 in regulation of this process will be discussed below. CK2 actually appears to be a multisite regulator of Wnt signaling, as it is capable of phosphorylating the regulated transcriptional co-factor β-catenin and the transcription factor TCF/LEF itself (Fig. 1).

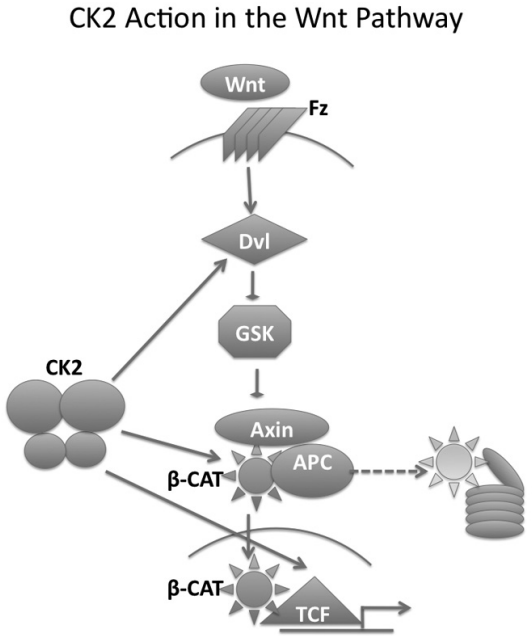

Figure 1. Schematic of Wnt signaling, and the multiple roles of CK2 in it. When Wnt ligands bind Frizzled (Fz) receptors and LRP co-receptors (not shown), a Dishevelled (Dvl) intermediate inhibits GSK3β activity. Active GSK3β phosphorylates β-catenin in its N-terminus, promoting its ubiquitination and proteasome-mediated degradation. Scaffold proteins Axin and APC (adenomatous polyposis coli gene product) form a negative regulatory complex that promotes β-catenin degradation. CK2 acts by phosphorylating and stabilizing Dvl and β-catenin, and promotes TCF DNA binding in the nucleus.

The NF-κB/Rel pathway

Like CK2, which is not a casein kinase *in vivo*, 'NF-κB' is a misnomer for an important family of transcriptional regulators. While it may have been originally identified as a nuclear factor regulating kappa light chain gene expression in B lymphocytes, we now know that NF-κB is not exclusively nuclear, is not restricted to lymphocytes, and that it regulates many downstream genes, not just kappa light chains.

NF-κB is a dimeric transcription factor consisting of p50, p52, p65/relA, relB, and c-rel subunits. NF-κB is maintained in a quiescent state in the cytoplasm by its inhibitory protein termed IκB, which itself consists of three isoforms, α, β, and γ. NF-κB was first found to have oncogenic activity in birds as a mutant truncated form, v-rel, expressed by avian reticuloendotheliosis virus. Other oncogenic viruses including HTLV-1 [7]

and EBV can also activate NF-κB, and we were among the first to appreciate that constitutive NF-κB activation is a common event in cancer [8], due to its anti-apoptotic activity, reviewed in [9] and pro-proliferative functions. Direct demonstration of the transforming potential of NF-κB in mammary tumorigenesis was shown through overexpression of c-rel in the mammary glands of transgenic mice using the MMTV promoter; more than 30% of the female mice developed mammary tumors [10]. Targets of NF-κB in tumorigenesis include c-myc [7], cyclin D1, bcl-xL, and others, and upregulation of these targets was seen in the MMTV-c-rel transgenic mammary tumors [10]. NF-κB has recently been identified as a key mediator of the epithelial-to-mesenchymal transition (EMT), which is felt to be a key process in the acquisition of a metastatic phenotype by tumor cells (recently reviewed in [11]). Snail and slug are transcription factors downstream of NF-κB that regulate the expression of genes such as E-cadherin, and vimentin. In addition, NF-κB directly regulates transcription of several matrix metalloproteinases (MMPs). Loss of the epithelial adhesion molecule E-cadherin and upregulation of vimentin and MMPs promotes motility and invasion. Recently, Sonenshein and colleagues have shown that the NF-κB subunit RelB mediates estrogen suppression of invasiveness in breast cancer cells [12].

Functional activation of NF-κB requires it to be released from its inhibitor, translocate to the nucleus, and there activate transcription. This occurs through the regulated degradation of IκB, which takes place through a multistep process in which phosphorylation signals ubiquitination and recognition by the proteasome. A series of kinases can start this cascade, including IKKα, IKKβ, IKK-i/IKKε, and CK2 itself [13, 14]. N-terminal phosphorylation by IKKα and IKKβ is referred to as canonical activation, while the alternative C-terminal phosphorylation-induced degradation by CK2 [15, 16] is considered to be a non-canonical pathway. A kinase-inactive form of CK2 can act as a dominant negative to reduce IκB phosphorylation and block NF-κB activation [13]. CK2 actually acts at multiple levels in NF-κB activation, as it targets not onlyIκB, but IKK-i/IKKε upstream [17] and NF-κB p65 itself [18,19] (Fig. 2).

