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CK2: The kinase controlling the Hsp90 chaperone machinery

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Abstract. CK2 is a ubiquitous and essential protein kinase with pleiotropic substrates and function, but it remains unclear how, when, and where CK2 activity is regulated in cells. Hsp90 is a major molecular chaperone that is required for the folding and function of its client proteins. A complex containing Hsp90 and its client protein includes co-chaperones such as steroid hormone receptor-specific FKBP52 and signaling kinase-specific Cdc37. Co-chaperones work cooperatively with Hsp90 to stabilize client proteins and to keep them in a conformation amenable to activation under appropriate conditions. In this review, critical roles of CK2 in the regulation of the Hsp90-mediated chaperone system are described. CK2 phosphorylates and modulates Hsp90 and its co-chaperones FKBP52 and Cdc37. CK2-dependent phosphorylation of Cdc37 is essential for the chaperoning function of Hsp90-Cdc37 for multiple signaling protein kinases. The tumor kinome appears to become addicted to the Hsp90-Cdc37 chaperone system, thus, targeting Hsp90, Cdc37, and CK2 is a promising strategy for cancer treatment. (Part of a Multi-author Review)

Keywords. CK2, molecular chaperone, Hsp90, Cdc37, FKBP52, protein kinase, cancer.

Introduction

CK2 is an essential and ubiquitous protein kinase with scanty known regulatory mechanism [1 – 5]. CK2 recognizes serine and threonine residues in an acidic environment and has been shown to phosphorylate both *in vivo* and *in vitro* a broad spectrum of endogenous and artificial substrates involved in transcription, translation, signal transduction, metabolic control, and many other cellular functions [6, 7]. A large number of CK2 substrates have been shown to function in a fashion dependent on CK2-mediated phosphorylation. However, it remains still unclear how, when, and where CK2 activity is regulated in cells. The lack of known regulatory mechanism and the pleiotropicity of the kinase make it difficult to pinpoint the physiological role of CK2 in the complicated cellular signaling network figure.
In this review, the intimate relationship between CK2 and the Hsp90 molecular chaperone system is described. In particular, the importance of an indispensable role of CK2-dependent phosphorylation of a kinase-specific Hsp90 co-chaperone, Cdc37, is emphasized. Hsp90 and Cdc37 are now established to be essential for the correct folding and function of many signaling protein kinases in cells. Thus, CK2 activity, via the phosphorylation of Cdc37, is required for the proper functions of a large number of diverse cellular signaling protein kinases. The interaction between CK2 and the Hsp90 molecular chaperone system makes CK2 even more pluripotent and pleiotropic. Finally, a possibility that the CK2-Cdc37-Hsp90 machinery could be a novel and efficient pharmacological molecular target for cancer chemotherapy is proposed.

Hsp90: A major molecular chaperone

Exposure of living cells to environmental stresses such as higher-than-normal temperature induces expression of a specific set of proteins called heat-shock proteins (HSPs) or stress proteins. Even in unstressed conditions, most stress proteins exist abundantly in cells and play essential roles in their growth and function. Stress proteins bind to cellular target pro-

teins, protect them from making aggregates or being destroyed or inactivated, thereby stabilize and maintain their structure and function [8]. 'Molecular chaperones' are a special set of well-conserved protein groups which assist the proper folding of cellular proteins and prevent unnecessary and harmful protein-protein interactions during the protein synthesis, or even after release from the ribosomes [9]. Therefore, most stress proteins can be categorized as molecular chaperones. Under stress condition, there is an increasing possibility of misfolding, aggregation, and inactivation of many proteins; thus, the induction of molecular chaperones by cellular stresses might be rational.

Hsp90 is one of the major molecular chaperones, and Hsp90 concentration in the cytosol is as high as 10–100 μM (1–10 mg/ml), sometimes occupying about 1% of total cytosolic proteins. Hsp90 is a part of a ubiquitously expressed multiprotein molecular chaperone system that is required for the folding, maturation, and stabilization of specific set of target proteins (called client proteins) [10–13]. The list of Hsp90 client proteins includes many functional proteins which are involved in a wide variety of cellular signal transduction systems. Generally, Hsp90 binds and stabilizes these signaling proteins, and keeps them in a conformation amenable to activation under appropriate conditions. Hsp90 possesses ATP-binding and ATPase activity, which is essential for its chaperone cycling mechanism and function [14, 15].

