

SUPPLEMENTARY DATA

Supplementary Table 1. (from fig. 5) BMP-7 acts partially through the SMAD pathway. C-peptide (ng/μg of DNA) following hNEPT treatment with BMP-4, THR-123, and BMP-7/BMP-7 + dorsomorphin.

Prep	<i>C-peptide (ng/μg of DNA)</i>					
	T=0	Control	BMP-4	THR-123	BMP-7	BMP-7 + DM
1	0.5	5.1	76.1		51.8	34.2
2	0.6	17.6	47.2			
3	0.1	11.8	56.6		40.3	22.8
4	0.1	29.1		54.6		
5	0.1	9.0		33.5		
6	1.2	7.6		18.4		
7	0.1	18			75.0	19.5

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Supplementary Table 2. Antibodies used.

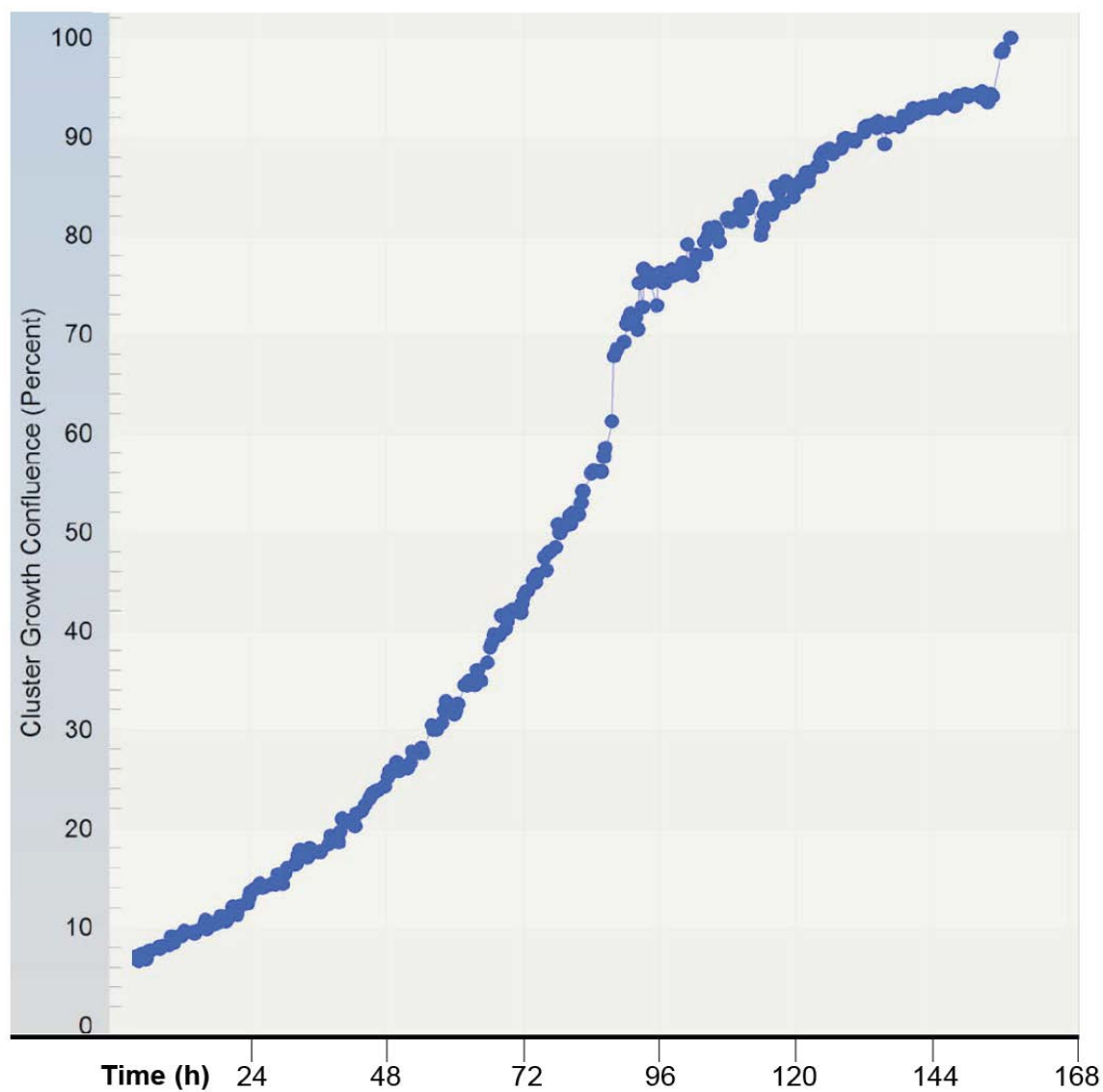
Secondary antibodies: Alexa Fluor 488 (706-545-148), 594 (706-515-148) and 647 (706-605-148) Conjugated AffinityPure Donkey Anti-Guinea Pig and Fluorescein (FITC) –conjugated Donkey antichickens (703-096-156), all from Jackson ImmunoResearch laboratories, Inc. Donkey anti-goat 568, donkey anti rabbit 488, donkey anti-mouse 594, donkey anti-mouse 647, goat anti-chicken 488, goat anti-rabbit 488, goat anti-mouse 488 and goat anti-rabbit 568, all from Life technologies: Nucleus was counter-stained with 4', 6-diamidino-2-phenylindole (DAPI, Life technologies D1306).