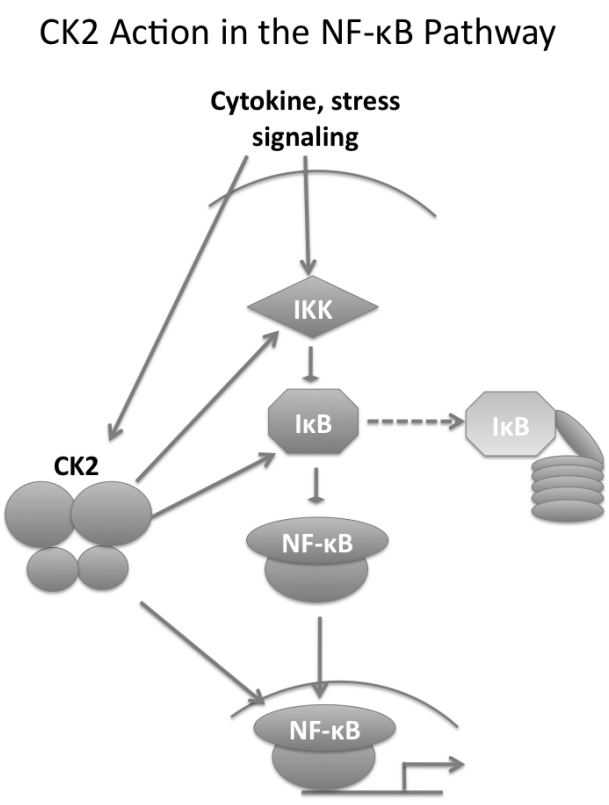

Figure 2. Schematic of NF-κB activation, and the role of CK2 in it. Cytokine and stress signaling promote kinase activation that, depending upon cell type, can include CK2 and/or IKK (IκB kinase). These kinases phosphorylate IκB in its C-terminal PEST domain, and signal its ubiquitination and proteasomal activation. This releases NF-κB which is than able to translocate to the nucleus and activate transcription.

Developmental roles of CK2 and its interaction with the Wnt and NF-κB pathways

CK2 in early embryogenesis

CK2 plays an essential role in early embryogenesis. This is quite apparent from mice with homozygous deletion of the β subunit, which is early embryonic lethal prior to implantation [20]. In this case, CK2 seems to act primarily in a cell-autonomous manner as no homozygous null CK2β embryonic stem (ES) cells could be obtained. This parallels findings in unicellular eukaryotes such as yeast, where temperature-sensitive alleles of CK2 can affect cell growth, morphology and viability [21]. Precisely which aspect of embryonic development is affected in the CK2β-null embryos is unknown; heterozygous CK2β+/– mice had no obvious phenotype but were born at lower rates than expected [22]. CK2β+/– ES cells seemed to have no alteration in growth rate or G1-, G2-checkpoints.

More information about Wnt signaling pathways regulated by CK2 during embryonic development comes from studies in *Xenopus laevis* embryos. In *Xenopus*, the contribution of gene products to embryonic development can be assessed by local injection of mRNA into the early embryo. Injection of an mRNA encoding a dominant-negative form of CK2α into the dorsal region of the embryo inhibits formation of dorsal structures such as the notochord, neural tube and somites [23]. CK2 overexpression in the ventral region of the embryo promotes the ectopic formation

of dorsal structures. Furthermore, endogenous CK2 protein is expressed from maternally derived mRNA in the appropriate dorsal location in the early embryo. Thus, CK2 is required for formation of the dorsal structures. Dorsal axis formation in *Xenopus* (and other organisms) is well-recognized to be dependent upon canonical Wnt signaling, as components of Wnt signaling promote dorsal axis formation and Wnt antagonists block it. Thus, we hypothesized that CK2 is acting via the Wnt pathway to promote dorsal axis formation. We found that CK2 overexpression increased levels of Wnt-responsive genes such as *siamois* and *Xnr3* [23], and dorsal expression of a peptide inhibitor of CK2 led to inhibition (Fig. 3). Research from a number of laboratories has shown that CK2 is involved in Wnt signaling in *Drosophila* [5] and mammalian cells [6,24–27].