Much of our current understanding of interactions between Hsp90 and its client proteins is modeled on extensive characterizations of steroid hormone receptors, one of the major Hsp90-client protein groups [16, 17]. Steroid hormone receptors exist in the cytosol as complexes with Hsp90, and this complex formation is essential for the hormone-binding activity of the receptors. Upon binding of a corresponding hormone, Hsp90 dissociates from the complex, and the receptor translocates into the nucleus where it functions as a DNA-binding transcription factor. On the other hand, steroid hormone receptors cannot achieve hormone-dependent transcription activity without binding to Hsp90. Thus, Hsp90 is not a simple activation molecule or an inhibitory factor, but involved in the full signal-transducing activity of steroid hormones. Another major group of Hsp90-client proteins consists of protein kinases that are involved in cellular signal transduction systems [10, 18–20]. They include many important protein kinases involved in cell growth and survival [21]. Hsp90 binds to client protein kinases and thus ensures that these kinases exist stably in cells and that they can perform their appropriate tasks when (and only when) necessary. The updating current list of Hsp90-client proteins can be downloaded from http://www.picard.ch/downloads/Hsp90-interactors.pdf

Essential association and regulatory phosphorylation of Hsp90 by CK2

An initial implication of the relationship between Hsp90 and CK2 was revealed when CK2 was found to be co-purified with a 90-kDa substrate [22], which was later identified as Hsp90 [23]. In addition, a purified preparation of Hsp90 contained CK2 which phosphorylated Hsp90, and CK2 was co-immunoprecipitated with Hsp90 from cell extracts [24]. In a buffer of low ionic strength, CK2 forms large inactive aggregates [25], but Hsp90 dissociates the aggregates, thus activating CK2 [24, 26]. Both α and β isoforms of Hsp90 can be constitutively phosphorylated by CK2 *in vitro* and *in vivo* at two serine residues in a highly charged region of the molecule [27]. Hsp90 was shown to enhance the kinase activity of eukaryotic initiation factor 2α kinase (eIF2αK), but the stimulatory effect was abolished after incubation of Hsp90 with a phosphatase PP1 [28]. Re-phosphorylation of dephosphorylated inactive Hsp90 by CK2 restored the biological activity of Hsp90 to stimulate eIF2αK [28], suggesting that CK2-dependent phosphorylation of Hsp90 is required for the chaperone activity of Hsp90 toward client kinases (Fig. 1). By contrast, hyperphosphorylation of Hsp90 in threonine residues caused by a treatment of cells with a phosphatase inhibitor okadaic acid has been shown to destabilize the association of Hsp90 with a client kinase v-Src [29]. A link between Hsp90 phosphorylation and its chaperoning function was also shown for another client protein, reovirus σ1 [30]. Thus, Hsp90 phosphorylation should be a part of the Hsp90 cycling mechanism and likely plays an important role in its chaperoning function (Fig. 1). Several protein kinases other than CK2 are also responsible for the phosphorylation of Hsp90. Double-stranded DNA-activated protein kinase was reported to phosphorylate two amino-terminal threonine residues unique to the Hsp90 α isoform [31]. A functional proteomic screening identified Hsp90α and Hsp90β as substrates for a serine/threonine protein kinase Akt [32], which is an Hsp90-client kinase [33]. In summary, it is clear that Hsp90 phosphorylation is intimately linked to its chaperoning function (Fig. 1), however, the precise molecular mechanism of the regulation and responsible kinases other than CK2 remain to be elucidated.

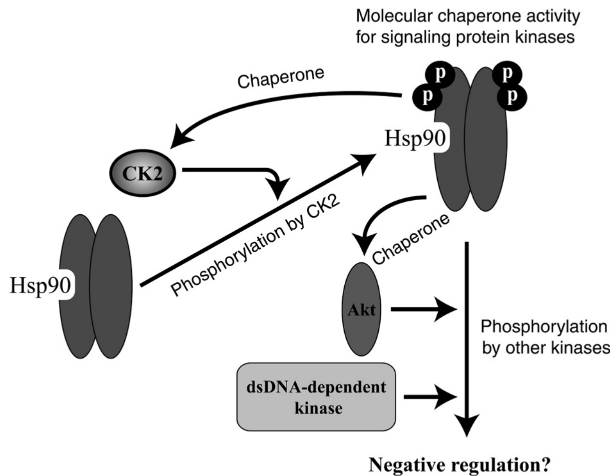

Figure 1. Essential phosphorylation of Hsp90 by CK2. CK2 phosphorylates Hsp90 at two serine residues in a highly charged region. CK2-dependent phosphorylation of Hsp90 is required for the chaperone activity of Hsp90 toward client proteins including CK2 itself. Hsp90 can be phosphorylated by Akt and dsDNA-dependent kinase.