Primary antibodies:

Name	Host	Source/ catalog number	Dilution/ Conc.
Amylase 2B	Mouse	SantaCruz (sc-46657)	1:200
α -Amylase	Rabbit	Sigma (A8273)	1:200
Cytokeratin 19	Mouse	Dako (M0888)	1:100
CA19-9	Mouse	Leica (NCL-L-CA19-9)	1:100
PDX1	Goat	R&D systems (AF2419)	15 mg/mL
NKX6.1	Mouse	DSHB (F55A12-s)	2 mg/mL
MafA	Mouse	Abcam (ab57807)	1:100
Insulin	Guinea pig	Dako (A0564)	1:250
C-peptide	Mouse	Chemicon (CBL94)	1:100
Anti-proinsulin C-peptide clone CPEP-0	Mouse	Millipore 05-1109	1:25
Glucagon	Rabbit	Biogenex (PU039-UP)	1:200
Glucagon	Rabbit	Dako (A0565)	1:250
Pancreatic Polypeptide	Rabbit	Chemicon (AB939)	1:200
Somatostatin	Rabbit	Dako (A0566)	1:200
Vimentin	Rabbit	GeneTex (GTX100619)	1:500
GFP	Chicken	Aves GFP-1020	1:500
BMPR1A (ALK3)	Mouse	Life Span Biosciences 1759/58348)	1:50
Phospho-Smad1/5/9	Rabbit	Cell Signaling 13820	1:250

