Supplemental Figure S2

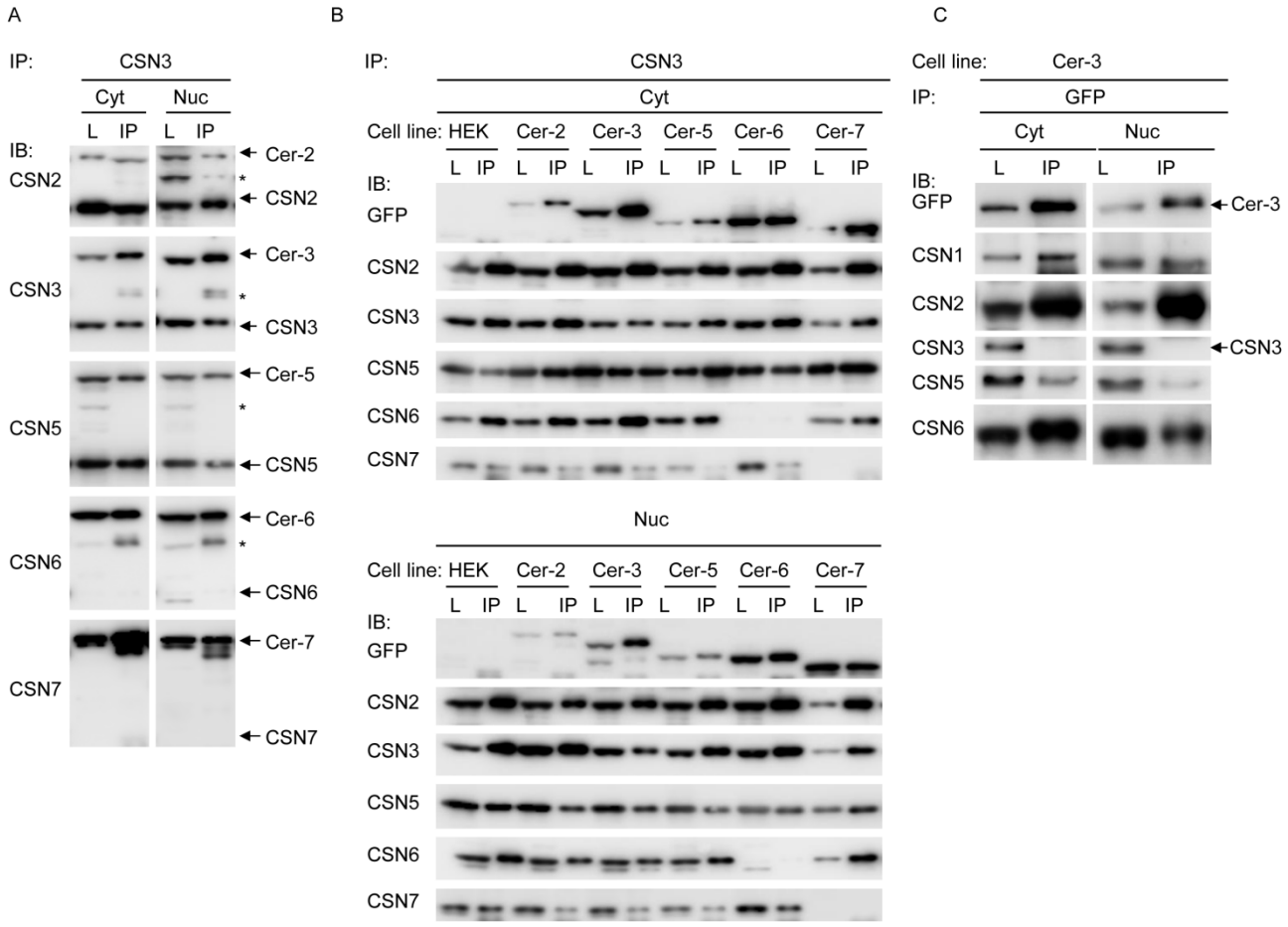

Fig S2. Fluorescent CSN subunits are integrated into the CSN complex. (A, B) Cellular proteins, extracted from the different fluorescently tagged cell lines were separated into cytosolic (Cyt) and nuclear (Nuc) fractions, and immunoprecipitated using a CSN3 antibody. Lysates from the different fractions (L) were run side-by-side with their corresponding CSN3-immunoprecipitated proteins (IP), and visualized using various antibodies, as indicated (IB). In each cell line, the incorporation of the fluorescent subunit within the endogenous complex was probed with an antibody directed against the tagged subunit, which recognized both forms. Asterisk denotes non-specific labeling (A). Immunoblotting with anti GFP and various CSN antibodies indicated that the fluorescent tag did not interfere with the assembly of the CSN complex (B). To further verify that Cer-CSN3 is incorporated into the endogenous complex, and was not pulled down independently by the CSN3 antibody,

13 cytosolic (Cyt) and nuclear (Nuc) fractions from the Cer-CSN3 cell line were immunoprecipitated
14 using an anti-GFP antibody. The antibody co-purified Cer-CSN3 together with the other CSN
15 subunits, except for endogenous CSN3, confirming the stoichiometric assembly of subunits within
16 the complex (C).

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