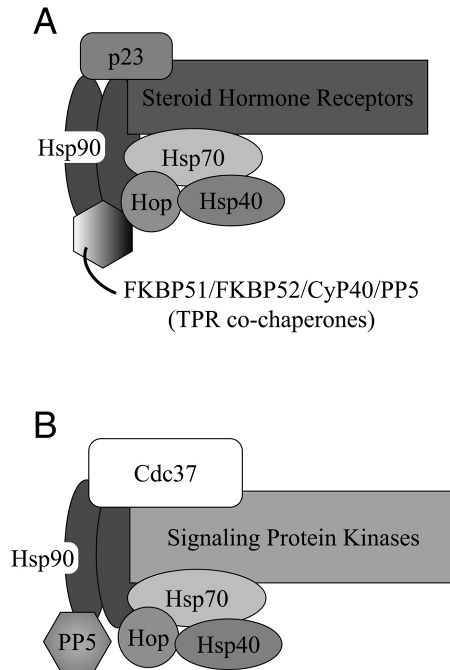

Figure 2. Hsp90, its co-chaperones, and client proteins. *(A)* Schematic illustration of a complex containing steroid hormone receptors and molecular chaperones. *(B)* Schematic illustration of a complex containing signaling protein kinases and molecular chaperones.

Phosphorylation by CK2 of FKBP52, a steroid hormone-specific Hsp90 co-chaperone

Hsp90 does not work alone. A complex containing Hsp90 and its client protein often includes other proteins called 'co-chaperones', which work cooperatively together with Hsp90 to 'chaperone' the client protein [8, 11–13, 34]. Hsp90 requires many specific co-chaperones for its function corresponding to a specific group of client proteins (Fig. 2). The mature steroid hormone receptor complexes contain any one of several co-chaperones that mutually compete for binding to Hsp90 [16, 17, 35]. These co-chaperones share a tetratricopeptide repeat (TPR) domain that mediates binding to the highly conserved C-terminal MEEVD sequence in Hsp90 (Fig. 2A). Among the TPR co-chaperones observed in steroid receptor complexes are immunophilins such as two members of the FK506-binding protein (FKBP) family – FKBP51 and FKBP52 (hsp56, p59, HBI) – and cyclophilin-40 (CyP40) [36, 37]. FKBP51 and FKBP52 are characterized by a conserved binding domain for the immunosuppressive drug FK506 (Fig. 3), and they show peptidylprolylisomerase activity as well as a chaperone-like activity *in vitro* [36–38]. FKBP52 has a stimulating effect on many steroid hormone receptors [38, 39]. The physiological importance of FKBP52 in steroid hormone signal transduction is supported by the fact that male mice lacking FKBP52 gene have many physiological features consistent with androgen insensitivity, while FKBP52-deficient female mice are infertile due to uterine defects caused by progesterone insensitivity [40, 41].

Despite the structural and biochemical similarities between FKBP51 and FKBP52, FKBP51 can inhibit FKBP52-mediated potentiation of steroid hormone receptor function; thus FKBP52 and FKBP51 have opposing effects on steroid hormone action [41].

One structural distinction between the two FKBPs lies in a linker loop (hinge region) between the first and second FK506-binding domains, where FKBP52 possesses a well-conserved putative CK2-phosphorylation sequence (Fig. 3), while FKBP51 does not. In fact, FKBP52 is a substrate of CK2 both *in vitro* and *in vivo*, and the conserved threonine residue in the linker loop region is the major site of phosphorylation by CK2 [42, 43] (Fig. 3). An *in vitro* reconstitution experiment suggested that CK2-phosphorylated FKBP52 has lower Hsp90-binding activity, indicating that phosphorylation by CK2 weakens FKBP52 function [42]. By contrast, in a reticulocyte lysate system, phosphorylation-incapable and phospho-mimicking mutations in the CK2-phosphorylation site did not change the binding of FKBP52 to Hsp90 as compared to wild type FKBP52 [43]. Thus, it remains unclear whether the phosphorylation of the linker loop region by CK2 affects the Hsp90-binding activity of FKBP52 *in vivo.* Interestingly, the activity of FKBP52 to potentiate steroid hormone receptors in yeast and in mouse cells is greatly diminished by the phospho-mimicking mutation in the CK2-phosphorylation site [43]. In

