

Loss of expression of the E3 ligase and tumour suppressor FBW7 attenuates vitamin D signaling.

Reyhaneh Salehi-Tabar^{1#}, Aiten Ismailova^{1#}, Zoe Carter¹, and John H. White^{1*}

¹Department of Physiology, McGill University, Montreal Qc, Canada

These authors contributed equally to this manuscript

* Corresponding author

Email: john.white@mcgill.ca

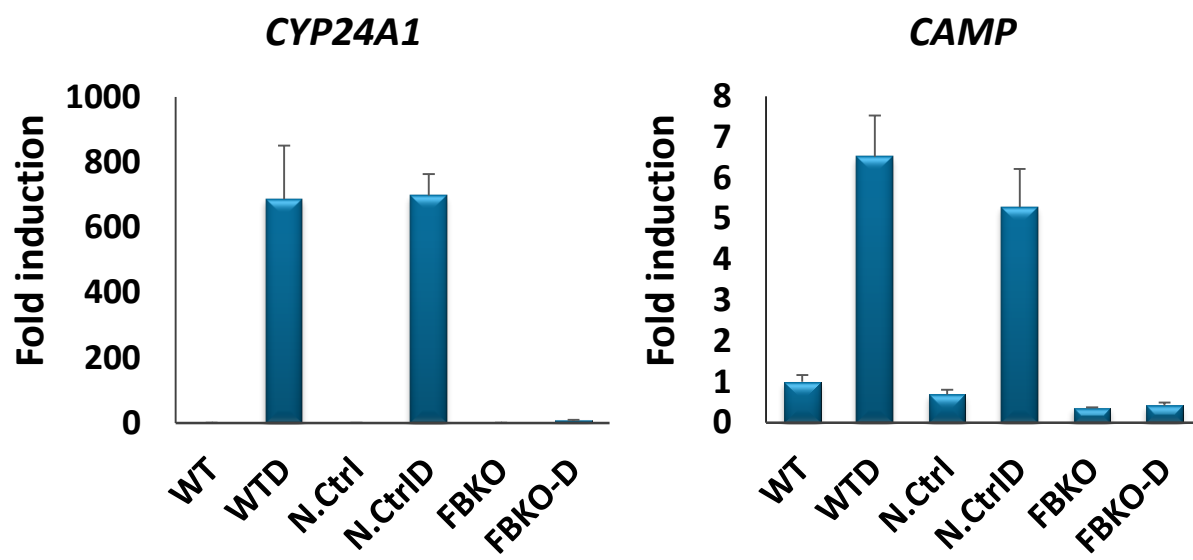


Fig S1: 1,25D-regulated expression of in wild-type and transduced cells in which *FBW7* ablation failed (Neg) or succeeded (FBKO).

Vitamin D receptor pathway (upregulated in GSEA from Wiki Pathways)

