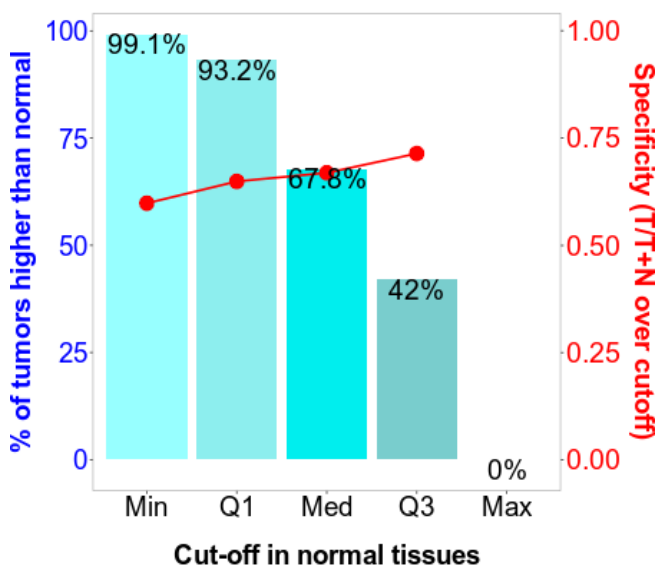


## Supplemental data

### CARMA3 promotes colorectal cancer cell motility and cancer stemness via YAP-mediated NF- $\kappa$ B activation

Ting-Yu Chan, Cheng-Tien Wu, Meei-Ling Sheu, Rong-Sen Yang and Shing-Hwa Liu

#### Supplementary File S1



**Supplementary Figure S1.** The sensitivity/specificity plot of CARD10 gene expression in colon cancer when compared tumor and normal RNA-sequencing data, which were downloaded from TNMplot (TNMplot.com: A Web Tool for the Comparison of Gene Expression in Normal, Tumor and Metastatic Tissues; <https://tnmplot.com/analysis/>). The bars represent the sensitivity: percentage of tumors which CARD10 gene expression is higher than normal samples (blue words). The specificity is shown as red words and red line.

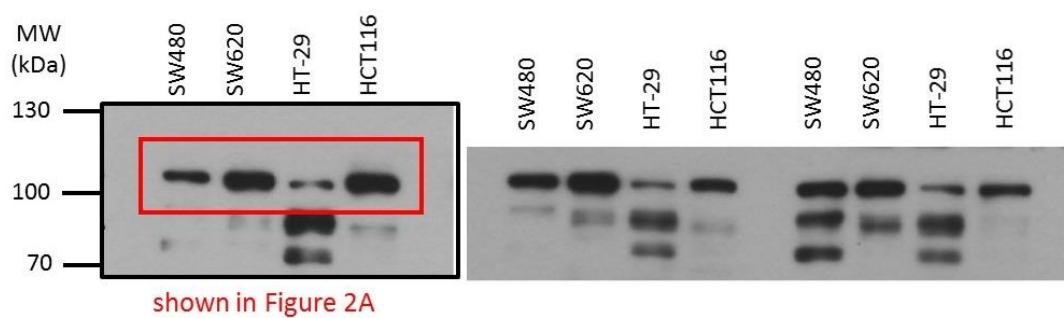
## Supplementary File S2

### Western blot raw data

#### 1. Figure S2A

Cell: SW480, SW620, HT-29, HCT116 cells

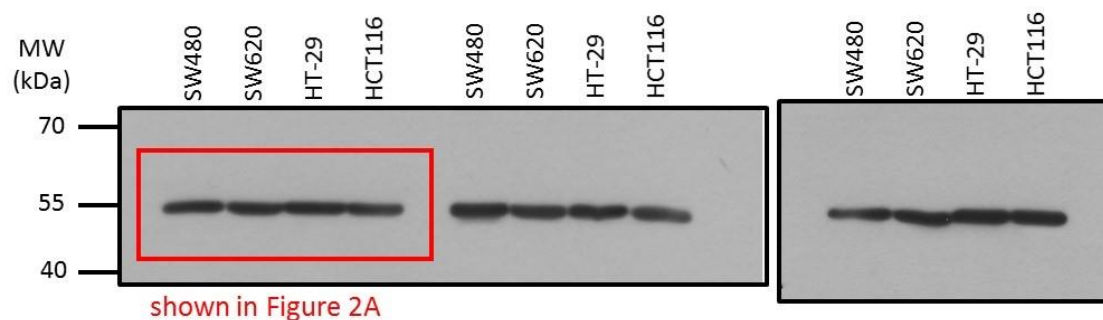
Protein: CARMA3 (116 kDa)



#### 2. Figure S2A

Cell: SW480, SW620, HT-29, HCT116 cells

Protein:  $\alpha$ -Tubulin (55 kDa)

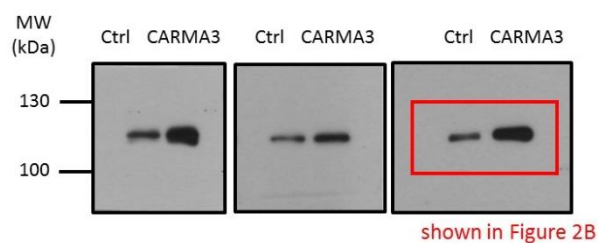


#### 3. Figure S2B-a

Overexpression of CARMA3 by plasmid transfection

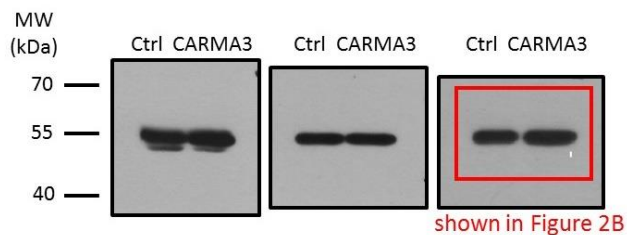
Cell: SW480 cell

Protein: CARMA3 (116 kDa)



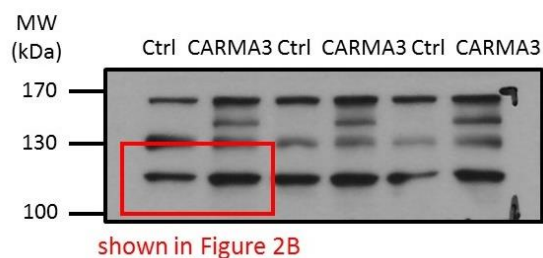
#### 4. Figure S2B-a

Overexpression of CARMA3 by plasmid transfection  
 Cell: SW480 cell  
 Protein:  $\alpha$ -Tubulin (55 kDa)



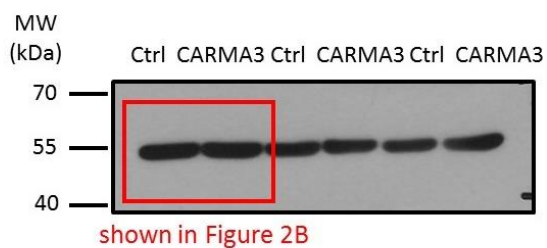
### 5. Figure S2B-a

Overexpression of CARMA3 by plasmid transfection  
 Cell: HT-29 cells  
 Protein: CARMA3 (116 kDa)



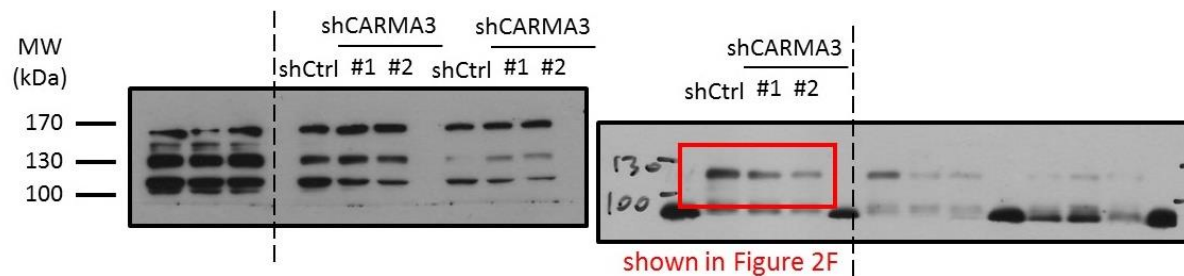
### 6. Figure S2B-b

Overexpression of CARMA3 by plasmid transfection  
 Cell: HT-29 cell  
 Protein:  $\alpha$ -Tubulin (55 kDa)



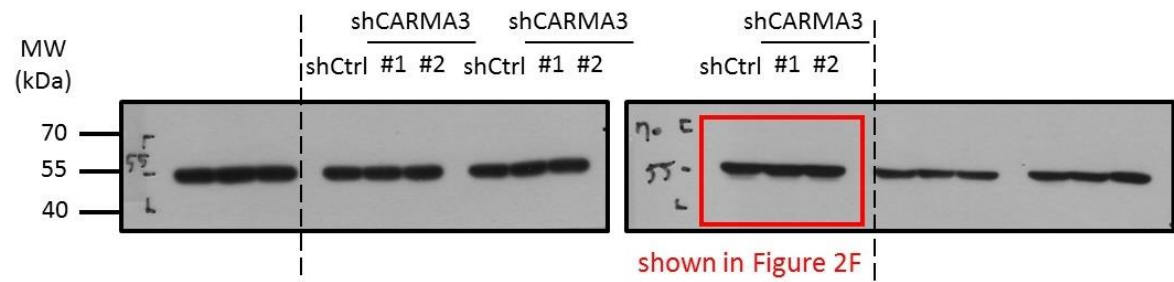
### 7. Figure S2F-a

Knockdown of CARMA3 by shRNA lentivirus infection  
 Cell: SW620 cell  
 Protein: CARMA3 (116 kDa)