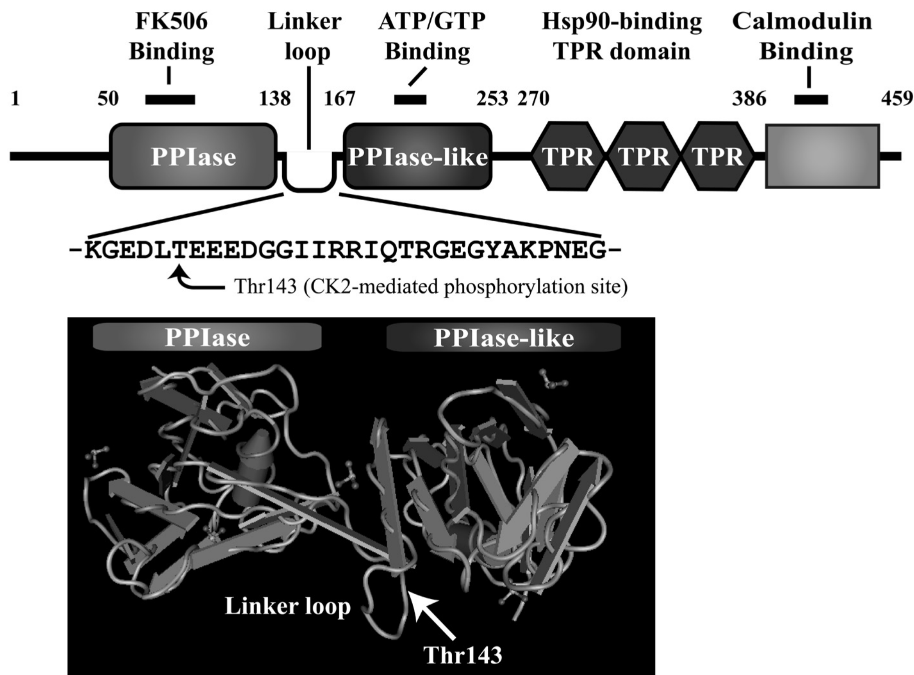

Figure 3. Structure and CK2-dependent phosphorylation site of a steroid hormone receptor specific co-chaperone FKBP52. Domain structure of FKBP52, the amino acid sequence surrounding the CK2-phosphorylation site of FKBP52, and a crystal structure image of a part of FKBP52 (PDB ID: 1Q1C / MMDB ID: 28163) are illustrated.

summary, the phosphorylation of the linker loop region by CK2 negatively regulates the physiological activity of FKBP52 (Fig. 3), however, the molecular mechanism of this functional regulation remains unclear.

Cdc37, a kinase-targeting co-chaperone of Hsp90

Signaling protein kinases are often associated with Hsp90, and these Hsp90-client kinases, in contrast to steroid hormone receptors, make complexes with a co-chaperone Cdc37 along with Hsp90, but not with FKBP51/52 [20, 44–46]. Cdc37 is rarely observed in Hsp90-client proteins other than protein kinases, and thus is called a kinase-targeting or kinase-specific co-chaperone (Fig. 2B). Cdc37 has a general role in kinase biogenesis and maturation, and functions either during or immediately after translation to protect nascent chains of a subset of protein kinases from misfolding, aggregation, and degradation [20, 44–46]. A recent analysis suggests that more than 75 % of the yeast kinome is affected by functional reduction of Cdc37 [47], but the molecular basis of specific and selective interaction of Cdc37 with kinases remains unclear. Cdc37 was initially isolated in a genetic screen for yeast mutants defective in cell cycle progression [48]. Mammalian p50, originally found to make complexes with $pp60^{v\text{-}src}$ and Hsp90, was identified as a homologue of Cdc37 [49]. The interaction of Cdc37 with protein kinases is primarily mediated by the N-terminal segment [50, 51], while binding site for Hsp90 has been mapped to the central and C-terminal region of Cdc37 [52–54], further supporting the notion that Cdc37 functions as a kinase-targeting co-chaperone of Hsp90 (Figs. 2b and 4). Cdc37 has a regulatory function and can arrest the ATPase-coupled Hsp90-chaperone cycle [53–55]. Cdc37 has also been demonstrated to possess chaperone activity on its own, independent of Hsp90 [56–58]. Cdc37 is essential for cell viability in yeast as well as in several other species [59, 60]. Although yeast Cdc37 shares only 20 % amino acid sequence identity with its mammalian counterpart, the first portion of the N-terminal domain is strongly conserved, and the roles of Cdc37 in cellular processes that mediate the stabilization and activation of many protein kinases appear to be the same for different species. The updated current list of Cdc37-client proteins can be downloaded from http://www.picard.ch/downloads/Cdc37interactors.pdf

Phosphorylation of Cdc37 by CK2

A temperature-sensitive mutant strain of *Saccharomyces cerevisiae,* which encodes Cdc37 with Ser14 to Leu14 substitution [61], was shown to be defective in supporting normal growth [62], suggesting an important role of the serine residue in the highly conserved N-terminal region for Cdc37 function. This serine residue is perfectly conserved among species, which corresponds to Ser13 in vertebrates (Fig. 4). The serine is followed by a cluster of acidic amino acids,