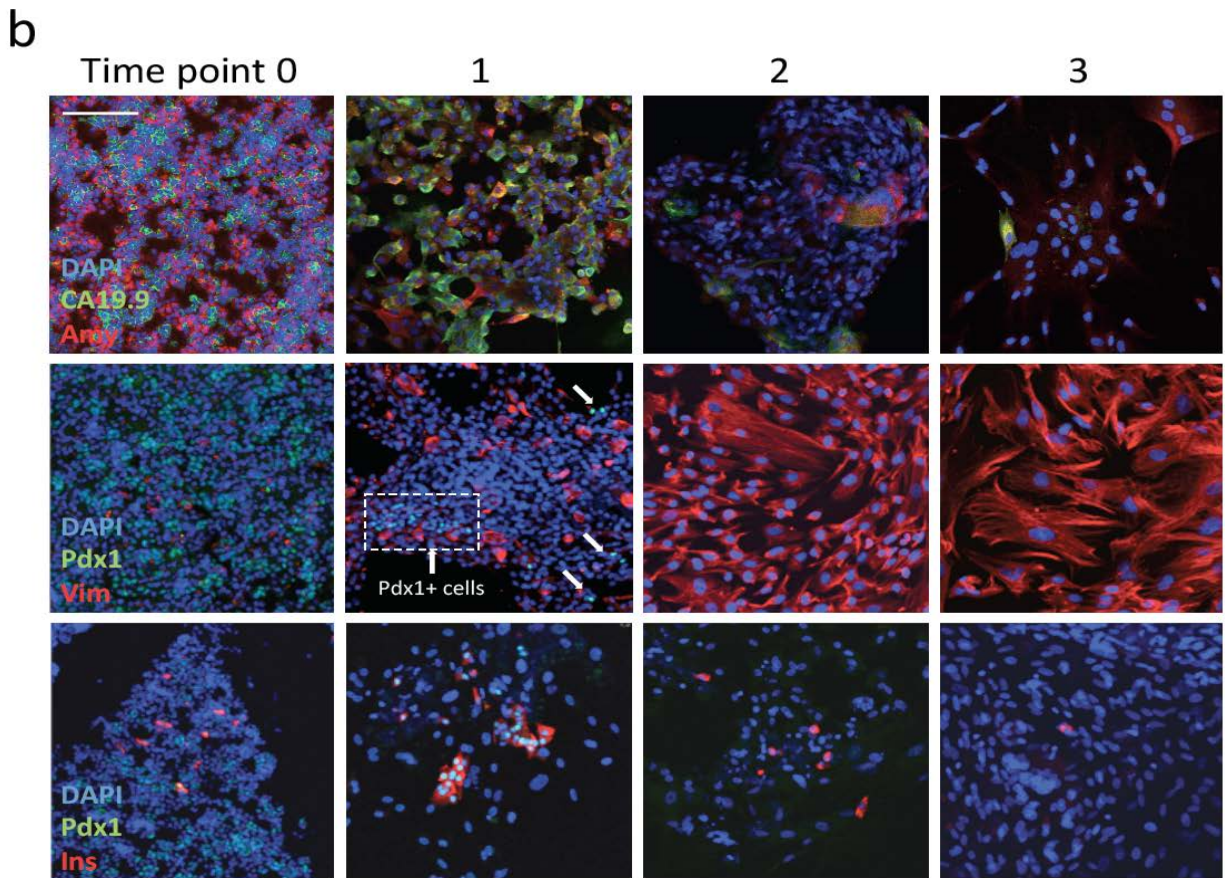
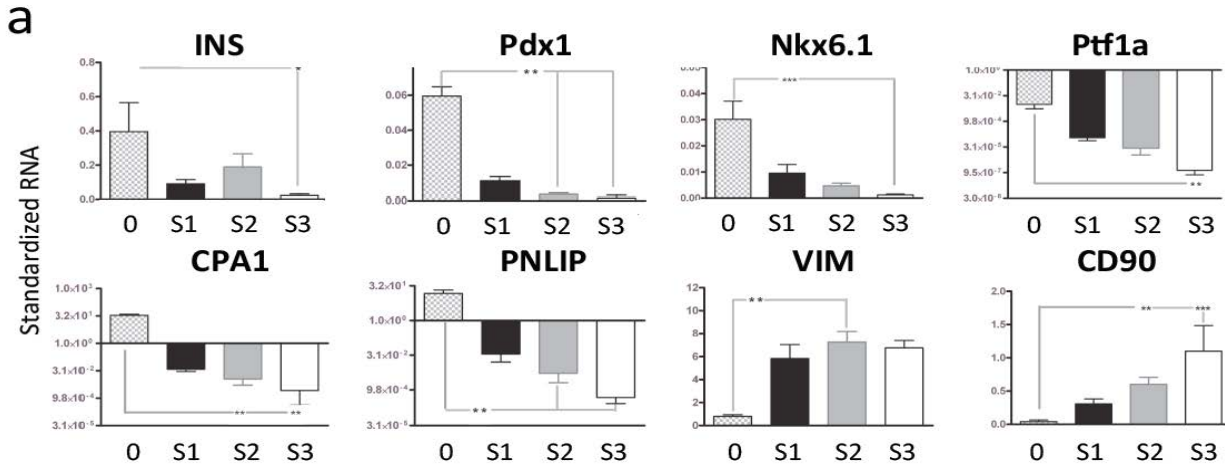
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Supplementary. Figure 1. Real-time imaging (Incucyte Zoom™)-assisted analysis of hNEPT colony growth upon BMP-7 exposure. X-axis: Time (hours). Y axis: Confluence percentage of a selected field.