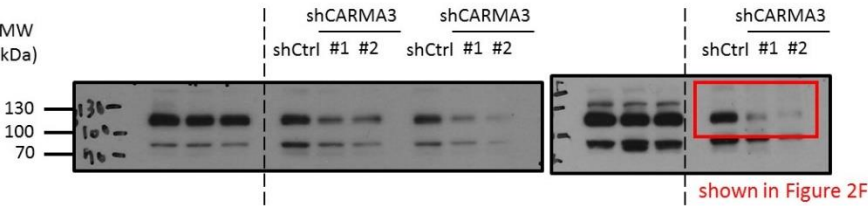
### 8. Figure S2F-a

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: SW620 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



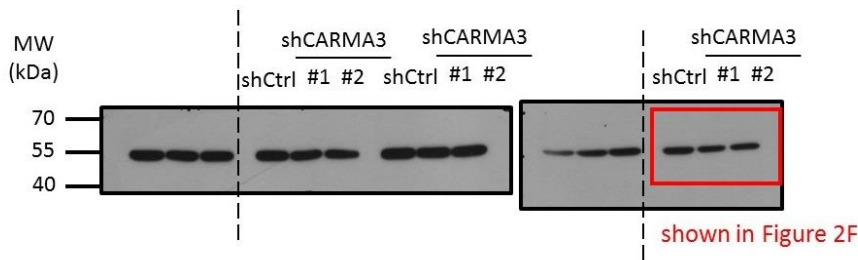
9. Figure S2F-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: CARMA3 (116 kDa)



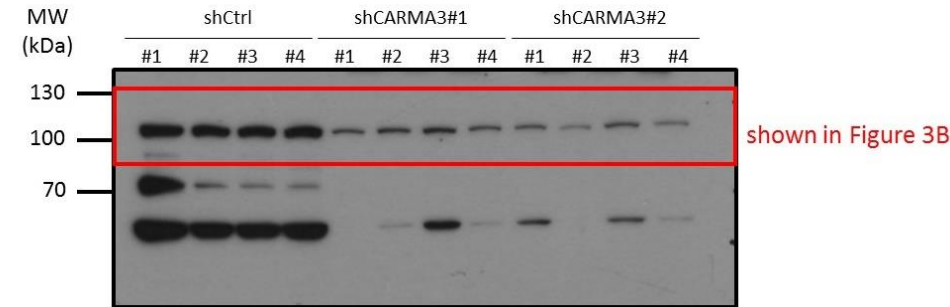
10. Figure S2F-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



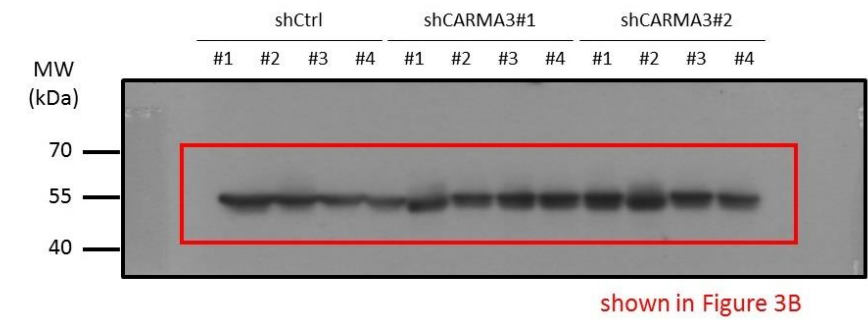
11. Figure S3B

Animal model  
Knockdown of CARMA3 in HCT116 cells by shRNA lentivirus infection  
Tumor tissues  
Protein: CARMA3 (116 kDa)



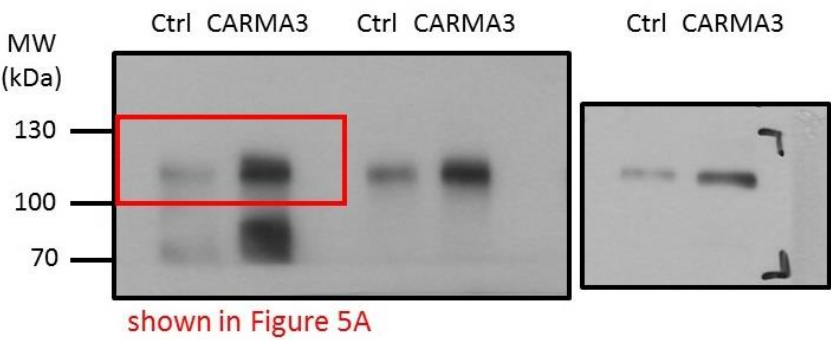
12. Figure S3B

Animal model  
Knockdown of CARMA3 in HCT116 cells by shRNA lentivirus infection  
Tumor tissues  
Protein:  $\alpha$ -Tubulin (55 kDa)



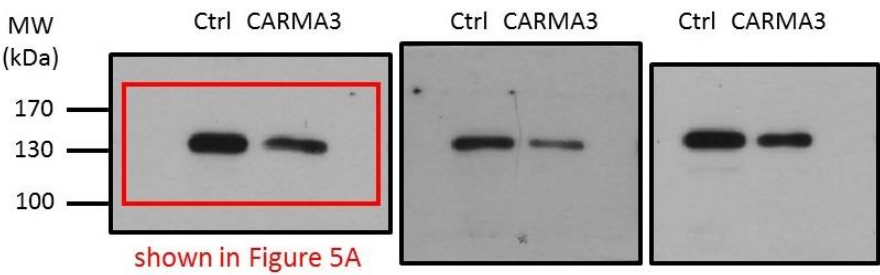
13. Figure S5A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: CARMA3 (116 kDa)



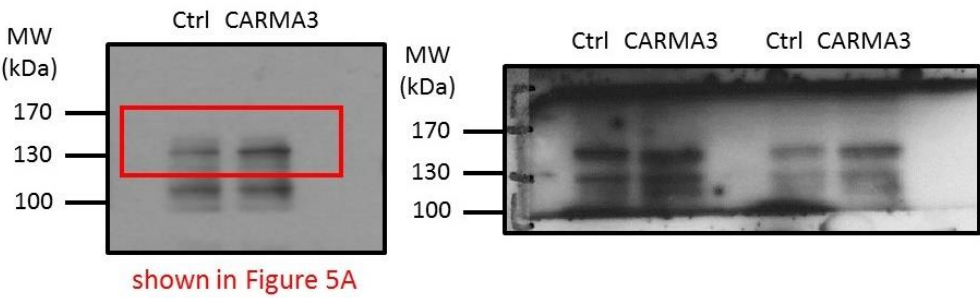
14. Figure S5A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: E-cadherin (135 kDa)



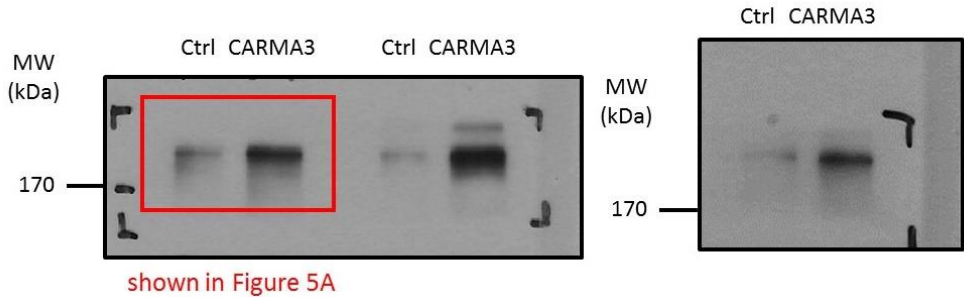
15. Figure S5A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: N-cadherin (140 kDa)



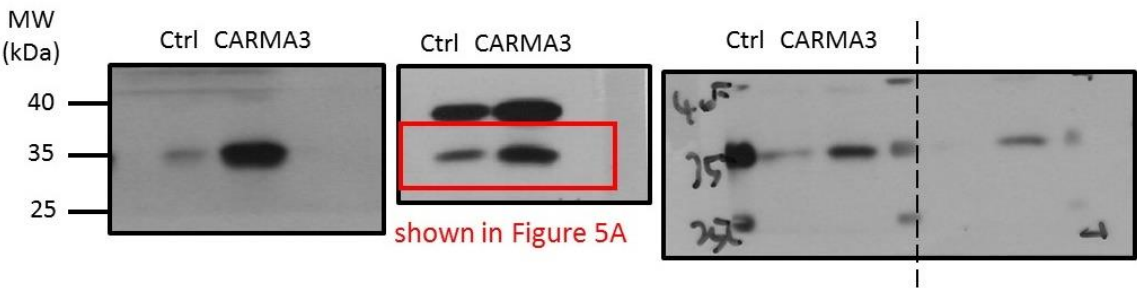
16. Figure S5A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: Fibronectin (240 kDa)