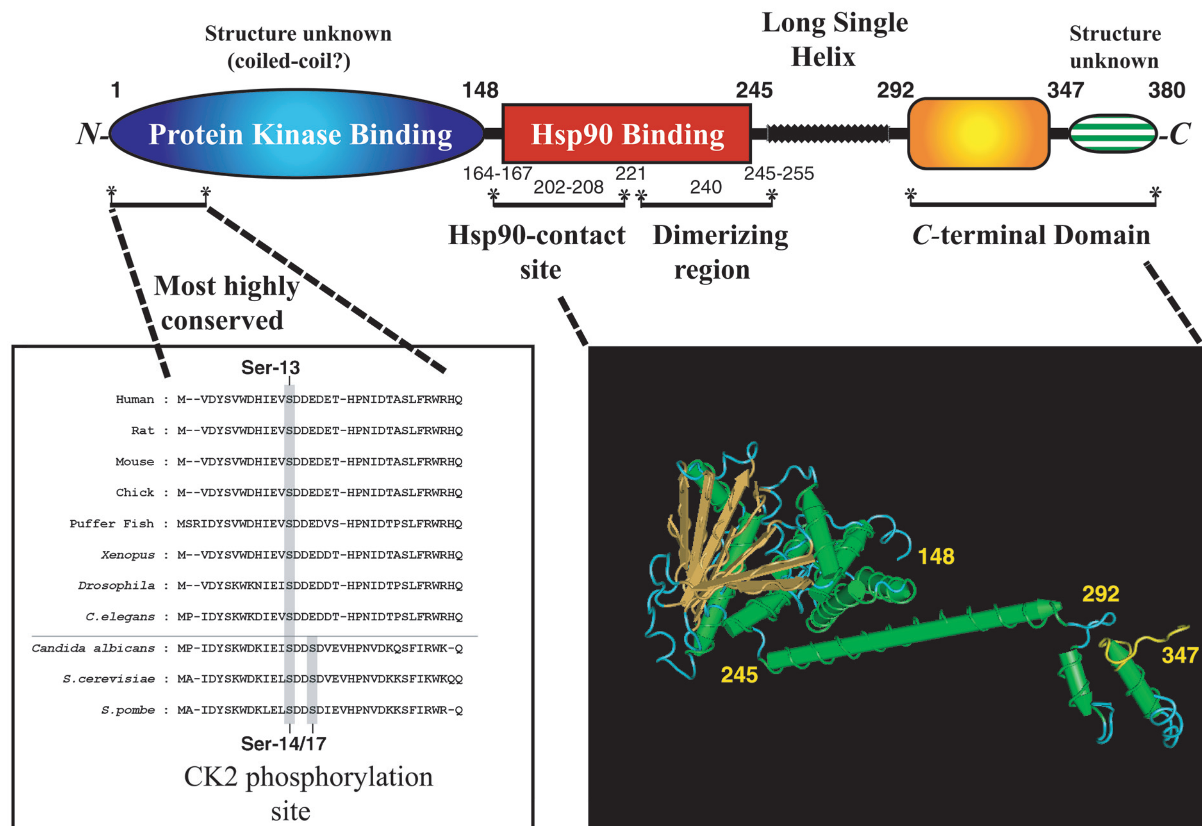

Figure 4. Structure and the CK2-dependent phosphorylation site of the kinase-targeting co-chaperone Cdc37. Domain structure of Cdc37, amino acid sequences surrounding the CK2-phosphorylation site of Cdc37, and a crystal structure image of a part of Cdc37 (PDB ID: 1US7 / MMDB ID: 26274)) are illustrated.

suggesting that this site can possibly be phosphorylated by CK2 (Fig. 4). Direct genetic interaction between Cdc37 and CK2 was discovered when Cdc37 was identified as a multicopy suppressor of a temperature-sensitive allele of the *S. cerevisiae* CKA1 gene encoding the catalytic subunit of CK2 [63]. In fact, purified recombinant Cdc37 of yeast [56] and mammalian [64] sources was radiolabeled when incubated with CK2 in the presence of [γ-^{32}P]ATP. Cdc37 is a phosphoprotein, and was phosphorylated in Ser13 when expressed in rabbit reticulocyte lysates [65] and in mammalian cultured cells [64, 66]. As compared with wild-type Cdc37, mutants in the CK2-dependent phosphorylation site showed negligible *in vivo* phosphorylation in both yeast and mammalian systems [63–65]. In addition, Cdc37 phosphorylation was greatly diminished in a CK2 knockout yeast strain [63] and in mammalian cells treated with a specific CK2 inhibitor [64, 67]. Moreover, the serine is the unique phosphorylation site of Cdc37 *in vivo,* and CK2 is the only kinase that phosphorylates Cdc37 under normal conditions both in yeast and in mammals. Taken together, Cdc37 is directly phosphorylated in Ser13 (mammalian)/Ser14 (yeast) by CK2 both *in vitro* and *in vivo.*

CK2-dependent phosphorylation of Cdc37 is required for multiple Hsp90-client signaling protein kinases