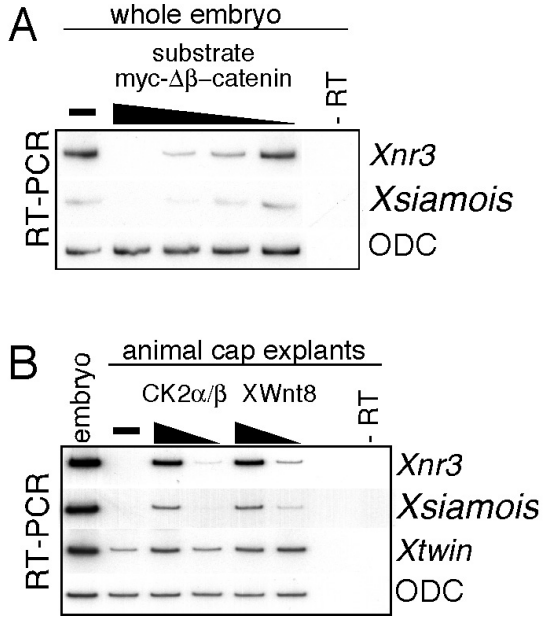

Figure 3. CK2 activity is necessary and sufficient for β-catenin/TCF target gene expression. (*A*) As a non-pharmacologic approach to inhibiting CK2 in *Xenopus laevis* embryos, a myc-tagged β-catenin fragment from residues 380–430, encompassing the CK2 consensus phosphorylation site at T393, was subcloned into pCS2-MT, from which mRNA was synthesized and injected into dorsal blastomeres at stage 2–3. 3 ng of mRNA completely inhibited formation of dorsal structures and Wnt target gene (Xsiamois and Xnr3) expression (lane 2) as assessed by RT-PCR using RNA from stage 10 embryos; ornithine decarboxylase (ODC) is the control. (*B*) Conversely, CK2α and β mRNA, like Xwnt8, induces β-catenin/TCF target genes in animal cap explants. Stage 2 embryos were injected with varying amounts of mRNA coding for CK2α and CK2β (lane 3, 1.1 and 0.9 ng and lane 4, 0.5 and 0.4 ng, respectively), mRNA for Xwnt8 (lane 5, 0.34 pg and lane 6, 0.17 pg) or left uninjected (lane 2). RT-PCR analysis was performed on RNA from animal caps at stage 10. ODC again serves as a normalization control. PCR samples from whole embryos (lane 1) and minus RT (lane 7) were also analyzed. Panel *b* is reprinted from [10] with permission of Elsevier.

CK2 appears to be capable of affecting the Wnt pathway at multiple levels. CK2 can phosphorylate intracellular Wnt signaling intermediates such as Dvl [5,28], β-catenin [28], APC [29] and TCF/LEF [26, 30, 31]. With respect to β-catenin, we have shown that CK2 associates with it, phosphorylates it, and regulates its stability in cells [6, 28]. A key site of phosphorylation appears to be threonine 393 [6]. Phosphorylation of T393 may increase β-catenin stability through diminished interaction with Axin, as a phosphomimetic mutant of β-catenin with an aspartic acid at position 393 has reduced binding to Axin and cannot be regulated by Axin in *Xenopus* embryos [Wu et al., *J. Cellular Biochem.*, in press]. Further downstream, CK2 is capable of regulating the interaction and transactivation activity of a β-catenin/LEF-1 complex though phosphorylation of LEF-1 at S42, S61 [24, 26]. Taken together, these results strongly implicate CK2 in a critical regulatory role in Wnt-dependent processes in embryonic development and in cancer.

Interestingly, in the fruit fly *Drosophila melanogaster*, the NF-κB pathway appears to regulate formation of the dorso-ventral axis, and here again CK2 activity appears to play a role. Dorso-ventral axis formation in *Drosophila* depends upon the nuclear translocation of the transcription factor Dorsal, a fly NF-κB (c-rel) homolog. Nuclear translocation of Dorsal is inhibited by a cytoplasmic interactor, Cactus, which is an IκB homolog (see http://www.sdbonline.org/fly/aignfam/dorslgrp.htm). CK2 regulates Cactus through phosphorylation of its PEST stability domain at serines 463, 467, and 468 [32, 33]. *Drosophila* embryos cannot be injected with mRNAs encoding developmental genes; rather, mutants are created through insertional mutagenesis. However, null alleles of CK2 in *Drosophila* are homozygous lethal [34, 35]. Functional temperature-sensitive alleles of *Drosophila* CK2α have been recently isolated [36] and should provide a genetic answer about the role of CK2 in dorso-ventral axis formation via regulation of Dorsal and in wing development via regulation of Wnt signaling.

CK2 in gametogenesis

CK2 has been implicated in gametogenesis in different model organisms. In mice and *Xenopus laevis*, CK2 transcript levels are higher in germ cells compared to somatic cells [37, 38]. The CK2α′ subunit is well-expressed in testis and is essential for male gametogenesis, as we have found that homozygous null male CK2α′–/– mice are infertile. Those spermatozoa that survived looked like those seen in the human infertility syndromes of 'globozoospermia' or round-headed sperm [38]. As in the CK2β-null phenotype, the CK2α′-null phenotype appears to be

due to a cell-autonomous defect in anterior head shaping that leads to increased apoptosis. Specific isoforms of CK2β are expressed in the testis of *Drosophila melanogaster* [39]. The presence of nuclear β-catenin and the activation of TCF-reporter in differentiating germ cells, particularly in spermatocytes and round spermatids, suggests that canonical Wnt-signaling activation could be downstream of CK2α′ effects on spermatogenesis[40]. Of note, CK2α–/– elongating spermatids had defective anterior head shaping beginning at step 9, and similar defects have been described in a mouse containing hypomorphic alleles of pygo2, a PHD finger protein that acts as a coactivator of β-catenin/TCF complexes [40]. If a relationship can be established between pygo2 and CK2α′, this may suggest that CK2α′, like pygo2, may have functions independent of the canonical Wnt pathway.

CK2 may also play a role in the growth and meiosis of female gametogenesis. In *Xenopus laevis* eggs, the relative amount of CK2 transcripts is high compared to other mRNAs [37], and CK2 activity increases 10- to 12-fold during oogenesis, while CK2α and β transcripts increase 2- to 3-fold [37]. This suggests that CK2 may be regulated at translational or post-translational sites during this period of oocyte growth. In *Xenopus*, CK2 may also regulate progesterone-activated oocyte meiosis as CK2β binds and inhibits the activity of Mos, a key meiotic regulatory kinase [41–43]. Overexpression of CK2β in *Xenopus* oocytes is able to inhibit the G2/M transition promoted by Mos. Interestingly, in *Ciona intestinalis*, the activity of monomeric CK2α oscillated through meiosis, while the regulatory subunit protein is only detectable before fertilization[44], suggesting that the CK2α catalytic subunit also plays a role in meiosis. In this regard, it is possible that CK2 activity plays a role in activating cdc25, the phosphatase that activates cdc2 [45]. In turn, cdc2 may affect CK2 activity [46].