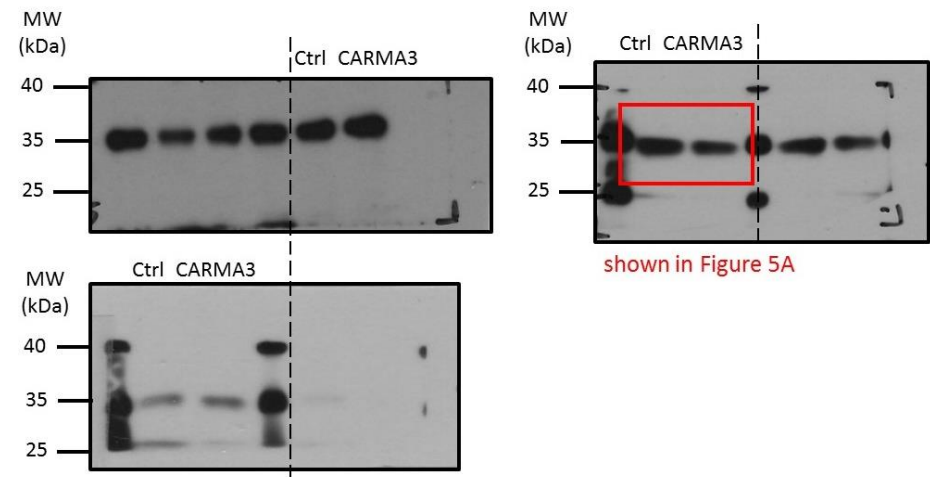
17. Figure S5A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: Slug (30 kDa)



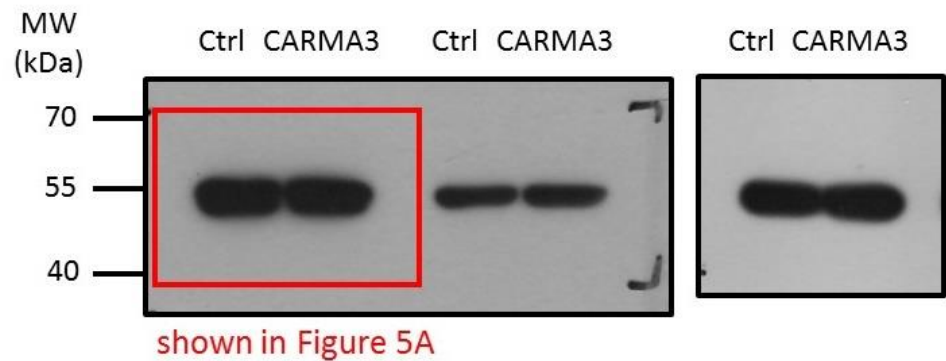
18. Figure S5A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: Snail (29 kDa)



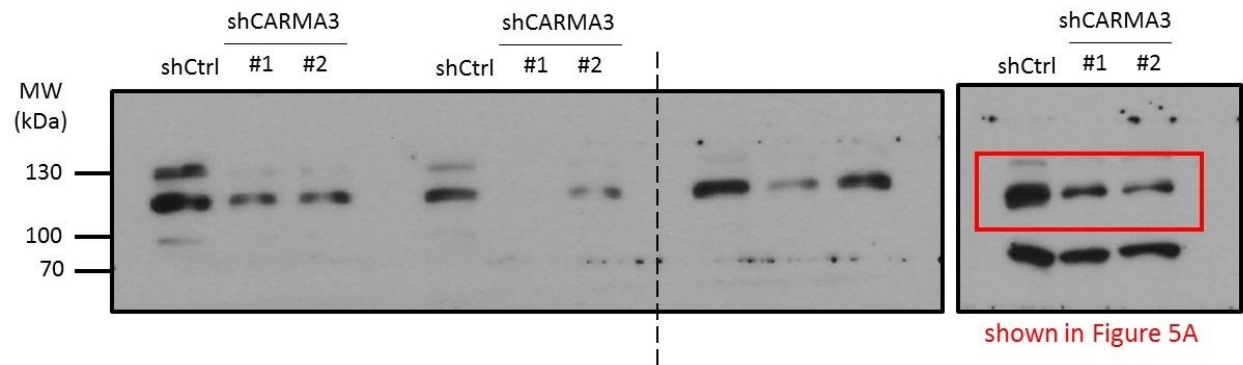
19. Figure S5A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



20. Figure S5A-b

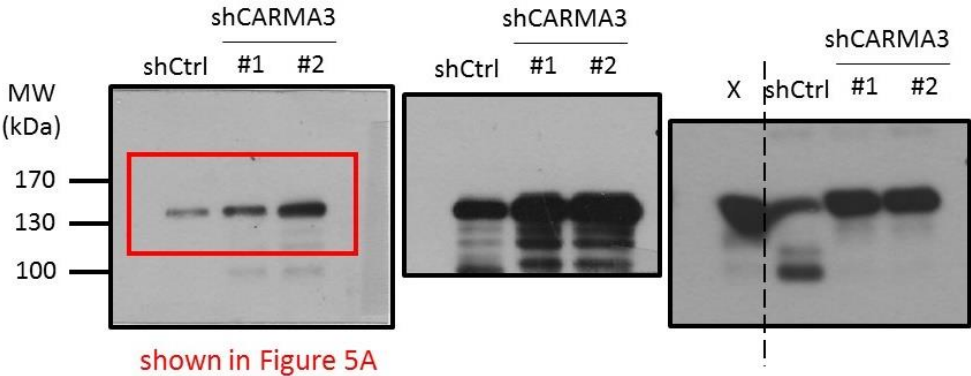
Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: CARMA3 (116 kDa)



21. Figure S5A-b

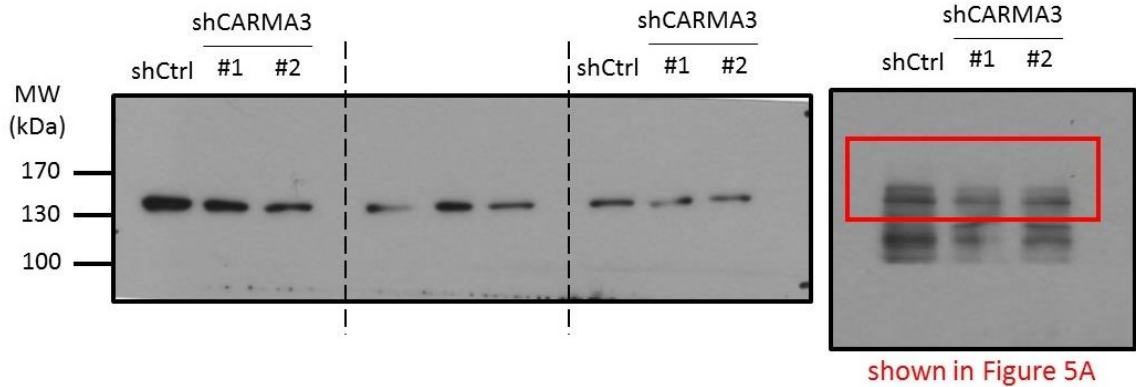


Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: E-cadherin (135 kDa)



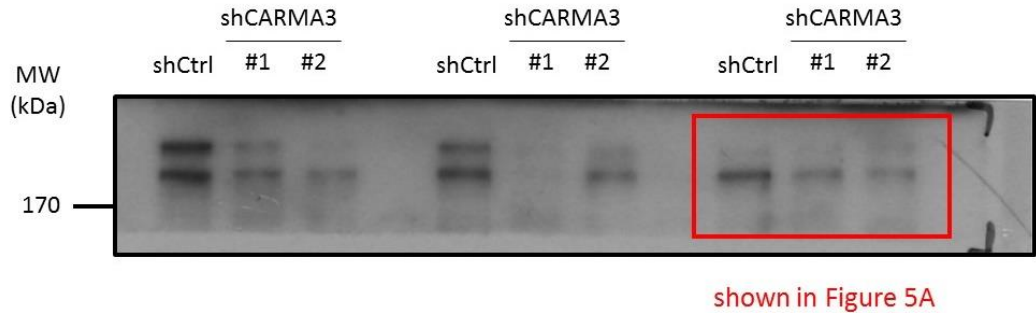
22. Figure S5A-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: N-cadherin (140 kDa)



23. Figure S5A-b

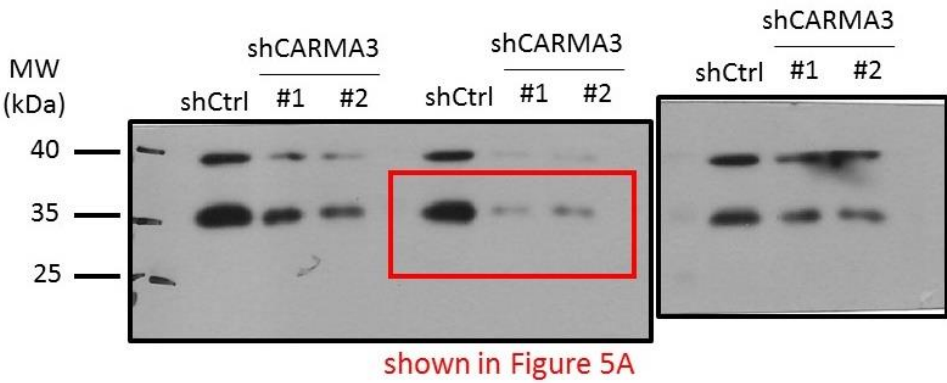
Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: Fibronectin (240 kDa)



24. Figure S5A-b

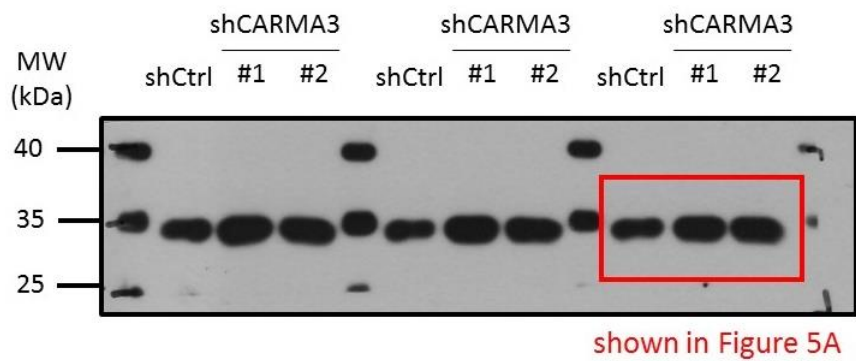


Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: Slug (30 kDa)