The functional, physiological, and general importance of the CK2-dependent phosphorylation of Cdc37 has been revealed by many different approaches. Mutants in the phosphorylation site of Cdc37 show a severely affected phenotype in yeast, displaying an extremely slow growth rate and an elongated, enlarged morphology [62, 63]. In a condition where Hsp90 activity is reduced, wild-type Cdc37 could rescue the viability of yeast, whereas the CK2-phosphorylation site mutant could not [63]. Genetic evidence indicated that the mutants were defective in supporting many kinases, including v-Src, Ste11, Kin28, and Mps1 [62, 63, 68]. The complex formation of eIF2αK and Cdc37 in rabbit reticulocyte lysates was reduced by the mutation in the CK2-phosphorylation site of Cdc37 [65]. The CK2-phosphorylation site was essential for the binding of Cdc37 to various kinases, including Raf1, Akt, Auror-

aB, Cdk4, v-Src, and MOK in mammalian cells [64, 66]. These client kinases were unstabilized and degraded when the CK2-phosphorylation site of Cdc37 was mutated. Cdc37 is involved in the MAPK-signaling routes responsive to osmotic and cell wall stresses via stabilizing Hog1p, and the phosphorylation site mutants of Cdc37 had reduced binding ability to Hog1p and showed an osmosensitive and cell wall stress-sensitive phenotype [69]. The CK2-dependent phosphorylation of Cdc37 in Ser13 was important for the recruitment of Hsp90 to protein kinase-Cdc37 complexes, but not for the Hsp90-binding activity of Cdc37 [64, 65]. The requirement of phospho-Ser13 (Ser14 in yeast) of Cdc37 for complex formation of such a diverse set of protein kinases suggests that Cdc37 with the CK2-phosphorylated serine is the form in which Cdc37 generally exists in Hsp90 complexes with client kinases [66, 70]. Finally, direct inhibition of CK2 *in vivo* by treating cells with a CK2-specific low molecular weight inhibitor suppressed the phosphorylation of Cdc37 and decreased the intracellular amounts of Cdc37-dependent protein kinases [64]. Taken altogether, phosphorylation of Cdc37 by CK2 in the conserved N-terminal region is important and indispensable for the essential role of Cdc37 in maintaining Hsp90-Cdc37-kinase heterocomplexes, and hence for the efficient activation and physiological function of client protein kinases [20, 46, 71] (Fig. 5).

Interestingly, CK2 itself is in turn a Cdc37 client kinase both *in vitro* [56] and *in vivo* [63]. Indeed, Hsp90 and Cdc37 are both required for optimum CK2 activity in yeast cells, but Cdc37 is the limiting component to maintain CK2 function [63]. This suggests that CK2 mediates a positive feedback loop with Cdc37 to promote multiple client protein kinases (Fig. 5).

Until recently, no *in vitro* system using purified proteins had been established to study protein kinase folding, and the molecular details of how they are chaperoned remain elusive. Checkpoint kinase 1 (Chk1), an Hsp90-client kinase, is a serine/threonine kinase that regulates DNA damage checkpoints. Chk1 expressed in *Escherichia coli* was used as a folding substrate for chaperone activity *in vitro,* enabling the identification of minimum chaperone components for client kinases [72]. Combination of multiple molecular chaperones, including all of Hsp90, Hsp70, Ydj1 (yeast Hsp40), Hop, and Cdc37, could not make properly folded active Chk1 kinase (see Fig. 2B). Importantly, addition of purified CK2 to this mixture induced 600-fold activation of Chk1, indicating a critical role for CK2 in the quality control of signaling protein kinases [72]. The stimulation was not observed when the phosphorylation-incapable mutant of Cdc37 was used, showing that activation by CK2 was due to the CK2-mediated phosphorylation of Cdc37. Although CK2 itself does not have molecular chaperone activity, CK2 should be distinctly a requisite member of the molecular chaperone system for the kinome.

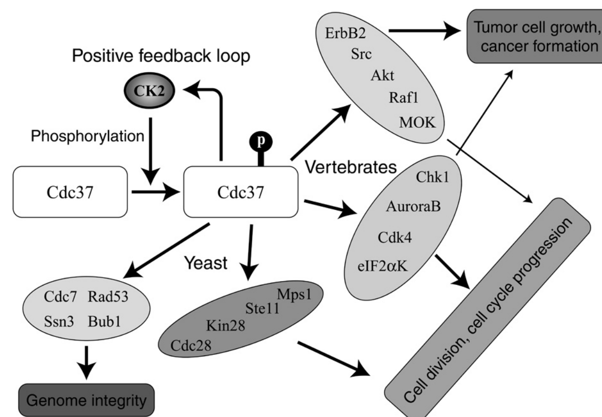

Figure 5. CK2 and Cdc37 constitute a positive feedback system to promote multiple client protein kinases in yeast and in vertebrates. CK2-dependent phosphorylation is critical for the essential role of Cdc37 in maintaining Hsp90-Cdc37-kinase heterocomplexes and for the physiological function of multiple client protein kinases. There are many other Cdc37-dependent signaling client kinases, which are omitted for simplicity.