CK2 in organogenesis

Studies on CK2α-ablated mice show that CK2 also plays a role in embryonic heart formation. Targeting of the more abundant CK2α subunit by homologous recombination results in mid-gestation lethality. CK2α-null embryos present structural defects in the heart that are probably the cause of the embryonic lethality [47]. CK2α–/– embryos display morphologically abnormal hearts with enlarged heart size, thin and disorganized endothelial lining, reduced trabeculation, and a thin atrial wall (Fig. 2) [47]. The signaling mechanism involved in CK2α-null phenotypes is not yet described. It is possible that CK2 may directly affects cardiogenic factors such as NKx2.5, as CK2 phosphorylation has been observed to potentiate DNA binding [48]. The target site is phosphorylated *in vivo* and it is conserved among all NK family homeobox genes. Ongoing experiments in our laboratories suggest that canonical Wnt signaling pathway is affected in CK2α-null embryos [Dominguez, unpublished data], which would be the first demonstration that CK2 regulates the canonical Wnt signaling *in vivo* in mammals. Canonical Wnt signaling has been involved in cardiogenesis. Canonical Wnts are expressed in early heart formation from the precardiac mesoderm, through the tubular and looping heart stages to the chambered heart suggesting that canonical Wnt signaling plays a role in different aspects of heart formation [49]. One particular aspect is the generation of the second heart field that will give rise to the right ventricle, among other parts. Conditional ablation of β-catenin in heart progenitors or precursors, shows that canonical Wnt signaling is involved in generation, expansion and migration of $Isl1^+$ progenitor cells in the second heart field [50]. CK2α-null embryos also display hypoplastic right ventricles [Dominguez, unpublished], suggesting that Isl1 progenitors maybe affected.

The CK2α-null embryos also display other phenotypes, such as open cranial neural tube, thinner neuroepithelium, abnormal otic vesicle, reduced branchial arches and hypoplastic limb buds, highlighting the specific role of CK2α in the development of these tissues. Some of these phenotypes may be linked to canonical Wnt signaling. Thus, the dorsal region of the developing otic vesicle, which is abnormal in CK2α–/– embryos, is controlled by Wnt signaling [50,51]. Wnt signals are required for elongation and patterning of the dorso-ventral axis of the limbs (e.g., reduction or absence of limbs are found in loss-of-function mutations of β-catenin in mice [50]). In addition, thinning of the dorsal neuroepithelium and abnormally shaped neural tubes are found in neuronally-targeted conditional β-catenin loss-of-function mice [50]. Not much is known about the role of Wnt signaling in branchial arch formation. It is interesting to note that avian embryonic branchial arches contain a kinase activity that is consistent with CK2, and whose activity is transiently upregulated during branchial arch development [52]. It is possible that CK2 also plays roles in non-canonical Wnt signaling pathways as dishevelled-null embryos display open neural tubes and heart defects [53]. In addition, phosphorylation of dishevelled by CK2 seems to play a key role in the regulation of non-canonical Wnt signaling pathways [54]. Lastly, it is possible that CK2 plays other developmental roles in embryogenesis, as CK2 phosphorylates many developmentally expressed proteins [55].

CK2 in cancer

As noted above, an important theme of cancer molecular pathogenesis is that reactivation of embryonic growth and differentiation pathways can lead to oncogenesis. We make use of this principle in clinical care when we measure embryonic proteins in the circulation, such as carcinoembryonic antigen in gastrointestinal tumors, or α-fetoprotein in germ cell tumors. From a mechanistic viewpoint, tumor growth and embryonic growth share functional properties including rapid cell proliferation, transdifferentiation, cell migration and invasion, angiogenesis, and others. The pathways discussed above in which CK2 acts to regulate development appear to play a role in tumorigenesis, and that has proven to be the case. We have focused our recent studies upon mammary tumorigenesis, but data from other systems and the prospects of inhibiting CK2 for treating cancer will be discussed. Hints that dysregulation of the widely expressed and broadly acting regulatory kinase CK2 could play a role in cancer go back decades to observations of high level CK2 expression in hematopoietic and solid tumors. To answer the question of whether dysregulated expression of CK2 could contribute to tumorigenesis, we overexpressed CK2α in the lymphoid compartment using an immunoglobulin promoter, and demonstrated that multiple lines of transgenic mice developed lymphomas [56]. Since these were monoclonal, it was apparent that CK2α overexpression required additional hits for transformation. These were explored *in vivo* through experiments combining CK2 with other lymphoid oncogenes such as myc [56] or tal-1 [57] or with loss of the tumor suppressor p53 [58]. In addition, we demonstrated that this effect was not confined to the lymphoid lineage, as overexpression in mammary tissue using the mouse mammary tumor virus (MMTV) promoter led to mammary tumors [59]. Upregulated CK2 expression has been documented in human breast cancer specimens and cell lines [14], and in carcinogen-induced mammary tumors [60]. Experiments since the early observations have focused upon identifying some of the pathways that CK2 preferentially activates in promoting tumorigenesis.

