Protein kinase CK2: A new view of an old molecular complex

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Supplementary data.

Model assumptions and equations.

In the context of cellular signaling, it has been shown that the recruitment of proteins to the cell membrane increases the number of molecular complexes made with membrane-associated substrates (Kholodenko et al., 2000) (see also Haugh, 2002 for a full account of the biophysical model.) Using a similar approach, we show here, that the interaction of CK2α with its partners, including the dimeric $CK2\beta_2$ regulatory subunit or other multi-molecular targets C, leads to a large difference in the number of complexes $CK2\alpha_2\beta_2$ and CK2αC depending on the biophysical conditions prevailing in the cell.

Let A be the CK2α subunit, B_2 the dimeric $CK2\beta_2$ regulatory subunit and C a multi-molecular complex. To reconcile *in vivo* and *in vitro* observations, we assume that:

1. interaction between CK2α and $CK2\beta_2$ subunits can take place everywhere in the cell bulk (Martel et al., 2002) (volume V_c.). At equilibrium, the law of mass action gives:

$$K = \frac{[A]^2[B_2]}{[A_2B_2]}$$

with $K \approx 29.2$ $(\text{nM})^2$.

2. CK2α can bind to the complex C only in a sub-compartment of the cell, which corresponds to the volume V_m. At equilibrium, concentrations are related to the dissociation constant K_O ('O' for 'other' CK2α partners) by:

$$K_O = \frac{[A_m][C]}{[A_m C]}$$

with K_O larger than $\sqrt{K}$. A_m represents the CK2α subunit in the sub-compartment where binding occurs.

3. the CK2α subunit is at equilibrium between the different cell compartments:

$$[A] = [A_m]$$

In cells, about 20 to 30% of the total volume is occupied by large macromolecules, which accounts for a reduction of the diffusion coefficients for proteins by factors ranging from 3 to 11 (Ellis, 2001). Therefore, we assume that the number of CK2α subunits exchanged between the bulk and the sub-compartment are negligible.

4. the total number of the different molecular species is conserved according to

$$\begin{aligned} P_T &= 2B_2 + 2A_2B_2 \\ P_{c,T} &= A + 2A_2B_2 \\ P_{m,T} &= A_m + A_m C_m \\ P_T &= P_{c,T} + P_{m,T} \\ C_T &= C + A_m C_m \end{aligned}$$

where a symbol in italics designates the number of molecules of the corresponding chemical species. The first relation codes for conservation of the CK2β_2 complex throughout the cell. Because we assume zero flux between the bulk and the sub-compartment, the total number of CK2α subunits is constant in both compartments (second and third equations.) The fourth relation ensures the conservation of the total CK2α subunits. The conservation of the complex C is given in the final equation.

To compare the relative importance of the CK2α binding to CK2β_2 or to other binding patners, we use number of molecules instead of concentration. The equilibrium relations give:

$$A_2B_2 = \frac{A^2.B_2}{(V_c)^2 K}$$

$$A_mC = \frac{A_m.C}{V_m K_O}$$

$$A_m = \frac{V_m}{V_c} A$$

Using the conservation relations and the equilibrium conditions, the number of CK2α subunits in both compartments is obtained as solution of two simultaneous equations:

$$(a) \qquad x + \frac{x^2}{\theta_1 + x^2} - \frac{V_c}{V_c + V_m} = 0$$

Eq. (1)

$$(b) \qquad y + \theta_2 \frac{y}{\theta_3 + y} - \frac{V_m}{V_c + V_m} = 0$$

where $x = \frac{A}{P_T}, y = \frac{A_m}{P_T}, \theta_1 = K\left(\frac{V_c}{P_T}\right)^2, \theta_2 = \frac{C_T}{P_T}, \theta_3 = \frac{V_m K_O}{P_T}$. Note that all the variables or parameters are in dimensionless form. The complexes normalized number of complexes A_2B_2 and A_mC are given by

$$\frac{A_2B_2}{P_T} = \frac{x^2}{\theta_1 + x^2}$$

$$\frac{A_mC}{P_T} = \theta_2 \frac{y}{\theta_3 + y}$$

where *x* and *y* are the unique solution of equation (1a,b) in the interval [0,1].

Model predictions.

The figure 3 (main text) is obtained for the following parameters: $K=29.2(\text{nM})^2$, $V_c=4.2$pl (volume of a sphere of 10μm) and $K_O = 100\sqrt{K}$; $P_T = 100\,V_c\sqrt{K}, C_T = 100\,V_c\sqrt{K}$. V_m , the volume of the sub compartment, ranges from *4.2*fl to *4.2*pl. As the ratio V_c/V_m

decreases (V_c=V_m *in vitro*), binding of CK2α to its two partners (CK2β_2 or C) favors the high affinity complex CK2$\alpha_2\beta_2$. Conversely, when CK2α binding to its low-affinity partner is confined to a restricted compartment (large V_c/V_m), equilibrium is shifted to the complex CK2αC. Equal amount of CK2$\alpha_2\beta_2$ and CK2αC (i.e. *r=1*) is obtained for $V_c/V_m \approx 30$.