De-phosphorylation of CK2-phosphorylated Cdc37 should also play an important role in the functional regulation of Cdc37. Isolated Cdc37 was rapidly de-phosphorylated upon incubation with a non-specific λ-phosphatase, but phospho-Ser13 in the Hsp90-Cdc37-Cdk4 complex was highly resistant to de-phosphorylation [70]. The result indicates that the N-terminus of Cdc37 is fully accessible from outside, but phospho-Ser13 in the Hsp90-Cdc37-kinase complex is buried or occluded in the ternary complex and inaccessible to the phosphatase. This supports the idea that phospho-Ser13 consists the binding surface between Cdc37-Hsp90 and client kinases [73]. PP5 is a protein phosphatase associated with Hsp90 through its TPR domain [36, 74]. PP5 de-phosphorylates phospho-Ser13 in Cdc37 only when both Cdc37 and PP5 are simultaneously bound to the same Hsp90 dimer [70] (Fig. 6). Collectively, phosphorylation by CK2 and de-phosphorylation by PP5 may drive a directional cycle of Cdc37 function in signaling kinase activation, and both should be regulated phenomena, equally important for Cdc37's biological function (Fig. 6). A longer incubation with λ-phosphatase compellingly de-phosphorylated Cdc37 in the Hsp90-Cdc37-kinase complex without disrupting it, suggesting that the phosphorylation is not essential for integrity of the complex, once assembled [70]. The crystal structure of Cdc37-Hsp90 complex was determined [53] (see Fig. 4), but the Cdc37 construct used in the study

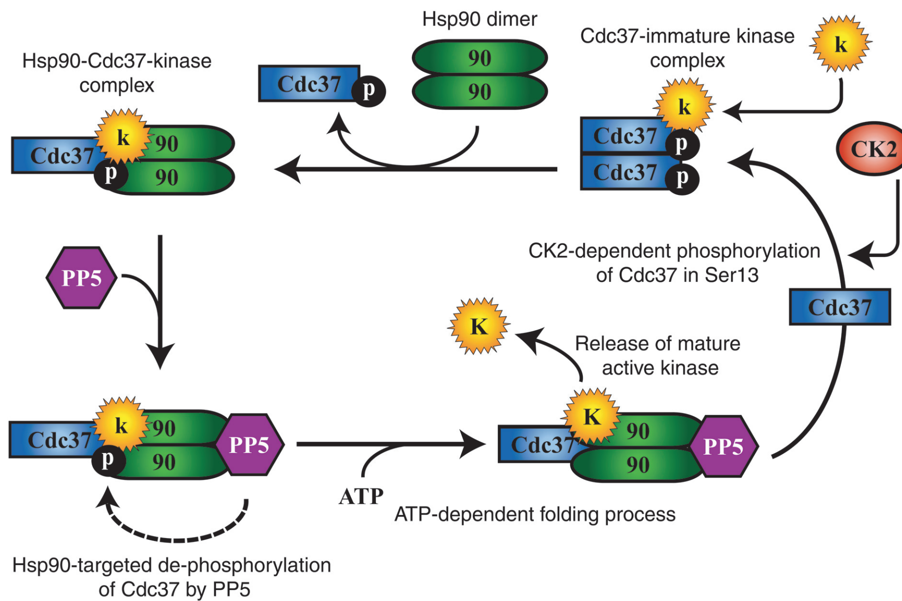

Figure 6. The functional cycle of Hsp90-Cdc37 chaperone machinery driven by CK2-dependent phosphorylation and PP5-dependent de-phosphorylation of Cdc37. Details are described in the text. This figure is modified from [70].

lacks the conserved N-terminal region containing the CK2-phosphorylation site. The 3D structure of Hsp90-Cdc37-Cdk4 complex determined by single-particle electron microscopy also failed to assign the N-terminal region of Cdc37 in the complex [73]. Therefore, the precise mechanism of the regulation of Cdc37 by CK2-dependent phosphorylation in molecular levels remains obscure.

A gene network analysis further supports the importance of the intimate relationship between CK2 and Cdc37. Using a genome-wide yeast synthetic gene array screening approach, 38 yeast genes were identified as having a genetic interaction with the phosphorylation site mutant of Cdc37 [75]. The list includes both α and β subunits of CK2 as well as several molecular chaperones, including yeast homologues of Hsp90, Hsp40, and Hop. In addition, the gene network analysis indicates that a group of kinase genes involved in genome integrity such as Rad53, Bub1, Cdc7, and Ssn3 have close links to the CK2-Cdc37 system [75]. Under the condition where Cdc37 activity is limited by insufficient CK2-dependent phosphorylation, these kinases might malfunction; thus, the robustness of the genome integrity system ensured by these kinases could be compromised (Fig. 5).