Strong associative and functional links suggest that CK2, NF-κB, and Wnt signaling can act together to promote tumorigenesis in the mammary gland. As discussed above, CK2 promotes constitutive and non-canonical IκB degradation and NF-κB activation, and also acts up and downstream in the pathway. In the Wnt pathway, CK2 acts at multiple levels as well, on Dvl, β-catenin, and on TCF/LEF itself. CK2-induced mammary tumors have upregulated NF-κB activity and nuclear β-catenin, and Wnt signaling [59]. Even when mammary tumors are induced with a chemical carcinogen rather than a specific oncogene, co-upregulation of these pathways is observed [60]. Recently, it was demonstrated that the CK2 inhibitor DMAT is toxic to tamoxifen-resistant sublines of human MCF7 breast tumor cells [61]. Another more specific inhibitor, TBB, reduced proliferation and β-catenin expression in ZR-75 breast cancer cells and other tumor cell types [27]; these investigators also showed that siRNA against CK2α had a similar capability in HEK293T cells.

Are the Wnt and NF-κB pathways the only pathways activated by CK2 in tumorigenesis? Are they required for CK2 transformation? The answer to the first question is certainly not, and to the second, perhaps. CK2 has many substrates, and as documented in other reviews in this issue, many of these are in growth control, metabolic, and apoptotic pathways. There is little doubt that the effects of dysregulated overexpression of CK2 in cancer cells affect many of these pathways and a cascade of events leads to transformation, in cooperation with other dysregulated gene expression in the cell (since our transgenic experiments indicate that multiple hits in addition to CK2 dysregulation are required for transformation). The question of whether NF-κB activation and Wnt activation are required for transformation in the presence of overexpressed or dysregulated CK2 is actually difficult to answer. In the whole animal context, major components of the Wnt and NF-κB pathways, like CK2 itself, are absolutely required for development, so one cannot show that CK2-mediated transformation is slowed in their absence. Components of these pathways could be knocked out in a lineage-specific fashion in the mammary gland using Cre-lox technology, or specifically knocked down in cells using shRNA or siRNA technology. In colon cancer cells [62], glioma cells [63], and hepatoma cells [64] knockdown of β-catenin leads to reduced proliferation and increased apoptosis. Knockdown of p65, p52, c-Rel and IKKgamma/NEMO by siRNA increased the doxorubicin sensitivity of HeLa cells [65]; knockdown of p65 in T cells leads to reduced Tax-induced activation of cyclin D2 and cdk6 and cell-cycle progression [66]. Experiments to carry out such knockdowns in the setting of CK2 overexpression will be carried out in the future, but it will be difficult to determine whether the effects are due to modulation of a single pathway or collaborative effects on parallel pathways controlling cell growth and apoptosis. Nonetheless, the ongoing study of pathways involved in both development and cancer will continue to shed light on control of both processes.

Acknowledgements. This research was supported by National Institutes of Health grants R01 CA71796 (DCS), R01 CA 87375 (GES), P01 ES011624 (GES), AHA Scientist Development Grant 0735521T (ID), and an award from the Karin Grunebaum Cancer Research Foundation (ID).