Figure legend

Figure S1 shows how the ratio function, as defined above, changes with $\theta = \frac{K_O}{\sqrt{K}}$, the relative affinity of CK2α to its two partners. When θ is in the range [1,10000] and for large V_c/V_m, CK2α is mainly engaged with its low-affinity partner, whatever the value of K_O. Conversely, at low V_c/V_m, CK2$\alpha_2\beta_2$ is predominant and represents the unique observable complex. These results support our conclusions and show the robustness of the prediction illustrated in figure 3 (main text). The different curves were obtained for $K_O \approx 2.78\sqrt{K}$ (curve a) to $K_O = 10000\sqrt{K}$ (curve e); the data shown in figure 3 (main text) corresponds to curve (d) with $K_O = 100\sqrt{K}$. Other parameters are those of the figure 3 (main text).

To demonstrate the robustness of the behaviour shown in figure 3 (main text), we have determined the boundary between the domains where CK2$\alpha_2\beta_2$< CK2αC or CK2$\alpha_2\beta_2$> CK2αC by solving equation 1 supplemented with the constraint *r=1* (i.e. $A_2B_2=A_mC$) for different values of the parameters (V_c/V_m,θ). Other parameters are those of figure 3 (main text). A limited space for the low-affinity reaction, representing 10% of the total bulk volume, results into an eighty-fold apparent decrease of the low-affinity dissociation constant. A further reaction confinement (5% and below) has a more drastic effect on the apparent dissociation constant which can counterpoise the tight association between CK2α and CK2β_2.

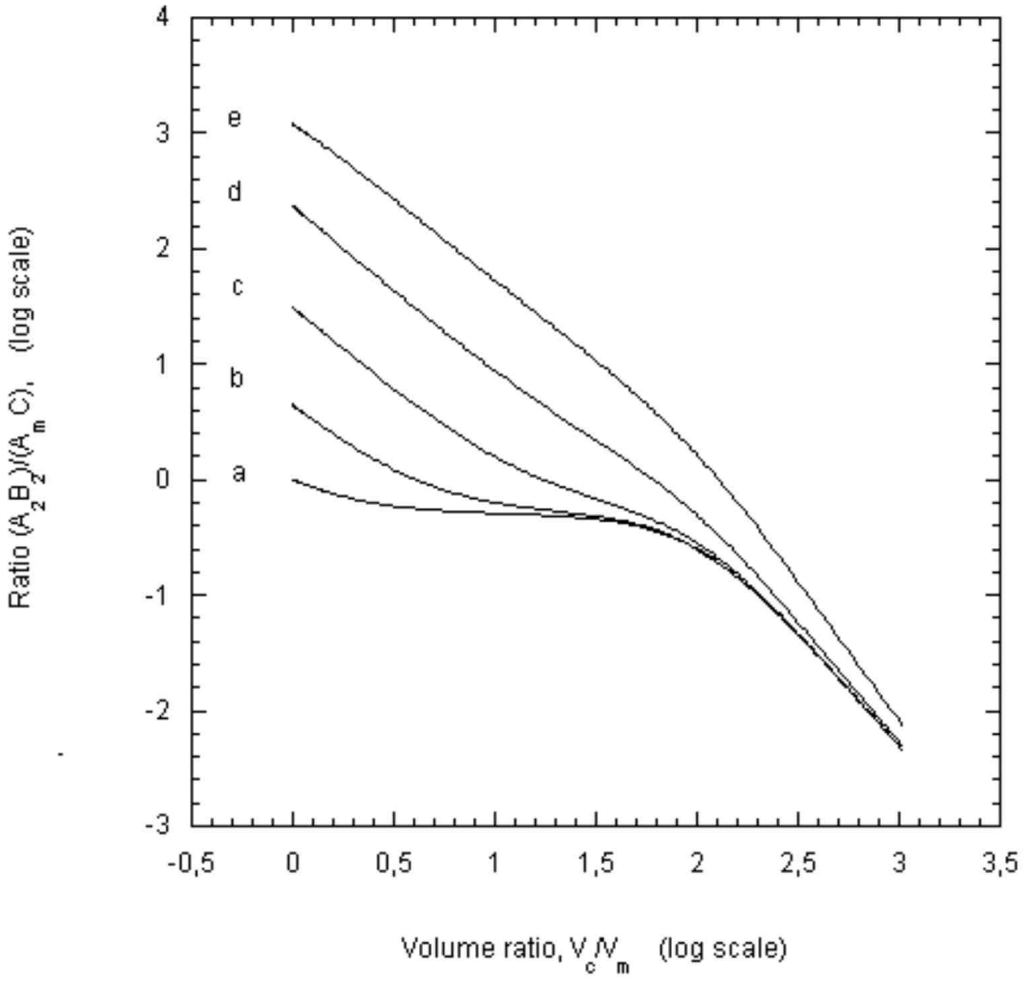
Ratio $(A_2B_2)/(A_mC)$, (log scale)
Volume ratio, V_c/V_m (log scale)
a
b
c
d
e
4
3
2
1
0
-1
-2
-3
-0,5
0
0,5
1
1,5
2
2,5
3
3,5

Figure S1

References

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