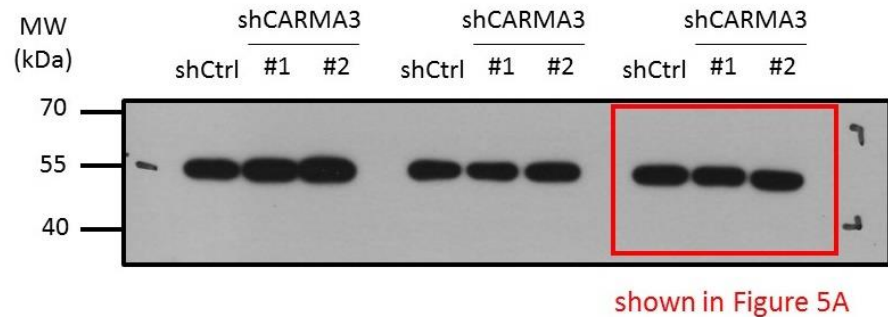
25. Figure S5A-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: Snail (29 kDa)



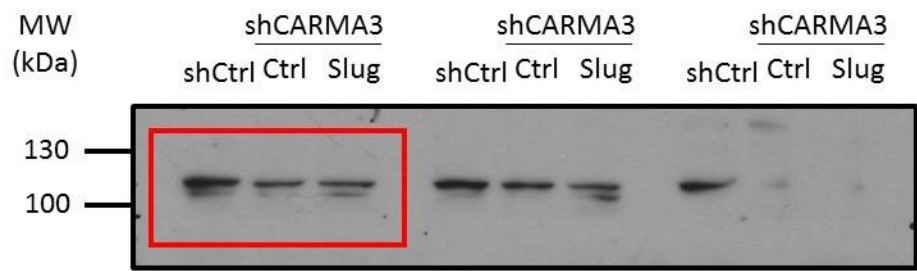
26. Figure S5A-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



27. Figure S5C

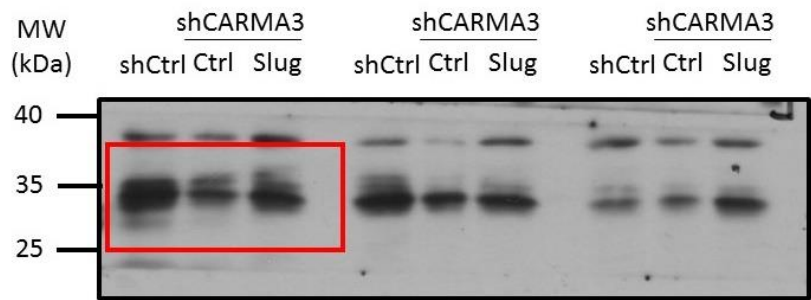
Re-overexpression of Slug in CARMA3-knocked down cells by plasmid transfection  
Cell: HCT116 cell  
Protein: CARMA3 (116 kDa)



shown in Figure 5C

28. Figure S5C

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: Slug (30 kDa)



shown in Figure 5C

29. Figure S5C

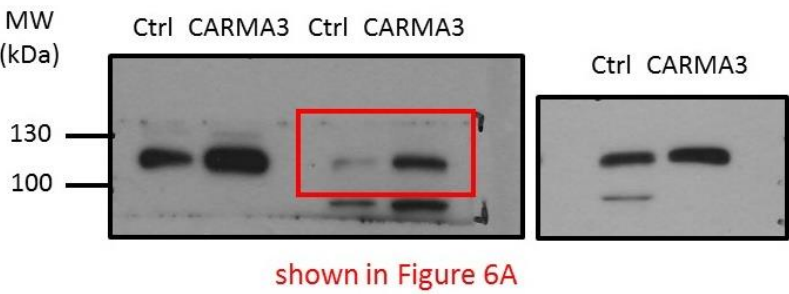
Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: α-Tubulin (55 kDa)



shown in Figure 5C

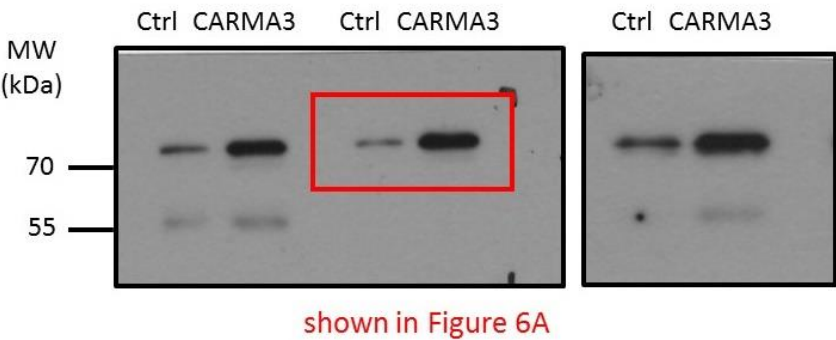
30. Figure S6A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: CARMA3 (116 kDa)



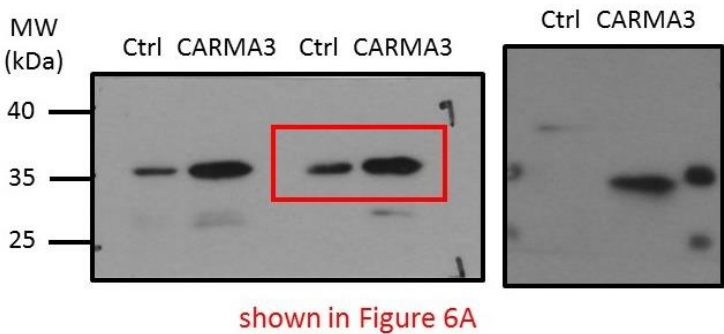
31. Figure S6A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: YAP (65-78 kDa)



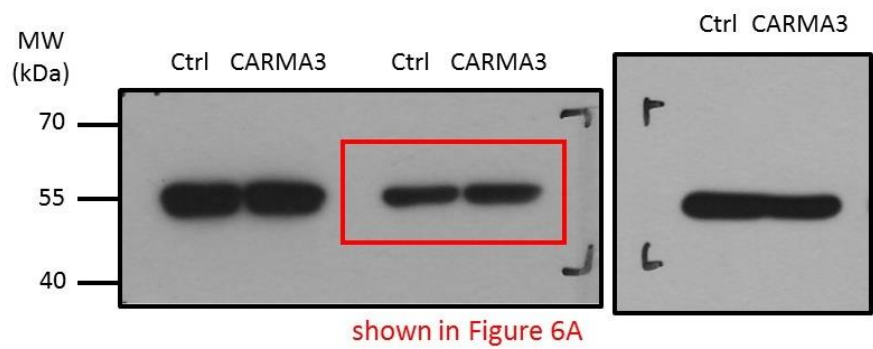
32. Figure S6A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein: Slug (30 kDa)



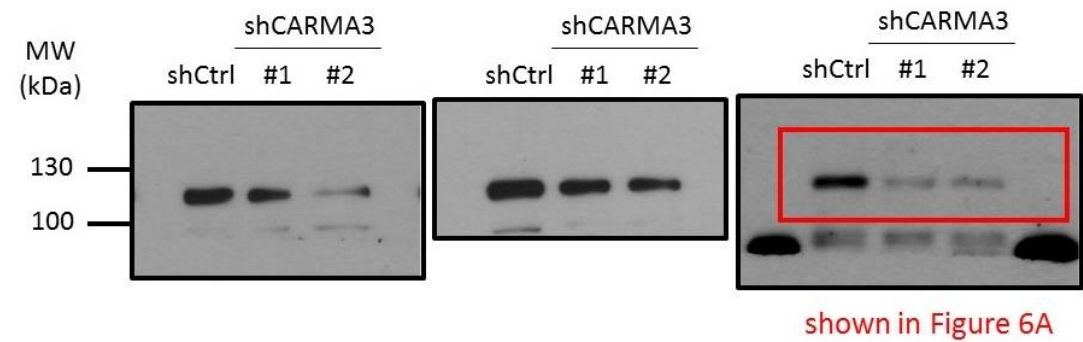
33. Figure S6A-a

Overexpression of CARMA3 by plasmid transfection  
Cell: SW480 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



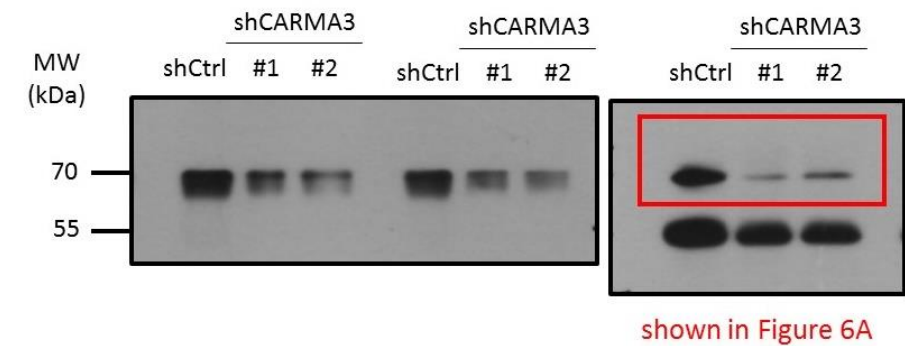
34. Figure S6A-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: CARMA3 (116 kDa)