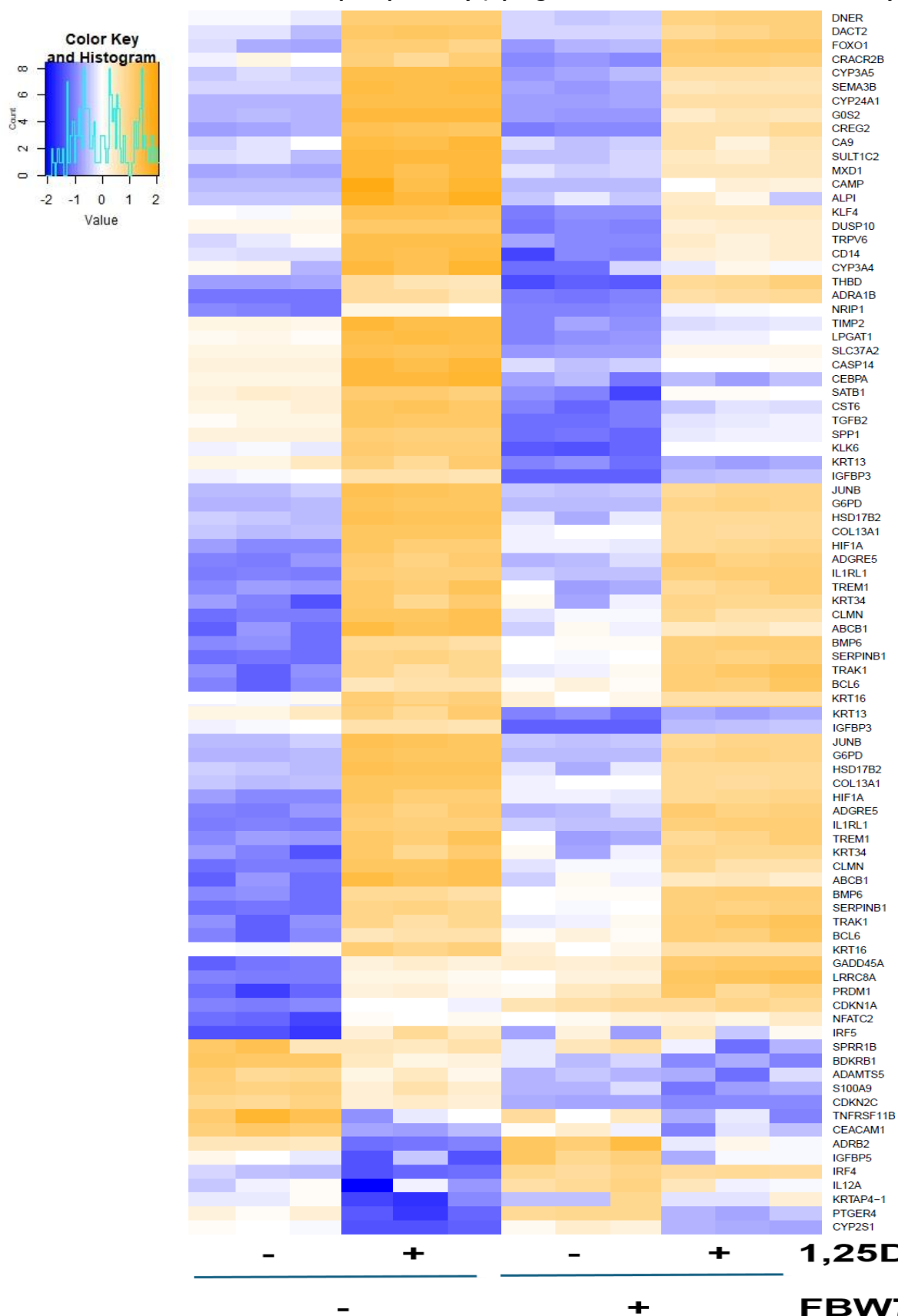


Fig. S2 : Heatmap of 1,25D-regulated DEGs with the VDR pathway in control and FBW7 KO cells.

A

Counts per gene

Condition	Donor	<i>RXRA</i>	<i>RXRB</i>	<i>RXRG</i>	Bulk average of all gene counts
C	1	26722	90	94	2428
	2	24401	41	81	2205
	3	28763	41	102	2556
D	1	17619	140	26	1630
	2	20904	6	23	1964
	3	26229	3	20	2376
KO	1	17592	40	8	2208
	2	17883	10	8	2349
	3	16093	9	5	2042
KO+D	1	17412	4	2	2435
	2	16538	6	3	2342
	3	18270	52	1	2511

B

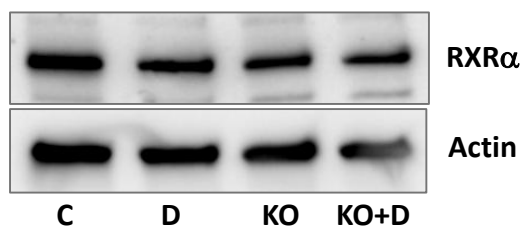


Fig. S3: Gene counts of *RXRA*, *RXRB* and *RXRG*, as well as average gene counts based on RNAseq for each donor of C, 1,25D (D), KO, and KO+D treated cells. **B.** Western blot of RXRα in wild-type and FBW7 KO cells. (See uncropped gel images in Supplementary Fig. S4)

