



|   |                      |                                    |
|---|----------------------|------------------------------------|
| □ | Unphosphorylated CKβ | $V_{\max}$ : $14.28 \pm 0.21$ U/mg |
|   |                      | $K_m$ : $0.26 \pm 0.015$ mM        |
| ■ | Phosphorylated CKβ   | $V_{\max}$ : $28.69 \pm 0.28$ U/mg |
|   |                      | $K_m$ : $0.55 \pm 0.016$ mM        |
| ◇ | S39D/S40D CKβ        | $V_{\max}$ : $28.39 \pm 0.40$ U/mg |
|   |                      | $K_m$ : $0.57 \pm 0.024$ mM        |

**S3 Fig. Effect of the phosphorylation mimic double-mutation on the catalytic activity of CKβ with ethanolamine as substrate, at constant ATP concentration (2 mM). Each data point represents the average of three independent measurements.**