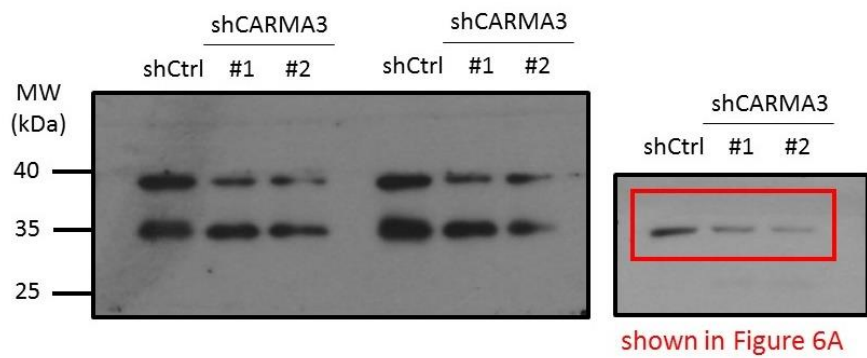
35. Figure S6A-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: YAP (65-78 kDa)



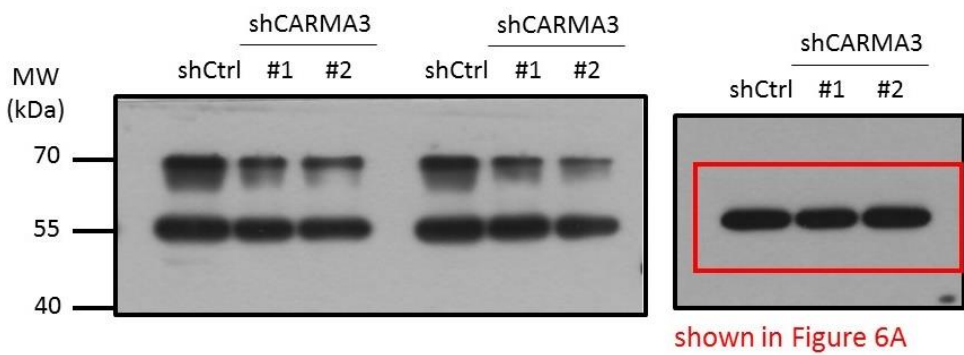
36. Figure S6A-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein: Slug (30 kDa)



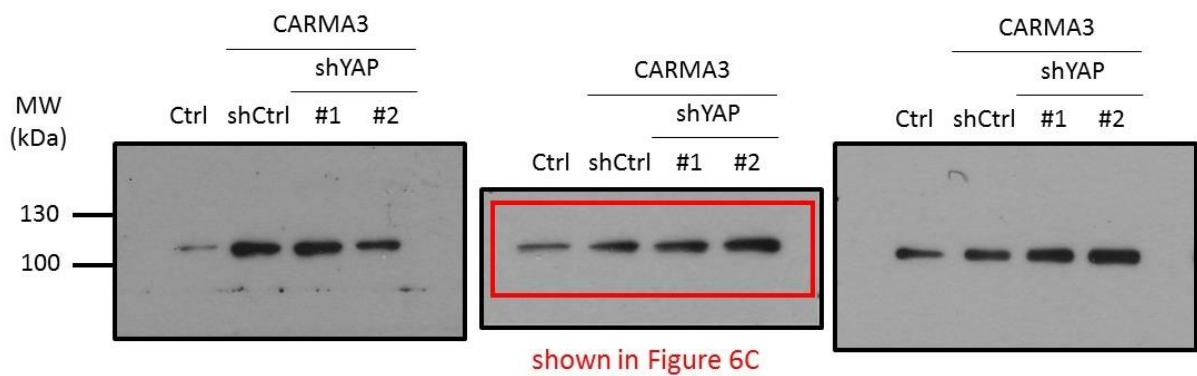
37. Figure S6A-b

Knockdown of CARMA3 by shRNA lentivirus infection  
Cell: HCT116 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



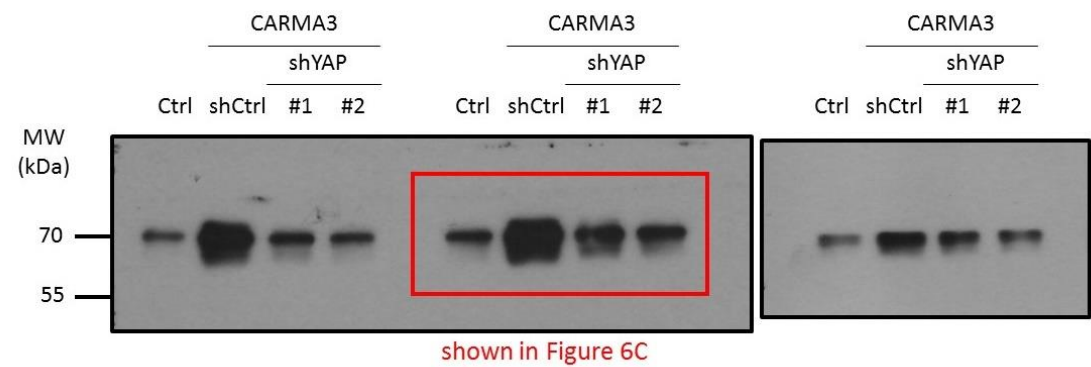
38. Figure S6C-a

Knockdown of YAP by shRNA lentivirus infection in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: CARMA3 (116 kDa)



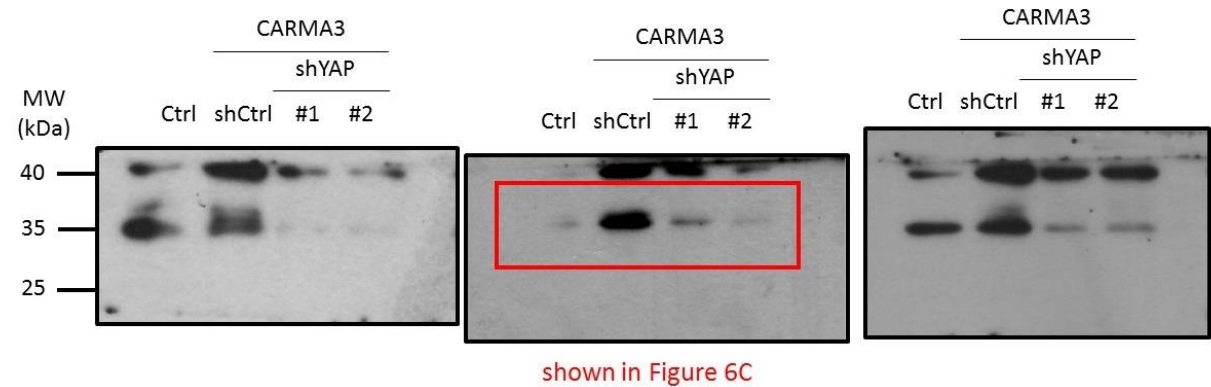
39. Figure S6C-a

Knockdown of YAP by shRNA lentivirus infection in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: YAP (65-78 kDa)



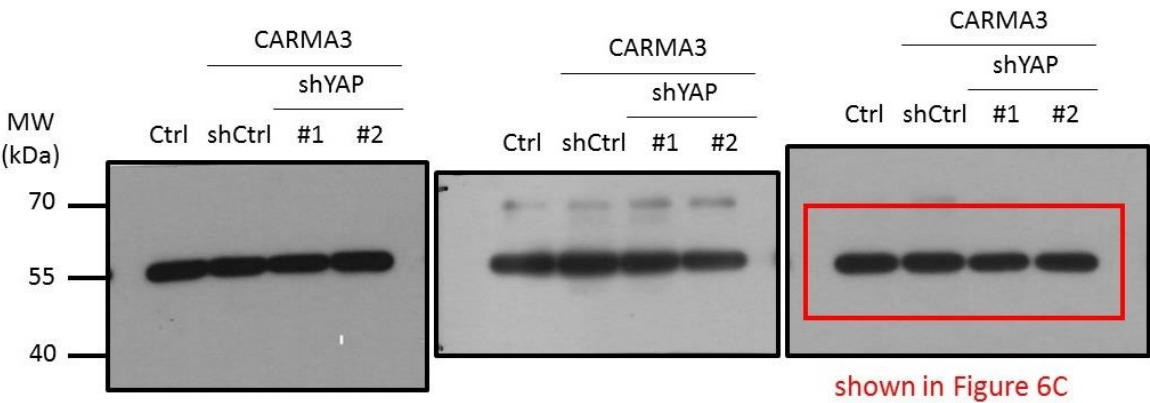
40. Figure S6C-a

Knockdown of YAP by shRNA lentivirus infection in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: Slug (30 kDa)



41. Figure S6C-a

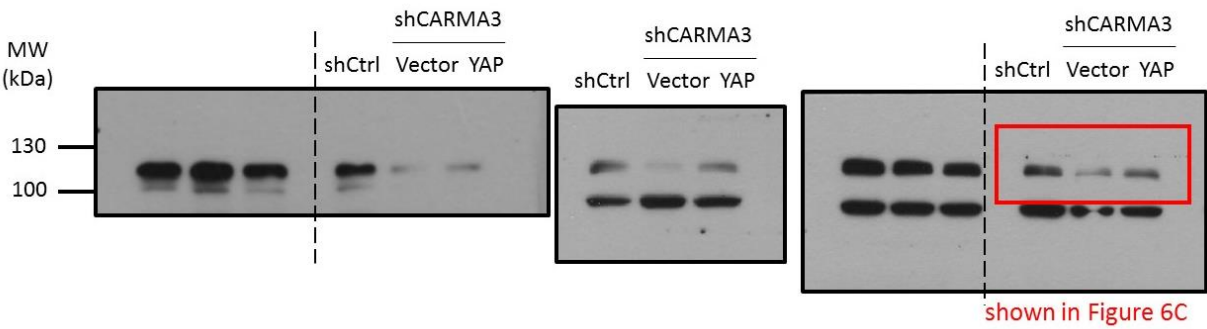
Knockdown of YAP by shRNA lentivirus infection in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