CK2, Cdc37, and Hsp90: A promising trinity as an anti-cancer drug target

The growth of cancer cells depends on multiple signal transduction systems that promote rapid cell division and cell cycle progression. Regulation of cell division and the cell cycle machinery is most often achieved by a variety of signaling protein kinases. Dysregulation or a mutation of a signaling protein kinase, which gives rise to abnormal stability and catalytic activity, eventually causing neoplastic cell growth. Therefore, signal-transducing protein kinases are favorable molecular targets for cancer chemotherapy. However, simply inhibiting a single protein kinase is not always effective in suppressing tumor cell growth. Cellular growth-signaling cascades constitute a complicated meshwork, and cells may escape the inhibition of a single pathway by using other signaling detours.

Hsp90 and Cdc37 are both required for activity and stability of many tumor-inducing signaling protein kinases, and tumors appear to become addicted to these chaperones [21, 46, 76–80]. The whole cancer kinome containing multiple signaling kinases can be inclusively suppressed by restraining the chaperoning function of Hsp90-Cdc37 (see Fig. 5). Attacking the Hsp90-Cdc37 chaperone system can be a more promising and effective way to inhibit tumor cell growth than targeting only one kinase. Geldanamycin, a low molecular weight benzoquinone compound, is a potent and specific inhibitor of Hsp90. Geldanamycin binds to the ATP-binding pocket of Hsp90 and inhibits the molecular chaperone activity of Hsp90 [81–83]. Treatment of cells with geldanamycin thus directs Hsp90-client proteins to proteasomal degradation, and thereby depletes client proteins from treated cells. Indeed, geldanamycin and its derivatives have been shown to suppress multiple Hsp90-dependent client protein kinases in cells and also to decrease solid tumors in animal models [14, 77, 79, 81, 83–86]. Several Hsp90 inhibitors are undergoing clinical trials for cancer chemotherapy [87, 88].

Cdc37 is increased in proliferating tissues and is heavily expressed in many clinical cancers [89]. Cdc37 is an overexpressed oncogene, and under compulsive Cdc37 expression cells become transformed and rapidly proliferate, resulting in tumor formation [90, 91]. Hsp90 and Cdc37 are both required for the cancer kinome, but Cdc37 might be the limiting component. It is predicted that the proliferation of mutation-activated kinases in tumors leads to a greater dependency on Cdc37 function. Since Cdc37 is required principally for protein kinases, a subset of Hsp90-client proteins, inhibition of Cdc37 function should specifically affect signaling protein kinases without touching most of other Hsp90-client proteins. This might be advantageous to avoid unexpected and undesirable side effects caused by general Hsp90 inhibitors. Therefore, disruption of Cdc37 activity disables many signaling protein kinases simultaneously, and is a promising new strategy for the treatment of cancer owing to its multi-targeting and specific nature, the increased expression of Cdc37 in rapidly dividing cells, and the ability of Cdc37 depletion to arrest tumor growth [92–94] (Fig. 5). Targeting Cdc37 in malignant tissues will be explored by siRNA-induced specific reduction of Cdc37 expression and pharmacological disruption of Hsp90-Cdc37-kinase interactions. In addition, modulation of CK2-dependent phosphorylation and PP5-dependent de-phosphorylation in the critical residue Ser13 within the client-interaction domain of Cdc37 will be an auspicious strategy for suppressing Cdc37 function. Although strong inhibition of CK2 might result in a pleiotropic outcome as in the case of Hsp90 inhibition, the feedback-loop mechanism of the Cdc37-CK2 couple may possibly favor specific disruption of the kinome chaperone system by modest CK2 inhibition. The important role of CK2 in tumor cell growth and a possibility of using CK2 inhibitors for cancer therapy will be precisely described elsewhere in this review series.

Concluding remarks

In this review, the critical roles of CK2 in regulation of Hsp90-mediated chaperone system are precisely described. CK2 phosphorylates and modulates Hsp90 and its co-chaperones FKBP52 and Cdc37. CK2-dependent phosphorylation of Cdc37 is essential for the chaperoning function of Hsp90-Cdc37 for multiple signaling protein kinases, indicating that CK2 is the kinase that chaperones molecular chaperones. The tumor kinome appears to become addicted to the Hsp90-Cdc37 chaperone system. Thus, targeting Hsp90, Cdc37, and CK2 is a promising strategy for cancer treatment.

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