

Supplementary Materials:

Binding of *Euplotes octocarinatus* centrin to peptide from Xeroderma Pigmentosum Group C Protein (XPC)

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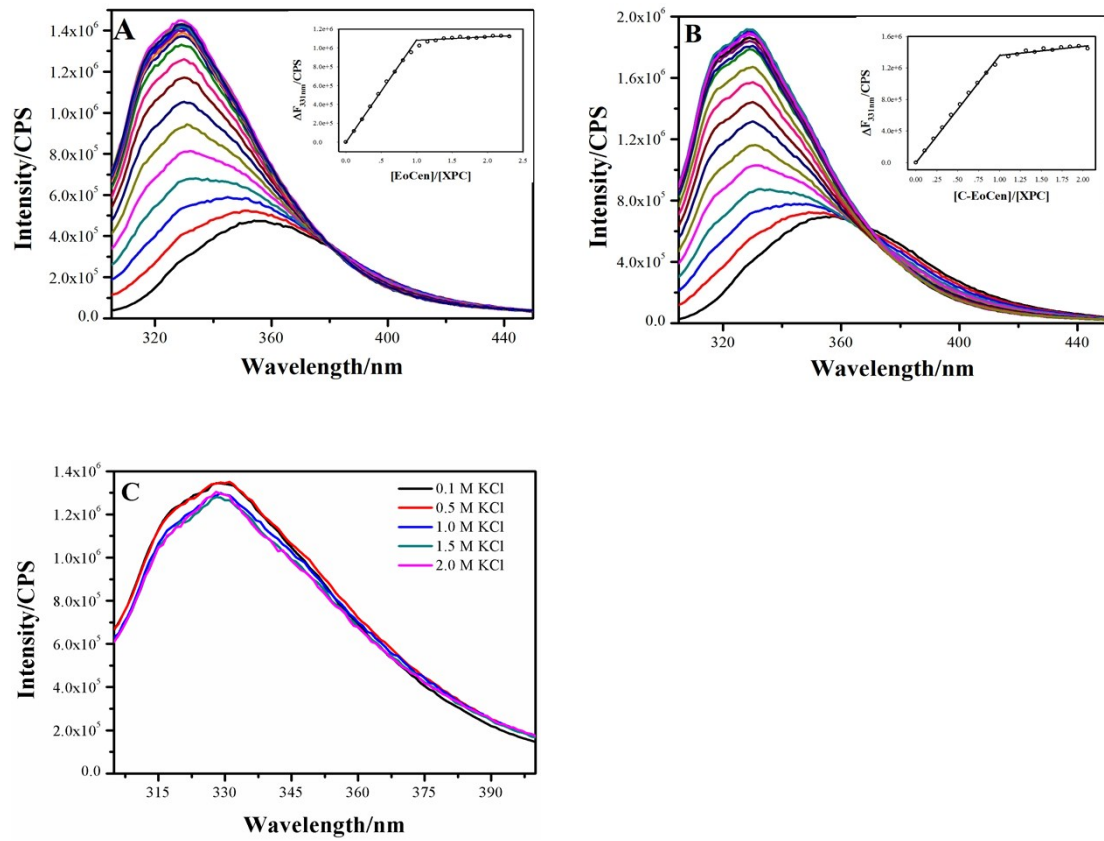


Fig. S1 Fluorescence spectra for the addition of EoCen (A) and C-EoCen(B) to the solution of XPC peptide in the presence of EDTA. Inset: $\Delta F_{331 \text{ nm}}$ vs the ratio of [protein]/[XPC]. Fluorescence intensity and maximum wavelength of complex formed by EoCen-XPC under different salt concentrations (C).

The method of calculation of the dissociation constant

In order to calculate the dissociation constant between XPC peptide and apoprotein, XPC peptide was titrated with apoproteins. The maximum emission fluorescence of the XPC peptide alone is situated at 356 nm. The new fluorescence emission at 331 nm appears after the formation of complex XPC-protein. So the increment value of fluorescence intensity at 331 nm, ΔF can be attributed to the contribution of the complex, XPC-protein. The concentrations of species for proteins and XPC peptide can be calculated by the following formulas (1)-(6):



$$K_d = \frac{[\text{protein}]_f [\text{XPC}]_f}{[\text{protein-XPC}]} \quad (2)$$

$$[\text{XPC}]_f = [\text{XPC}]_t - [\text{protein-XPC}] \quad (3)$$

$$\frac{[\text{protein-XPC}]}{[\text{XPC}]_t} = \frac{[\text{protein}]_f}{K_d + [\text{protein}]_f} \quad (4)$$

$$\frac{\Delta F}{\Delta F_{\max}} = \frac{F - F_0}{F_{\max} - F_0} = \frac{[\text{protein-XPC}]}{[\text{XPC}]_t} \quad (5)$$

$$[\text{protein}]_f = \frac{K_d \cdot [\text{protein-XPC}]}{[\text{XPC}]_t - [\text{protein-XPC}]} \quad (6)$$