42. Figure S6C-b

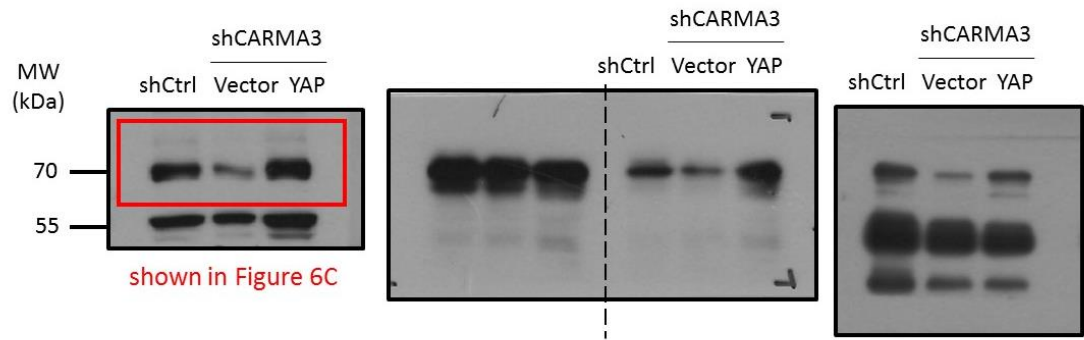


Overexpression of YAP by plasmid transfection in CARMA3-silenced cells  
Cell: HCT116 cell  
Protein: CARMA3 (116 kDa)



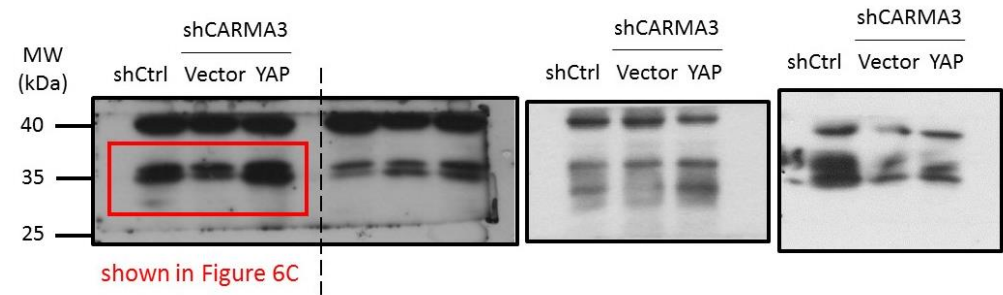
43. Figure S6C-b

Overexpression of YAP by plasmid transfection in CARMA3-silenced cells  
Cell: HCT116 cell  
Protein: YAP (65-78 kDa)



44. Figure S6C-b

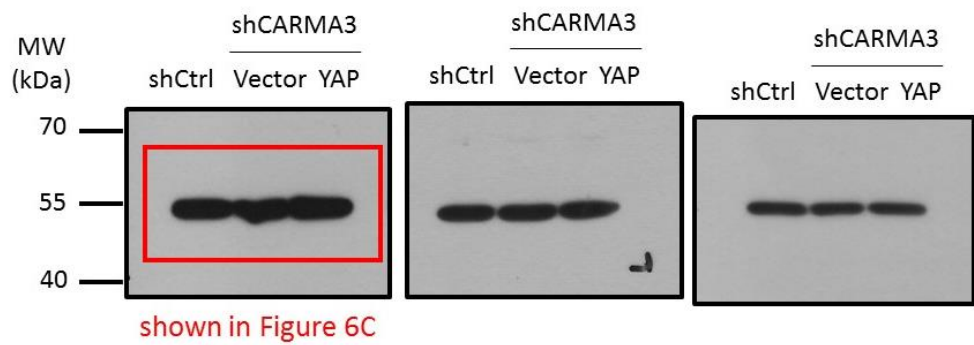
Overexpression of YAP by plasmid transfection in CARMA3-silenced cells  
Cell: HCT116 cell  
Protein: Slug (30 kDa)



45. Figure S6C-b

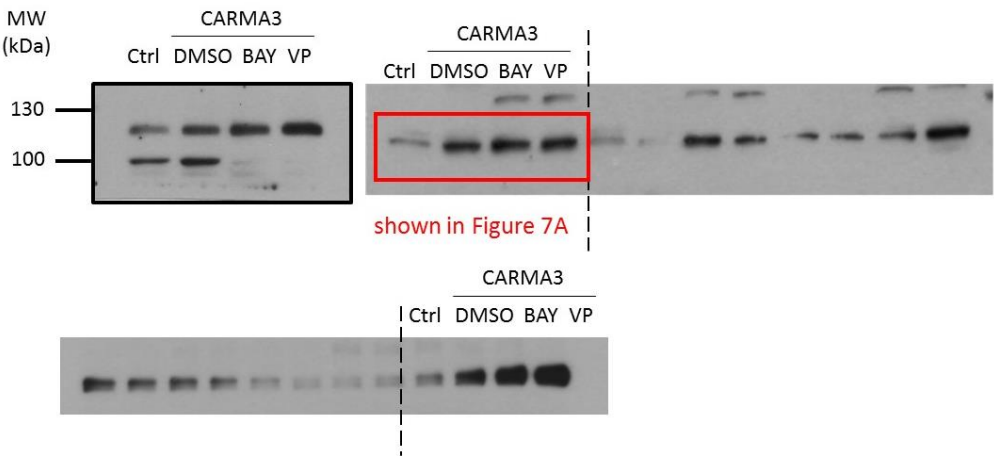


Overexpression of YAP by plasmid transfection in CARMA3-silenced cells  
Cell: HCT116 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



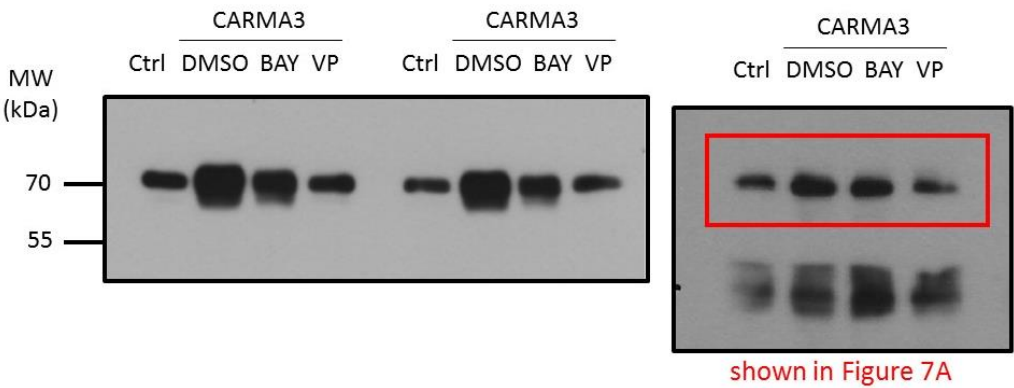
46. Figure S7A

Treatment with BAY or VP in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: CARMA3 (116 kDa)



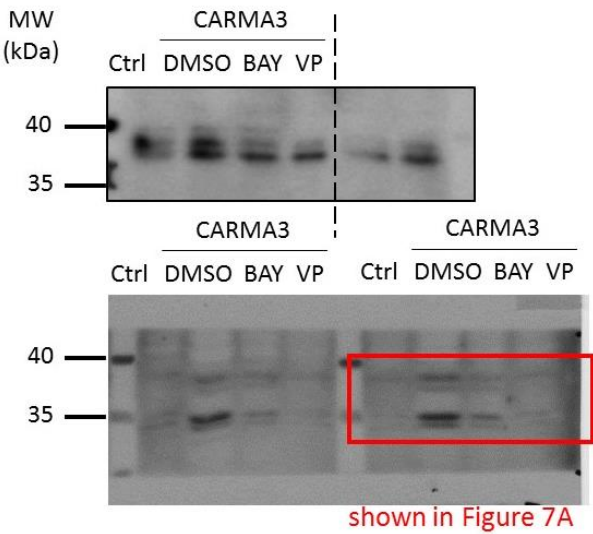
47. Figure S7A

Treatment with BAY or VP in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: YAP (65-78 kDa)



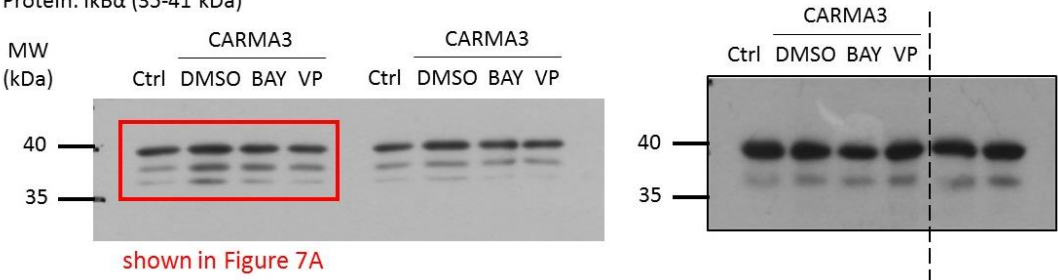
48. Figure S7A

Treatment with BAY or VP in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: p-IkB $\alpha$  (40 kDa)