1 Miller, J. R. and Moon, R. T. (1996) Signal transduction through beta-catenin and specification of cell fate during embryogenesis. Genes Dev. 10, 2527–2539.
2 Giles, R. H., van Es, J. H. and Clevers, H. (2003) Caught up in a Wnt storm: Wnt signaling in cancer. Biochim. Biophys. Acta 1653, 1–24.
3 Cadigan, K. M. and Nusse, R. (1997) Wnt signaling: a common theme in animal development. Genes Dev. 11, 3286–3305.
4 Moon, R. T., Kohn, A. D., De Ferrari, G. V. and Kaykas, A. (2004) WNT and beta-catenin signalling: diseases and therapies. Nat. Rev. Genet. 5, 691–701.
5 Willert, K., Brink, M., Wodarz, A., Varmus, H. and Nusse, R. (1997) Casein kinase 2 associates with and phosphorylates dishevelled. EMBO J. 16, 3089–3096.
6 Song, D. H., Dominguez, I., Mizuno, J., Kaut, M., Mohr, S. C. and Seldin, D. C. (2003) CK2 phosphorylation of the armadillo repeat region of beta-catenin potentiates Wnt signaling. J. Biol. Chem. 278, 24018–24025.
7 Duyao, M. P., Kessler, D. J., Spicer, D. B., Bartholomew, C., Cleveland, J. L., Siekevitz, M. and Sonenshein, G. E. (1992) Transactivation of the c-myc promoter by human T cell leukemia virus type 1 tax is mediated by NF kappa B. J. Biol. Chem. 267, 16288–16291.
8 Sovak, M. A., Bellas, R. E., Kim, D. W., Zanieski, G. J., Rogers, A. E., Traish, A. M. and Sonenshein, G. E. (1997) Aberrant nuclear factor-kappaB/Rel expression and the pathogenesis of breast cancer. J. Clin. Invest. 100, 2952–2960.
9 Sonenshein, G. E. (1997) Rel/NF-kappa B transcription factors and the control of apoptosis. Semin. Cancer Biol. 8, 113–119.
10 Romieu-Mourez, R., Kim, D. W., Shin, S. M., Demicco, E. G., Landesman-Bollag, E., Seldin, D. C., Cardiff, R. D. and Sonenshein, G. E. (2003) Mouse mammary tumor virus c-rel transgenic mice develop mammary tumors. Mol. Cell. Biol. 23, 5738–5574.
11 Min, C., Eddy, S. F., Sherr, D. H. and Sonenshein, G. E. (2008) NF-kappaB and epithelial to mesenchymal transition of cancer. J. Cell. Biochem. 104, 733–744.
12 Wu, M., Min, C., Wang, X., Yu, Z., Kirsch, K. H., Trackman, P. C. and Sonenshein, G. E. (2007) Repression of BCL2 by the tumor suppressor activity of the lysyl oxidase propeptide inhibits transformed phenotype of lung and pancreatic cancer cells. Cancer Res. 67, 6278–6285.
13 Romieu-Mourez, R., Landesman-Bollag, E., Seldin, D. C. and Sonenshein, G. E. (2002) Protein kinase CK2 promotes aberrant activation of nuclear factor-kappaB, transformed phenotype, and survival of breast cancer cells. Cancer Res. 62, 6770–6778.
14 Romieu-Mourez, R., Landesman-Bollag, E., Seldin, D. C., Traish, A. M., Mercurio, F. and Sonenshein, G. E. (2001) Roles of IKK kinases and protein kinase CK2 in activation of nuclear factor-kappaB in breast cancer. Cancer Res. 61, 3810–3818.
15 Chu, Z. L., McKinsey, T. A., Liu, L., Qi, X. and Ballard, D. W. (1996) Basal phosphorylation of the PEST domain in the I(kappa)B(beta) regulates its functional interaction with the c-rel proto-oncogene product. Mol. Cell. Biol. 16, 5974–5984.
16 McElhinny, J. A., Trushin, S. A., Bren, G. D., Chester, N. and Paya, C. V. (1996) Casein kinase II phosphorylates I kappa B alpha at S-283, S-289, S-293, and T-291 and is required for its degradation. Mol. Cell. Biol. 16, 899–906.
17 Eddy, S. F., Guo, S., Demicco, E. G., Romieu-Mourez, R., Landesman-Bollag, E., Seldin, D. C. and Sonenshein, G. E. (2005) Inducible IkappaB kinase/IkappaB kinase epsilon expression is induced by CK2 and promotes aberrant nuclear factor-kappaB activation in breast cancer cells. Cancer Res. 65, 11375–11383.
18 Chantome, A., Pance, A., Gauthier, N., Vandroux, D., Chenu, J., Solary, E., Jeannin, J. F. and Reveneau, S. (2004) Casein kinase II-mediated phosphorylation of NF-kappaB p65 subunit enhances inducible nitric-oxide synthase gene transcription *in vivo*. J. Biol. Chem. 279, 23953–23960.
