NDRG1 attenuates epithelial-mesenchymal transition of nasopharyngeal cancer cells via blocking Smad2 signaling

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Supplemental Materials

Supplemental Table:

Table S1. The differentially expressed proteins identified by MALDI-TOF-TOF spectrometry between NPC 5-8F and 5-8F-LN cell lines.

MSDB ID	Protein full name	5-8F/5-8F-LN
LAMIN	Prelamin-A/C	2.01898
K2C7	Keratin, type II cytoskeletal 7	1.8887
NDRG1	N-myc downstream-regulated gene 1 protein	1.50555
EFTU	Elongation factor Tu,mipochondrial	1.64925
FAS	Tumor necrosis factor receptor superfamily member 6	1.536
SPB4	Serpin B4	2.83408
ACADS	Short-chain specific acyl-CoA dehydrogenase, mitochondrial	1.52603
ACTA	Alpha-actin-2	2.11457
IPYR	Inorganic pyrophosphatase	1.83611
CTSD	Cathepsin D	1.50259
GDIR1	Rho GDP-dissociation inhibitor 1	2.09702

Supplemental Figure 1

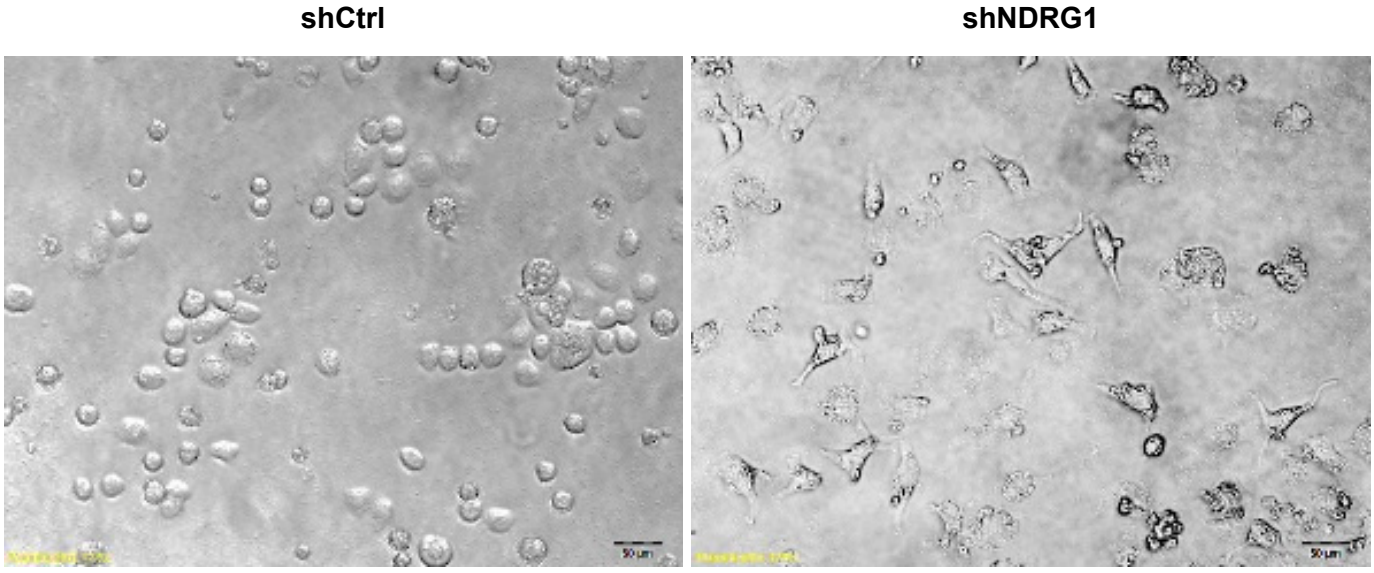

Figure S1: Knockdown of NDRG1 by shRNA in 5-8F cells caused a morphological alteration that resembles an EMT.