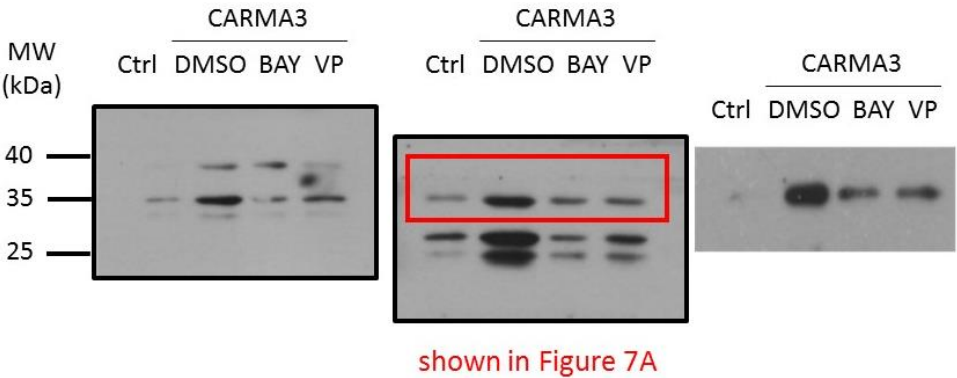
49. Figure S7A

Treatment with BAY or VP in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: IkB $\alpha$  (35-41 kDa)



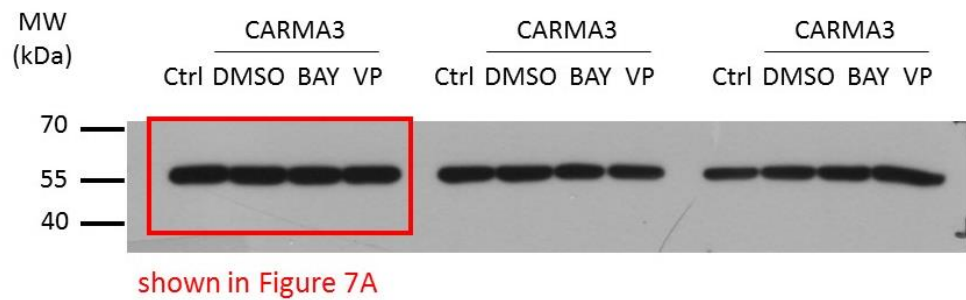
50. Figure S7A

Treatment with BAY or VP in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: Slug (30 kDa)



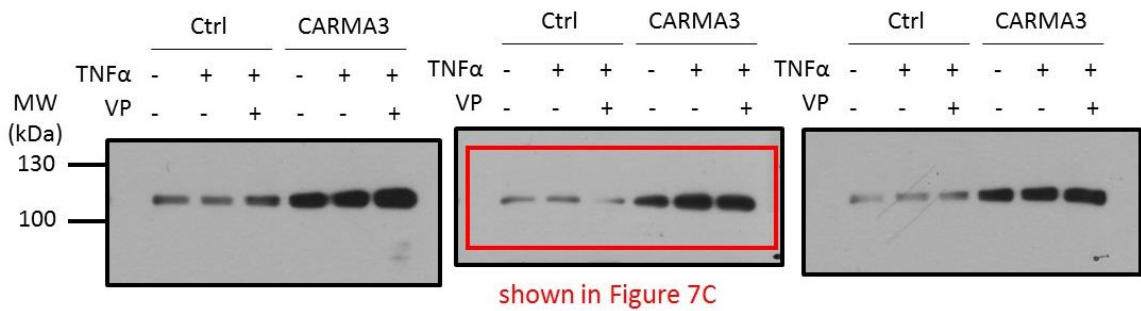
51. Figure S7A

Treatment with BAY or VP in CARMA3-overexpressed cells  
 Cell: SW480 cell  
 Protein:  $\alpha$ -Tubulin (55 kDa)



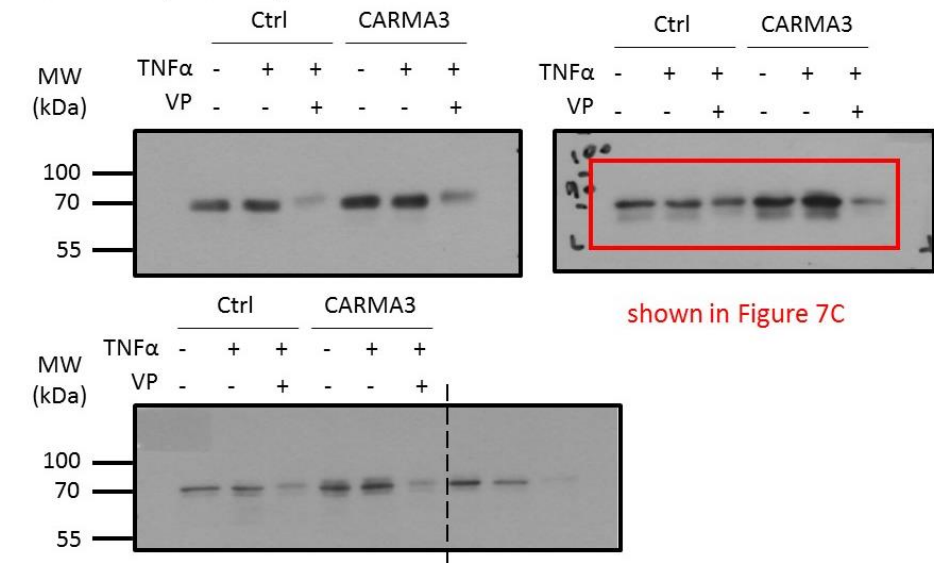
52. Figure S7C

Treatment with TNF $\alpha$  or VP in CARMA3-overexpressed cells  
 Cell: SW480 cell  
 Protein: CARMA3 (116 kDa)



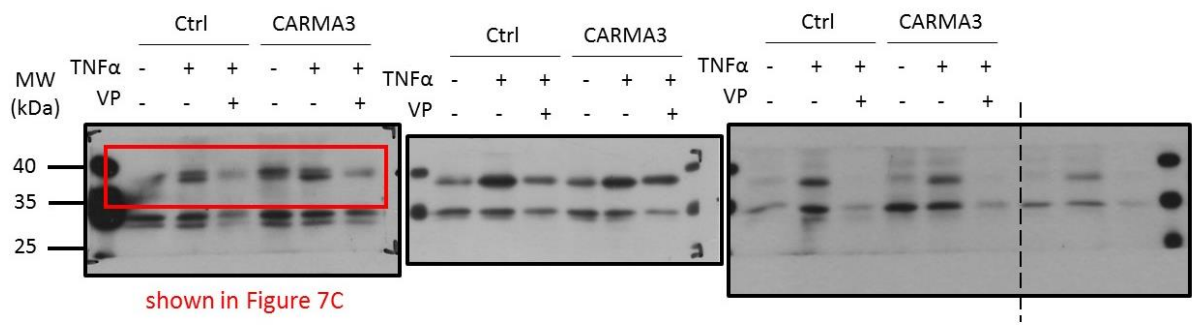
53. Figure S7C

Treatment with TNF $\alpha$  or VP in CARMA3-overexpressed cells  
 Cell: SW480 cell  
 Protein: YAP (65-78 kDa)



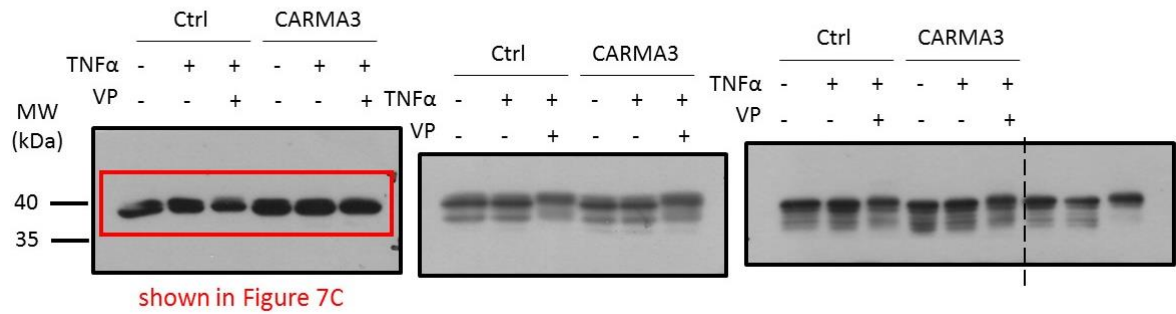
54. Figure S7C

Treatment with TNFα or VP in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: p-IκBα (40 kDa)



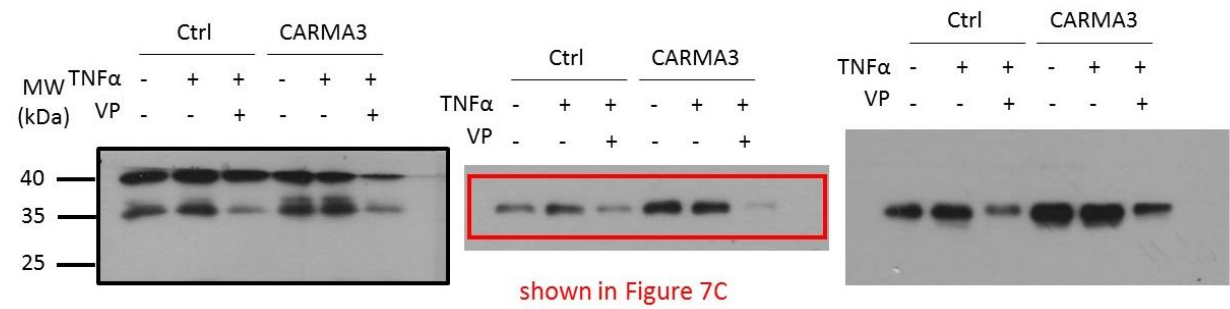
55. Figure S7C

Treatment with TNFα or VP in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: IκBα (35-41 kDa)