Table S4: Enriched activated diseases & functions FBW7 KO vs Control

Diseases or Functions Annotation	T	p-value	T ×	Predicted Activation State	T ×	Activation z-score
Blood pressure		1.01e-13		Increased		3.094
Organismal death		1.16e-53		Increased		2.915
Hypertrophy of heart		4.84e-17		Increased		2.897
Organ Degeneration		5.55e-26		Increased		2.895
Migration of smooth muscle cells		2.86e-11		Increased		2.791
Abnormal morphology of muscle		6.56e-12		Increased		2.758
Development of gastrointestinal tract		4.57e-18		Increased		2.743
Formation of solid tumor		1.49e-238		Increased		2.733
Melanoma		1.51e-147		Increased		2.733
Liver carcinoma		1.98e-68		Increased		2.721
Nonpituitary endocrine tumor		1.13e-177		Increased		2.712
Extraintestinal functional disorder		2.40e-23		Increased		2.708
Lymphoreticular neoplasm		1.12e-56		Increased		2.683
Generation of tumor		1.25e-224		Increased		2.677
Hepatobiliary carcinoma		2.46e-70		Increased		2.647
Non-hematological solid tumor		6.73e-265		Increased		2.639
Hypertrophy		3.61e-20		Increased		2.611
Familial cardiovascular disease		7.33e-13		Increased		2.595
Solid tumor		4.32e-237		Increased		2.581
Cell movement of tumor cells		1.94e-11		Increased		2.571
Enlargement of heart		4.28e-15		Increased		2.553
Seizure disorder		1.56e-12		Increased		2.524
Myeloid or lymphoid neoplasm		4.12e-76		Increased		2.514
Acute leukemia		1.59e-35		Increased		2.479
Progression of tumor		2.49e-13		Increased		2.473
Abnormal morphology of cardiovascular system		4.31e-22		Increased		2.468
Migration of muscle cells		5.31e-12		Increased		2.465
Upper abdominal cancer		4.99e-119		Increased		2.460
Tumorigenesis of lymphocytes		1.29e-48		Increased		2.458
Migration of cancer cells		1.23e-11		Increased		2.457
Uveal or cutaneous melanoma		3.79e-134		Increased		2.452
Seizures		2.03e-11		Increased		2.437
Nervous system neoplasm		4.52e-154		Increased		2.435
Endocrine cancer		4.45e-176		Increased		2.425
Growth of solid tumor		7.64e-27		Increased		2.416
Permeability of vascular system		5.32e-11		Increased		2.403
Development of head and neck tumor		3.59e-27		Increased		2.401
Intrathoracic organ tumor		2.86e-74		Increased		2.384
Thoracic neoplasm		1.56e-74		Increased		2.384
Abnormal morphology of mediastinum		1.07e-17		Increased		2.378

Table S5: Enriched suppressed diseases & functions FBW7 KO vs Control

Diseases or Functions Annotation	Υ	p-value	$\Upsilon \times$	Predicted Activation State	$\Upsilon \times$	Activation z-score
Size of body		7.44e-25		Decreased		-2.975
Cell movement of leukocytes		4.11e-27		Decreased		-2.918
Cell movement of blood cells		4.24e-28		Decreased		-2.703
Migration of blood cells		3.45e-27		Decreased		-2.666
Leukocyte migration		4.35e-27		Decreased		-2.621
Aggregation of cells		3.73e-19		Decreased		-2.611
Assembly of intercellular junctions		1.76e-11		Decreased		-2.560
Learning		1.65e-15		Decreased		-2.541
Cell movement of phagocytes		6.96e-21		Decreased		-2.476
Quantity of cells		2.89e-32		Decreased		-2.437
Transport of metal ion		6.34e-12		Decreased		-2.411
Transport of cation		1.89e-13		Decreased		-2.381
Transport of inorganic cation		8.82e-13		Decreased		-2.370
Cell movement of antigen presenting cells		5.15e-14		Decreased		-2.368
Formation of intercellular junctions		9.74e-12		Decreased		-2.331
Assembly of cell junction		1.52e-13		Decreased		-2.314
Formation of cell-cell contacts		8.88e-12		Decreased		-2.303
Aggregation of blood cells		1.65e-13		Decreased		-2.231
Synthesis of nitric oxide		2.31e-11		Decreased		-2.227
Metabolism of reactive oxygen species		1.47e-17		Decreased		-2.199
Cognition		8.49e-16		Decreased		-2.181
Formation of plasma membrane		3.43e-12		Decreased		-2.148
Cell movement of myeloid cells		1.88e-23		Decreased		-2.123
Development of central nervous system		2.73e-20		Decreased		-2.092
Synthesis of reactive oxygen species		5.95e-17		Decreased		-2.075
Cell movement of macrophages		4.78e-12		Decreased		-2.074
Progressive motor neuropathy		9.88e-14		Decreased		-2.062
Progressive neuropathy		1.40e-13		Decreased		-2.062
Transport of ion		2.86e-13		Decreased		-2.054
Inflammatory response		1.97e-25		Decreased		-2.043
Formation of cellular membrane		3.04e-12		Decreased		-2.022
Differentiation of epithelial tissue		5.07e-17		Decreased		-2.017