Where $[\text{protein}]_t$ and $[\text{XPC}]_t$ represent the total concentration of protein and XPC peptide, respectively. $[\text{XPC}]_f$, $[\text{protein}]_f$ and $[\text{protein-XPC}]$ represent the concentration of free XPC peptide, free protein and the complex protein-XPC, respectively. F_0 , F and F_{\max} represent the fluorescence intensity of proteins in the absence of peptide, in the presence of peptide, and the saturated intensity (at the saturation concentration), at 331 nm, respectively.

Firstly, $[\text{protein}]_t$ took place of $[\text{protein}]_f$. Fit of $[\text{protein-XPC}]/[\text{XPC}]_t$ vs $[\text{protein}]_t$ was performed using SigmaPlot 10.0 software to a single-site binding model. Secondly, according to K_d and equation (6), $[\text{protein}]_f$ was calculated. Then fit of $[\text{protein-XPC}]/[\text{XPC}]_t$ vs $[\text{protein}]_f$ was performed again by using the obtained $[\text{protein}]_f$. The rest could be done in the same manner until the K_d value approached the approximate value for the n th time and the $(n+1)$ th time. That is so-called iteration method.

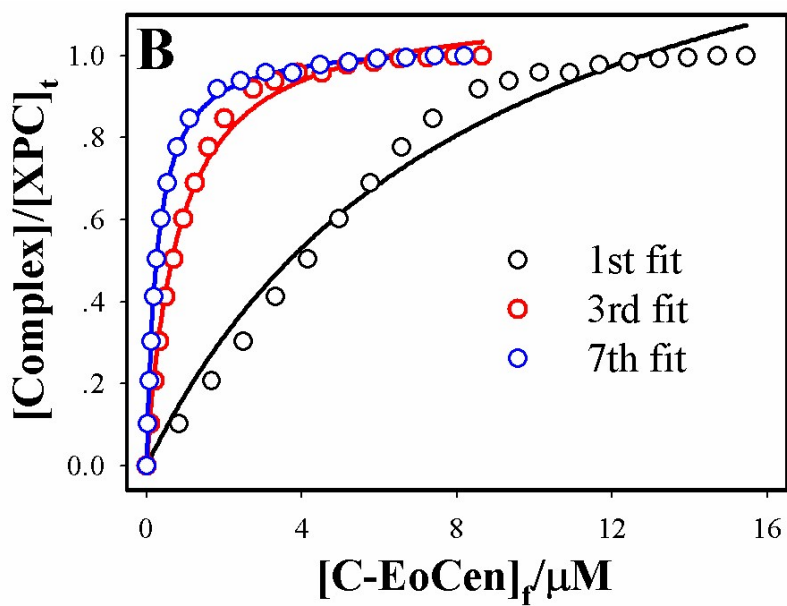
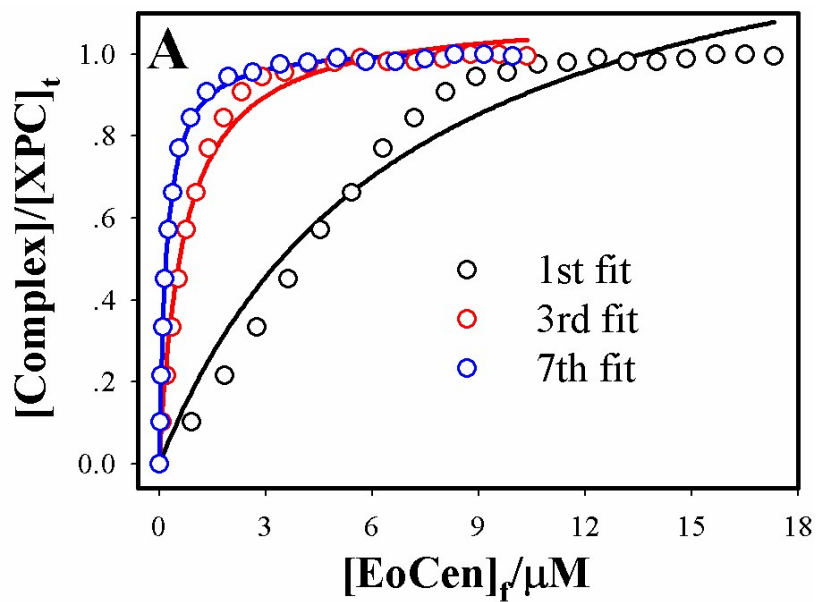


