Supplemental figures

S1A) The different cell types were transfected with TOPflash or FOPflash and Renilla control vectors and were either left unstimulated or stimulated with rWnt-5a. The relative luciferase units (RLU) and the subsequent ratio of TOP/FOP were calculated for each sample.

S1B) Time kinetics of treatment with the CKI inhibitor indicates that the total levels of E-cadherin or β−catenin protein were not reduced upon addition of the CKI inhibitor at the times used in the immuno-coprecipitation experiments.

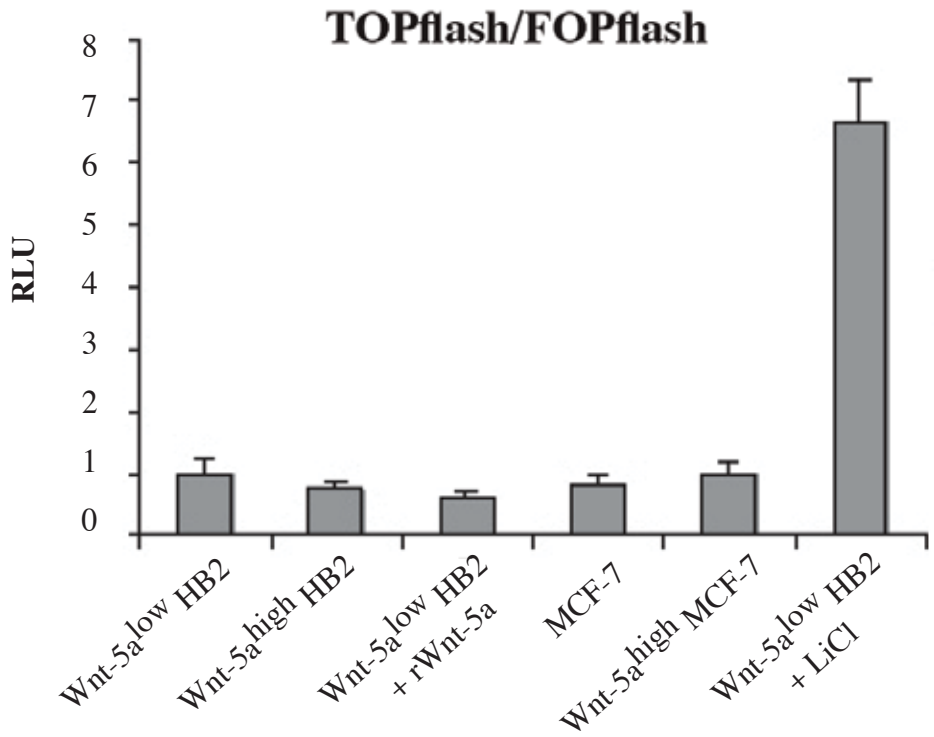
S1C) The reverse experiments, immunoprecipitation of E-cadherin and coprecipitation of β-catenin, show a 1:1 ratio of β-catenin/E-cadherin but the levels of coprecipitated Ser45 phosphorylated β-catenin is increased upon Wnt-5a stimulation.

S1D) Immuno-coprecipitation of CKIα and β-catenin with Axin. Both proteins show enhanced binding with Axin upon Wnt-5a stimulation. The blots were reprobed with Axin specific antibodies.

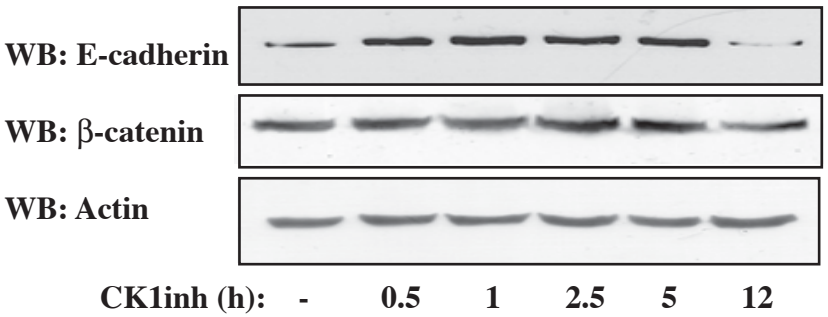
S1E) Immuno-coprecipitation of β-catenin and E-cadherin with α-catenin. β-catenin and E-cadherin show enhanced binding to α-catenin upon Wnt-5a stimulation. The blots were reprobed with α-catenin specific antibodies.