Supplemental Figure 2

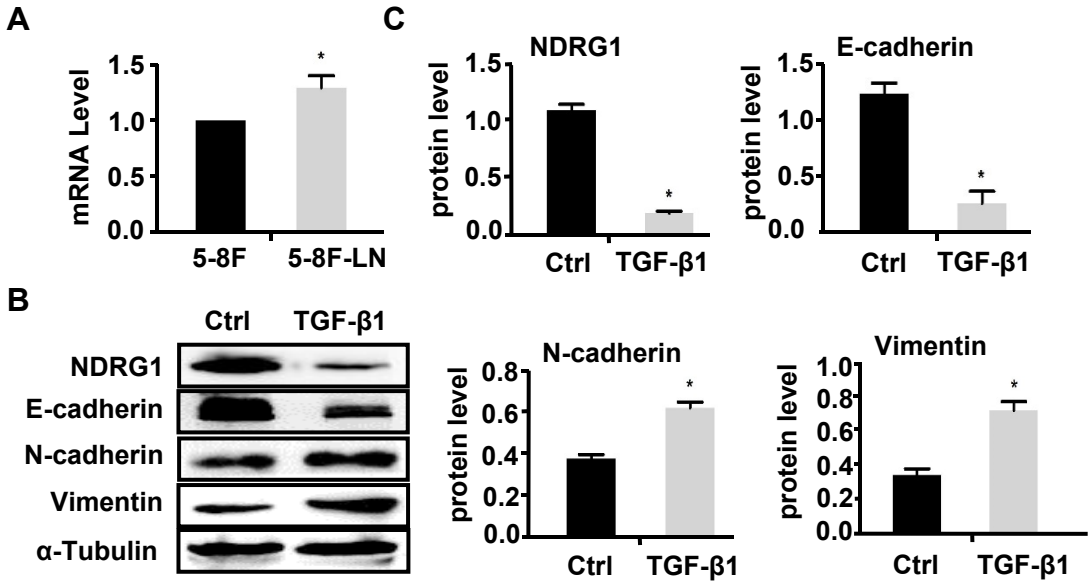

Figure S2. TGF-β1 appeared to be involved in the EMT of 5-8F cells. (A) TGF-β1 mRNA expression was increased in 5-8F-LN compared to 5-8F cells as detected by quantitative RT-PCR. *P<0.05 compared to 5-8F cells, n=3. (B) TGF-β1 blocked NDRG1 expression while inducing EMT of 5-8F cells as shown by downregulation of E-cadherin and upregulation of N-cadherin and Vimentin. 5-8F cells were treated with TGF-β1 (5ng/ml) for 48 hours. Western blotting was performed to detect the expression of indicated proteins. (C) Quantification of protein levels shown in B by normalized to α-Tubulin. *P<0.01 compared to 5-8F cells treated with vehicle (Ctrl), n=3.

Supplemental Figure 3

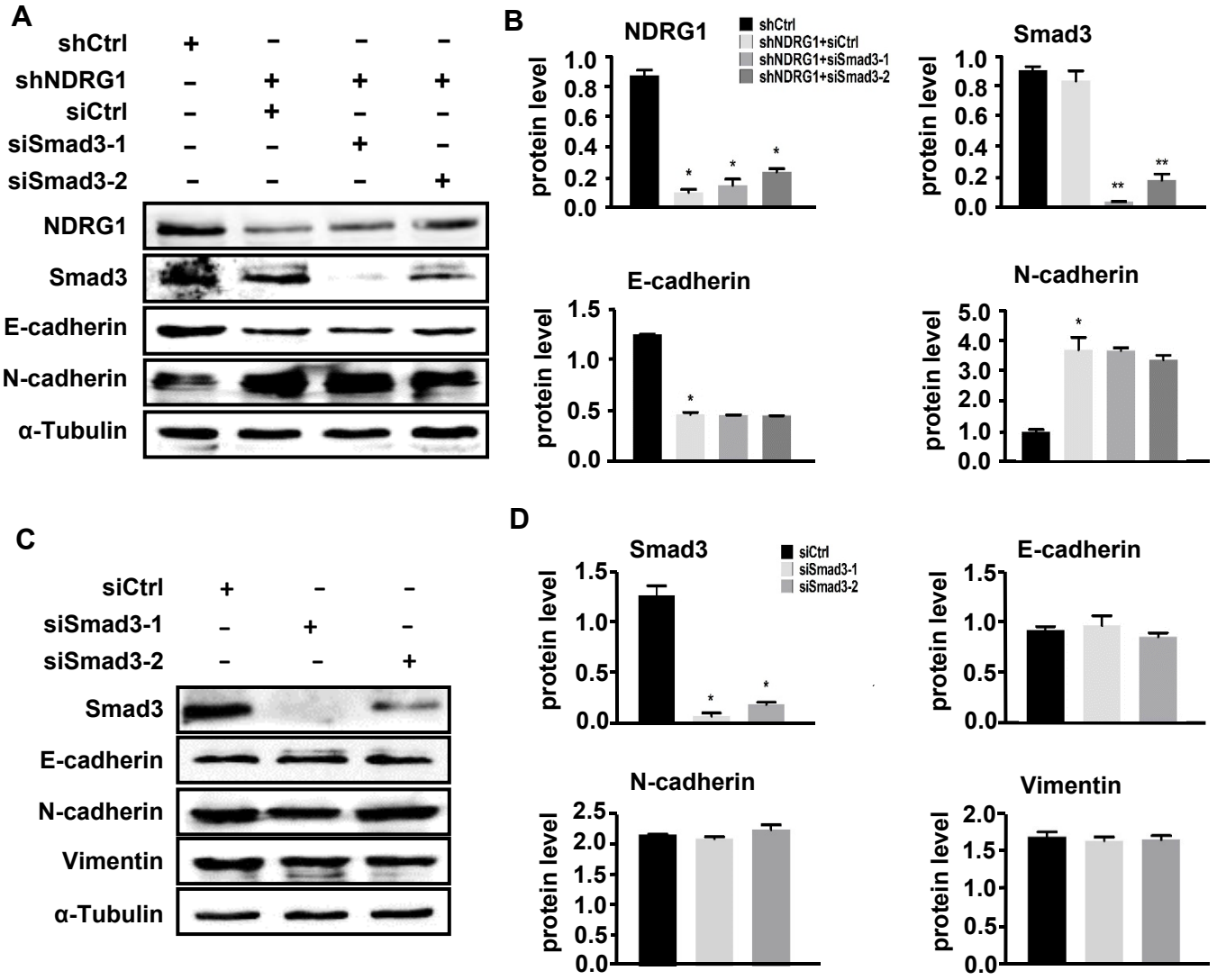

Figure S3: Smad3 signaling was not essential for NDRG1 knockdown-induced EMT of NPC 5-8F cells. (A) Knockdown of Smad3 did not alter E-Cadherin and N-cadherin expression in NDRG1-deficient 5-8Fcells. (B) Quantification of protein levels shown in A by normalized to α-Tubulin. *P<0.01 compared to NDRG1 control shRNA (shCtrl)-treated groups; **P<0.001 compared to Smad3 control siRNA (siCtrl)-treated groups for each individual protein, n=3. (C) Knockdown of Smad3 did not alter the expression of E-Cadherin, N-cadherin, and Vimentin in 5-8F-LN cells . (D) Quantification of protein levels shown in C by normalized to α-Tubulin. *P<0.001 compared to Smad3 control siRNA (siCtrl)-treated groups for Smad3 protein, n=3.