19 Wang, D., Westerheide, S. D., Hanson, J. L. and Baldwin, A. S., Jr. (2000) Tumor necrosis factor alpha-induced phosphorylation of RelA/p65 on Ser529 is controlled by casein kinase II. J. Biol. Chem. 275, 32592–32597.
20 Buchou, T., Vernet, M., Blond, O., Jensen, H. H., Pointu, H., Olsen, B. B., Cochet, C., Issinger, O. G. and Boldyreff, B. (2003) Disruption of the regulatory beta subunit of protein kinase CK2 in mice leads to a cell-autonomous defect and early embryonic lethality. Mol. Cell. Biol. 23, 908–915.
21 Canton, D. A. and Litchfield, D. W. (2006) The shape of things to come: an emerging role for protein kinase CK2 in the regulation of cell morphology and the cytoskeleton. Cell Signal. 18, 267–275.
22 Blond, O., Jensen, H. H., Buchou, T., Cochet, C., Issinger, O.-G. and Boldyreff, B. (2005) Knocking out the regulatory beta subunit of protein kinase CK2 in mice: Gene dosage effects in ES cells and embryos. Mol. Cell. Biochem. 274, 31–37.
23 Dominguez, I., Mizuno, J., Wu, H., Song, D. H., Symes, K. and Seldin, D. C. (2004) Protein kinase CK2 is required for dorsal axis formation in Xenopus embryos. Dev. Biol. 274, 110–124.
24 Hammerlein, A., Weiske, J. and Huber, O. (2005) A second protein kinase CK1-mediated step negatively regulates Wnt signalling by disrupting the lymphocyte enhancer factor-1/beta-catenin complex. Cell. Mol. Life Sci. 62, 606–618.
25 Gao, Y. and Wang, H. Y. (2006) Casein kinase 2 is activated and essential for wnt/beta -catenin signaling. J. Biol. Chem. 281, 18394–18400.
26 Wang, S. and Jones, K. A. (2006) CK2 controls the recruitment of Wnt regulators to target genes *in vivo*. Curr. Biol. 16, 2239–2244.
27 Tapia, J. C., Torres, V. A., Rodriguez, D. A., Leyton, L. and Quest, A. F. (2006) Casein kinase 2 (CK2) increases survivin expression via enhanced beta-catenin-T cell factor/lymphoid enhancer binding factor-dependent transcription. Proc. Natl. Acad. Sci. USA 103, 15079–15084.
28 Song, D. H., Sussman, D. J. and Seldin, D. C. (2000) Endogenous protein kinase CK2 participates in Wnt signaling in mammary epithelial cells. J. Biol. Chem. 275, 23790–23797.
29 Homma, M.K., Li, D., Krebs, E. G., Yuasa, Y. and Homma, Y. (2002) Association and regulation of casein kinase 2 activity by adenomatous polyposis coli protein. Proc. Natl. Acad. Sci. USA 99, 5959–5964.
30 Miravet, S., Piedra, J., Miro, F., Itarte, E., Garcia de Herreros, A. and Dunach, M. (2002) The transcriptional factor Tcf-4 contains different binding sites for beta-catenin and plakoglobin. J. Biol. Chem. 277, 1884–1891.
31 Pukrop, T., Gradl, D., Henningfeld, K. A., Knochel, W., Wedlich, D. and Kuhl, M. (2001) Identification of two regulatory elements within the high mobility group box transcription factor XTCF-4. J. Biol. Chem. 276, 8968–8978.
32 Packman, L. C., Kubota, K., Parker, J. and Gay, N. J. (1997) Casein kinase II phosphorylates Ser468 in the PEST domain of the Drosophila IkappaB homologue cactus. FEBS Lett. 400, 45–50.
33 Liu, Z. P., Galindo, R. L. and Wasserman, S. A. (1997) A role for CKII phosphorylation of the cactus PEST domain in dorsoventral patterning of the Drosophila embryo. Genes Dev. 11, 3413–3422.
34 Lin, J. M., Kilman, V. L., Keegan, K., Paddock, B., Emery-Le, M., Rosbash, M. and Allada, R. (2002) A role for casein kinase 2alpha in the Drosophila circadian clock. Nature 420, 816–820.
35 Jauch, E., Wecklein, H., Stark, F., Jauch, M. and Raabe, T. (2006) The Drosophila melanogaster DmCK2beta transcription unit encodes for functionally non-redundant protein isoforms. Gene 4, 142–152.
36 Kuntamalla, P. P., Kunttas-Tatli, E., Karandikar, U., Bishop, C. P. and Bidwai, A. P. (2008) Drosophila protein kinase CK2 is rendered temperature-sensitive by mutations of highly conserved residues flanking the activation segment. Mol. Cell. Biochem. 323, 49–60.
37 Wilhelm, V., Rojas, P., Gatica, M., Allende, C. C. and Allende, J. E. (1995) Expression of the subunits of protein kinase CK2 during oogenesis in Xenopus laevis. Eur. J. Biochem. 232, 671–676.