Fig. S2 Fit of $[\text{protein-XPC}]/[\text{XPC}]_t$ as a function of free concentration of EoCen (A) and C-EoCen(B) to a single-site binding model using iteration method by Sigma Plot 10.0.

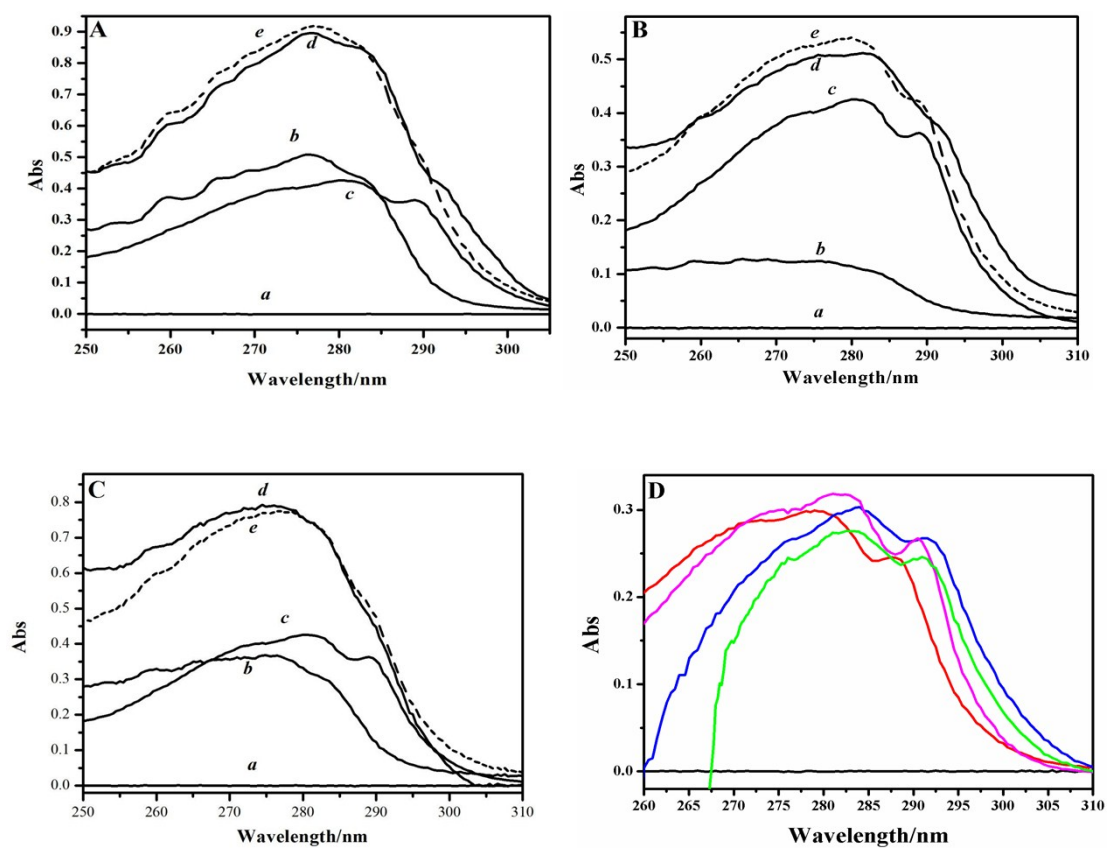


Fig. S3. UV spectra of interaction between XPC peptide and WT-EoCen (A), C-EoCen (B), N-EoCen (C), respectively. (a) Hepes, (b) apoprotein, (c) XPC, (d) 1:1 complex of apoprotein with XPC peptide, (e) the calculated sum spectra of same concentration of apoprotein and XPC peptide. (D) UV spectra of Tryptophan in different solvent: H₂O (red line), DMSO (blue line), DMF (green line), dioxane (magenta line).