S2A) Statistics of the Wnt-5a/CKI induced β-catenin/E-cadherin complex formation in two different breast cancer cell lines MCF-7 and 4T1. The blots shown in Fig. 6 D, are representative of several separate experiments and OD measurements of the band intensities were performed to quantify the differences. Error bars indicate SEM, n=8, $p<0.05$ *, $p<0.01$**.

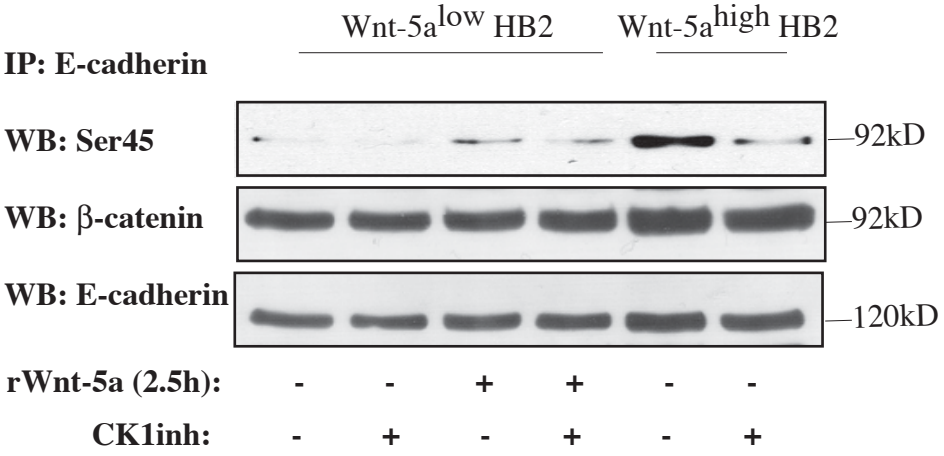
A


TOPflash/FOPflash
RLU
8
7
6
5
4
3
2
1
0
Wnt-5a low HB2
Wnt-5a high HB2
Wnt-5a low HB2 + rWnt-5a
MCF-7
Wnt-5a high MCF-7
Wnt-5a low HB2 + LiCl

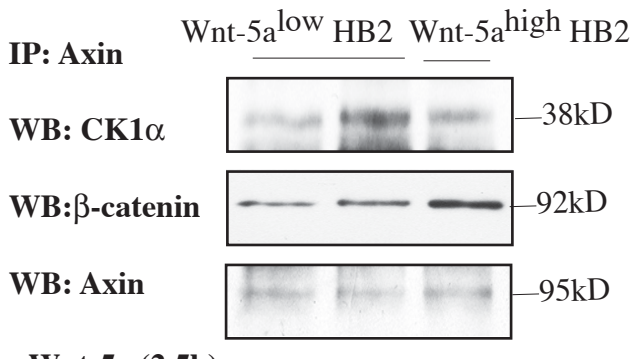
B


WB: E-cadherin
WB: β-catenin
WB: Actin
CK1inh (h): - 0.5 1 2.5 5 12

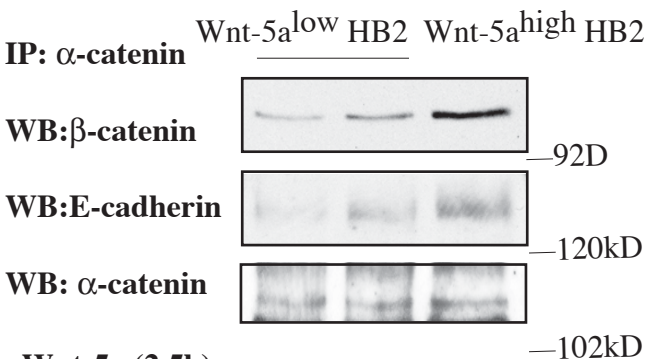
C


Wnt-5a low HB2
Wnt-5a high HB2
IP: E-cadherin
WB: Ser45
WB: β-catenin
WB: E-cadherin
92kD
92kD
120kD
rWnt-5a (2.5h): - - + + - -
CK1inh: - + - + - +

D


Wnt-5a low HB2
Wnt-5a high HB2
IP: Axin
WB: CK1α
WB:β-catenin
WB: Axin
38kD
92kD
95kD
rWnt-5a (2.5h): - + -

E


Wnt-5a low HB2
Wnt-5a high HB2
IP: α-catenin
WB:β-catenin
WB:E-cadherin
WB: α-catenin
92D
120kD
102kD
rWnt-5a (2.5h): - + -

Supplementary 2

A