38 Xu, X., Toselli, P. A., Russell, L. D. and Seldin, D. C. (1999) Globozoospermia in mice lacking the casein kinase II alpha′ catalytic subunit. Nat. Genet. 23, 118–121.
39 Kalmykova, A. I., Dobritsa, A. A. and Gvozdev, V. A. (1997) The beta CK2tes gene encodes the tissue-specific regulatory subunit of casein kinase 2 in Drosophila melanogaster. Biochemistry (Mosc) 62, 455–460.
40 Nair, M., Nagamori, I., Sun, P., Mishra, D. P., Rheaume, C., Li, B., Sassone-Corsi, P. and Dai, X. (2008) Nuclear regulator Pygo2 controls spermiogenesis and histone H3 acetylation. Dev. Biol. 320, 446–455.
41 Stanford, J. S., Lieberman, S. L., Wong, V. L. and Ruderman, J. V. (2003) Regulation of the G2/M transition in oocytes of Xenopus tropicalis. Dev. Biol. 260, 438–448.
42 Lieberman, S. L. and Ruderman, J. V. (2004) CK2 beta, which inhibits Mos function, binds to a discrete domain in the N-terminus of Mos. Dev. Biol. 268, 271–279.
43 Chen, M., Li, D., Krebs, E. G. and Cooper, J. A. (1997) The casein kinase II beta subunit binds to Mos and inhibits Mos activity. Mol. Cell. Biol. 17, 1904–1912.
44 Russo, G. L., Tosto, M., Mupo, A., Castellano, I., Cuomo, A. and Tosti, E. (2004) Biochemical and functional characterization of protein kinase CK2 in ascidian Ciona intestinalis oocytes at fertilization. Cloning and sequence analysis of cDNA for alpha and beta subunits. J. Biol. Chem. 279, 33012–33023.
45 Bibby, A. C. and Litchfield, D. W. (2005) The multiple personalities of the regulatory subunit of protein kinase CK2: CK2 dependent and CK2 independent roles reveal a secret identity for CK2beta. Int. J. Biol. Sci. 1, 67–79.
46 Mulner-Lorillon, O., Cormier, P., Labbe, J. C., Doree, M., Poulhe, R., Osborne, H. and Belle, R. (1990) M-phase-specific cdc2 protein kinase phosphorylates the beta subunit of casein kinase II and increases casein kinase II activity. Eur. J. Biochem. 193, 529–534.
47 Lou, D. Y., Dominguez, I., Toselli, P., Landesman-Bollag, E., O'Brien, C. and Seldin, D. C. (2008) The alpha catalytic subunit of protein kinase CK2 is required for mouse embryonic development. Mol. Cell. Biol. 28, 131–139.
48 Kasahara, H. and Izumo, S. (1999) Identification of the *in vivo* casein kinase II phosphorylation site within the homeodomain of the cardiac tisue-specifying homeobox gene product Csx/Nkx2.5. Mol. Cell. Biol. 19, 526–536.
49 Eisenberg, L. M. and Eisenberg, C. A. (2006) Wnt signal transduction and the formation of the myocardium. Dev. Biol. 293, 305–315.
50 Grigoryan, T., Wend, P., Klaus, A. and Birchmeier, W. (2008) Deciphering the function of canonical Wnt signals in development and disease: conditional loss- and gain-of-function mutations of beta-catenin in mice. Genes Dev. 22, 2308–2341.
51 Riccomagno, M. M., Takada, S. and Epstein, D. J. (2005) Wnt-dependent regulation of inner ear morphogenesis is balanced by the opposing and supporting roles of Shh. Genes Dev. 19, 1612–1623.
52 Jones, N. C., Farlie, P. G., Minichiello, J. and Newgreen, D. F. (1999) Detection of an appropriate kinase activity in branchial arches I and II that coincides with peak expression of the Treacher Collins syndrome gene product, treacle. Hum. Mol. Genet. 8, 2239–2245.
53 Hamblet, N. S., Lijam, N., Ruiz-Lozano, P., Wang, J., Yang, Y., Luo, Z., Mei, L., Chien, K. R., Sussman, D. J. and Wynshaw-Boris, A. (2002) Dishevelled 2 is essential for cardiac outflow tract development, somite segmentation and neural tube closure. Development 129, 5827–5838.
54 Bryja, V. E., Schambony, A., Cajanek, L. S., Dominguez, I., Arenas, E. and Schulte, G. (2008) beta-Arrestin and casein kinase 1/2 define distinct branches of non-canonical WNT signalling pathways. EMBO Rep. 9, 1244–1250.
55 Meggio, F. and Pinna, L. A. (2003) One-thousand-and-one substrates of protein kinase CK2? FASEB J. 17, 349–368.
56 Seldin, D. C. and Leder, P. (1995) Casein kinase II alpha transgene-induced murine lymphoma: relation to theileriosis in cattle [see comments]. Science 267, 894–897.
57 Kelliher, M. A., Seldin, D. C. and Leder, P. (1996) Tal-1 induces T cell acute lymphoblastic leukemia accelerated by casein kinase IIα. EMBO J. 15 5160–5166.
58 Landesman-Bollag, E., Channavajhala, P. L., Cardiff, R. D. and Seldin, D. C. (1998) p53 deficiency and misexpression of protein kinase CK2alpha collaborate in the development of thymic lymphomas in mice. Oncogene 16, 2965–2974.
59 Landesman-Bollag, E., Romieu-Mourez, R., Song, D. H., Sonenshein, G. E., Cardiff, R. D. and Seldin, D. C. (2001) Protein kinase CK2 in mammary gland tumorigenesis. Oncogene 20, 3247–3257.
60 Currier, N., Solomon, S. E., Demicco, E. G., Chang, D. L., Farago, M., Ying, H., Dominguez, I., Sonenshein, G. E., Cardiff, R. D., Xiao, Z. X., Sherr, D. H. and Seldin, D. C. (2005) Oncogenic signaling pathways activated in DMBA-induced mouse mammary tumors. Toxicol. Pathol. 33, 726–737.
61 Yde, C. W., Frogne, T., Lykkesfeldt, A. E., Fichtner, I., Issinger, O. G. and Stenvang, J. (2007) Induction of cell death in antiestrogen resistant human breast cancer cells by the protein kinase CK2 inhibitor DMAT. Cancer Lett. 256, 229–237.
62 Huang, W. S., Wang, J. P., Wang, T., Fang, J. Y., Lan, P. and Ma, J. P. (2007) ShRNA-mediated gene silencing of beta-catenin inhibits growth of human colon cancer cells. World J. Gastroenterol. 13, 6581–6587.
63 Pu, P., Zhang, Z., Kang, C., Jiang, R., Jia, Z., Wang, G. and Jiang, H. (2008) Downregulation of Wnt2 and beta-catenin by siRNA suppresses malignant glioma cell growth. Cancer Gene Ther. 16, 351–361.
64 Apte, U., Zeng, G., Thompson, M. D., Muller, P., Micsenyi, A., Cieply, B., Kaestner, K. H. and Monga, S. P. (2007) beta-Catenin is critical for early postnatal liver growth. Am. J. Physiol. Gastrointest. Liver Physiol. 292, G1578–1585.
65 Tapia, M. A., Gonzalez-Navarrete, I., Dalmases, A., Bosch, M., Rodriguez-Fanjul, V., Rolfe, M., Ross, J. S., Mezquita, J., Mezquita, C., Bachs, O. et al. (2007) Inhibition of the canonical IKK/NF kappa B pathway sensitizes human cancer cells to doxorubicin. Cell Cycle 6, 2284–2292.
66 Iwanaga, R., Ozono, E., Fujisawa, J., Ikeda, M. A., Okamura, N., Huang, Y. and Ohtani, K. (2008) Activation of the cyclin D2 and cdk6 genes through NF-kappaB is critical for cell-cycle progression induced by HTLV-I Tax. Oncogene 27, 5635–5642.