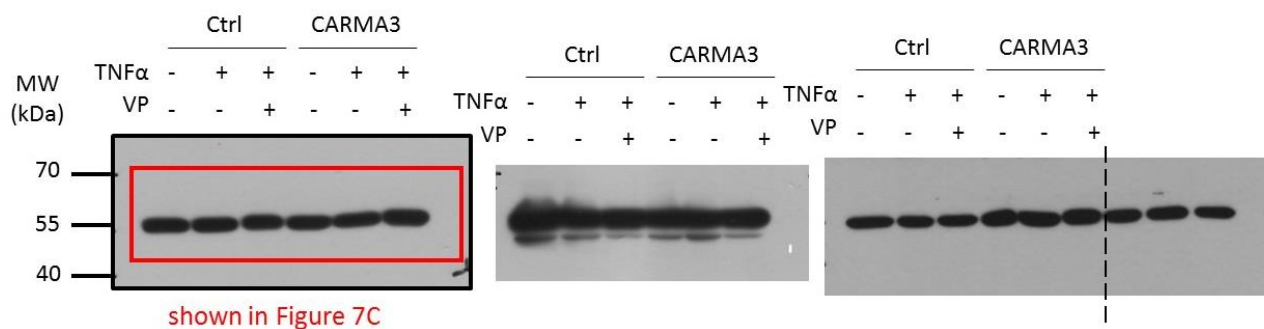
56. Figure S7C

Treatment with TNFα or VP in CARMA3-overexpressed cells  
Cell: SW480 cell  
Protein: Slug (30 kDa)

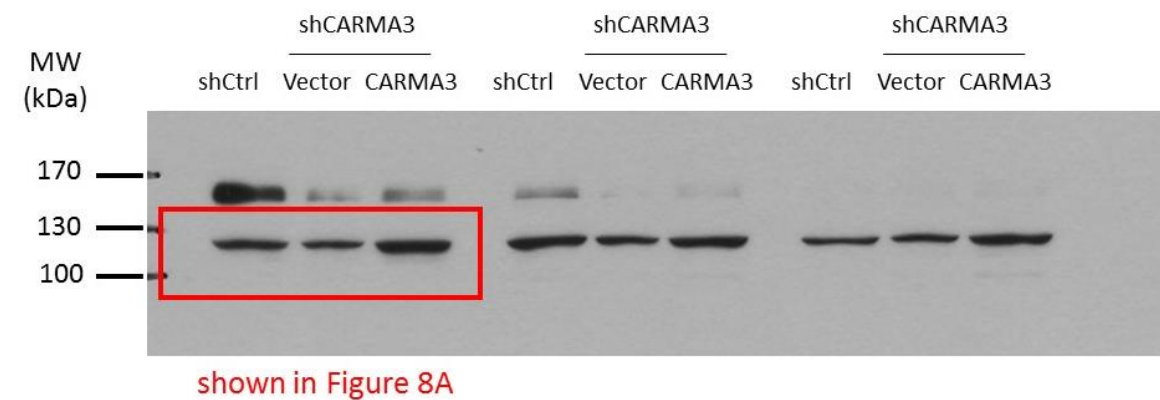


57. Figure S7C

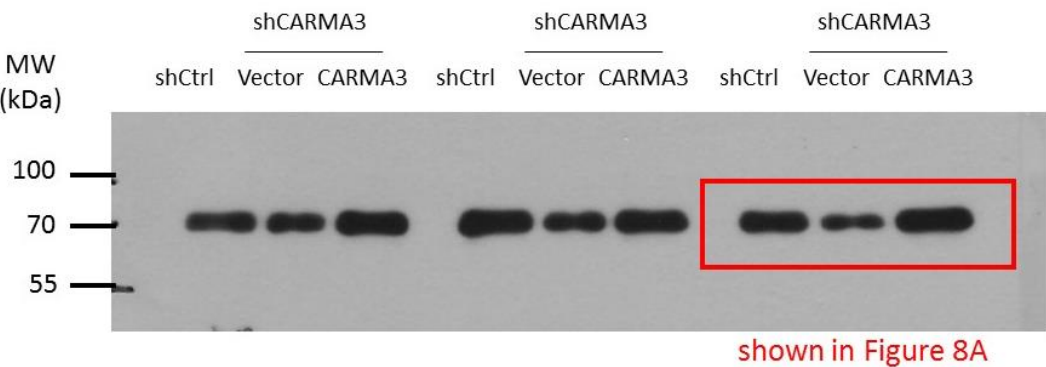
Treatment with TNFα or VP in CARMA3-overexpressed cells  
 Cell: SW480 cell  
 Protein: α-Tubulin (55 kDa)



58. Figure S8A  
 Overexpression of CARMA3 by plasmid transfection in CARMA3-silenced cells  
 Cell: HCT116 cell  
 Protein: CARMA3 (116kDa)

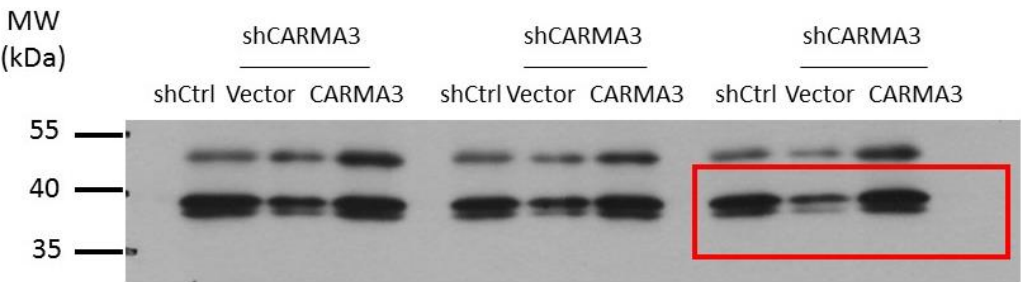


59. Figure S8A  
 Overexpression of CARMA3 by plasmid transfection in CARMA3-silenced cells  
 Cell: HCT116 cell  
 Protein: YAP (65-78 kDa)



60. Figure S8A

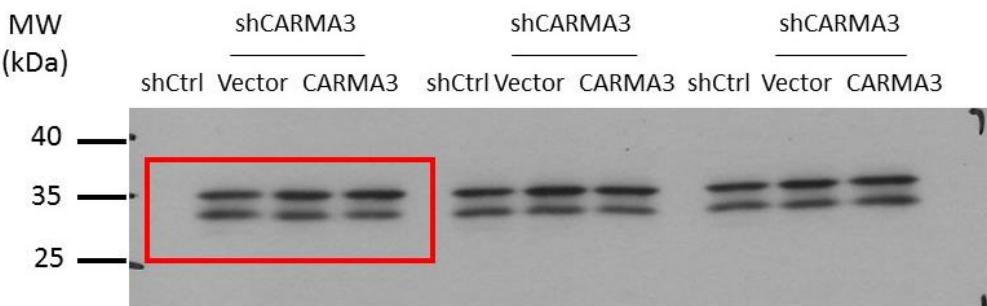
Overexpression of CARMA3 by plasmid transfection in CARMA3-silenced cells  
Cell: HCT116 cell  
Protein: p-IkBα (40 kDa)



shown in Figure 8A

61. Figure S8A

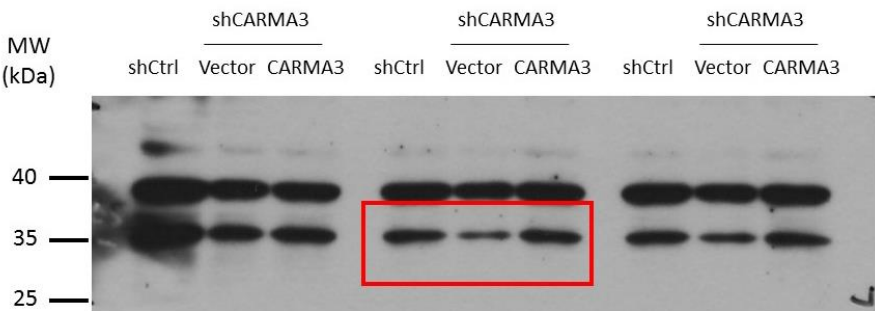
Overexpression of CARMA3 by plasmid transfection in CARMA3-silenced cells  
Cell: HCT116 cell  
Protein: IkBα (35-41 kDa)



shown in Figure 8A

62. Figure S8A

Overexpression of CARMA3 by plasmid transfection in CARMA3-silenced cells  
Cell: HCT116 cell  
Protein: Slug (30 kDa)

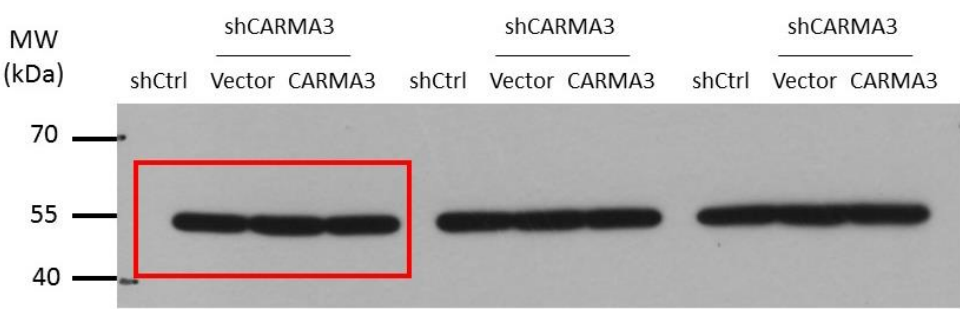


shown in Figure 8A

63. Figure S8A



Overexpression of CARMA3 by plasmid transfection in CARMA3-silenced cells  
Cell: HCT116 cell  
Protein:  $\alpha$ -Tubulin (55 kDa)



shown in Figure 8A