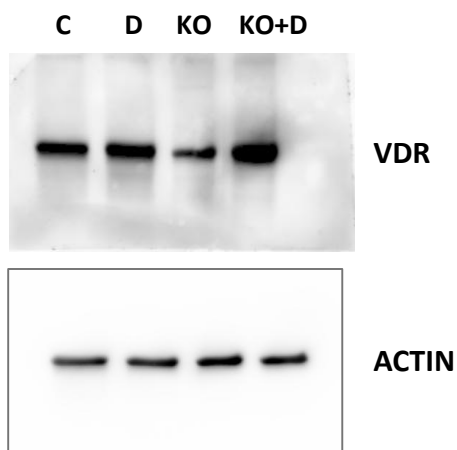
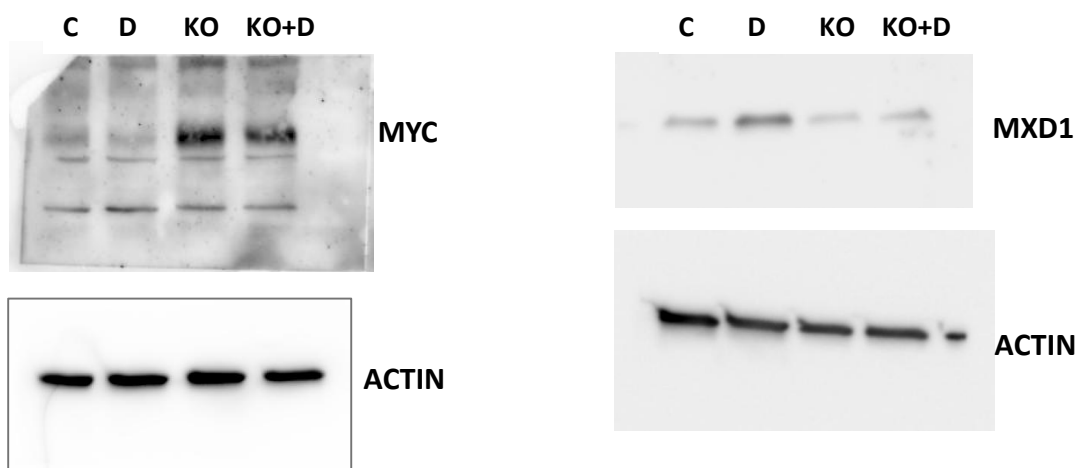
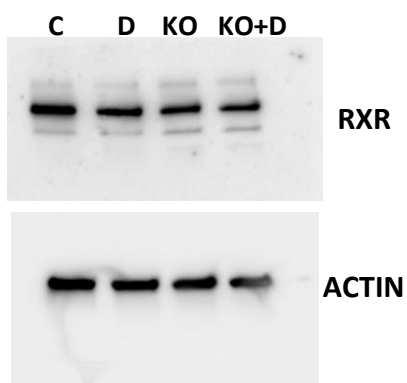
A**B****C**

Fig. S4. A. Uncropped images of figure 1E. **B.** Uncropped images of figure 4B. **C.** Uncropped images of figure S3B. Actin controls are from the same blot.