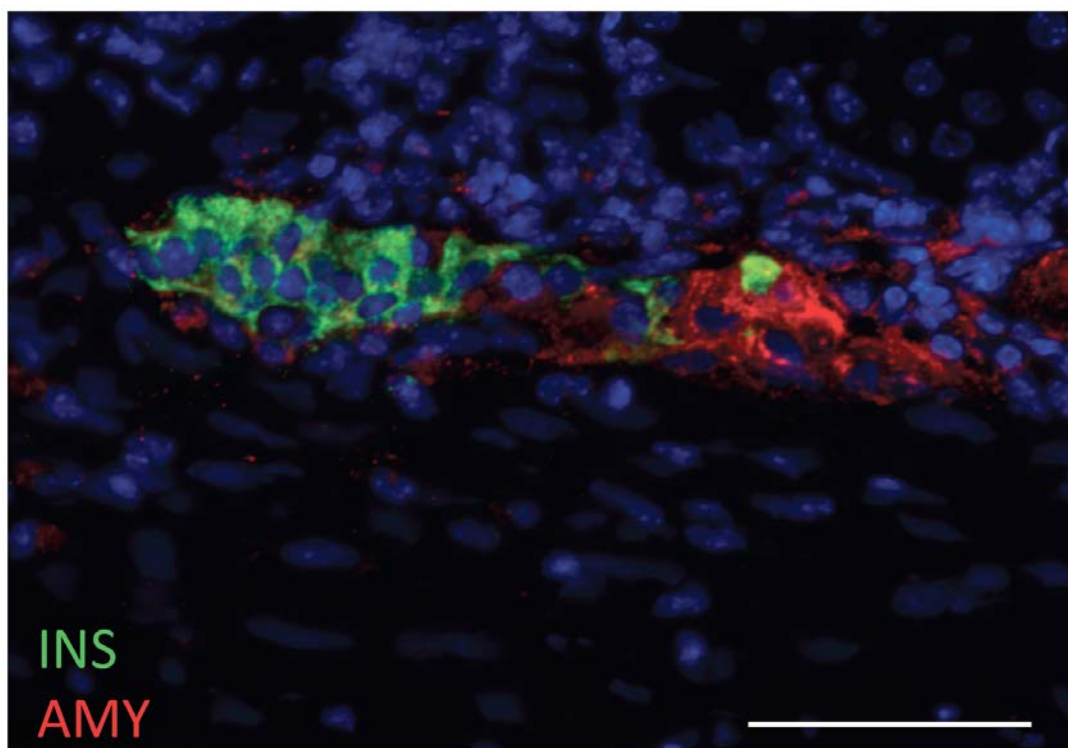
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Supplementary. Figure 2. Long-term hNEPT culture results in phenotypic changes consistent with EMT. (A) TLDA qRT-PCR stage-wise analysis of gene expression of hNEPT as a function of time in longterm culture. Markers: INS (insulin), PDX1, NKX6.1, Ptf1a, CPA1 (carboxypeptidase A), PNLIP (pancreatic lipase), VIM (vimentin) and CD90. X-axis, stages of culture: 0, after isolation; S1, stage 1 (days 6-8); S2, stage 2 (days 10-12); S3, stage 3 (days 15-17). Of note, the experimental design



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Supplementary. Figure 3. IF analysis of explanted kidney capsule grafts. Insulin (green) and amylase (red) expression in engrafted BMP-7-treated hNEPT (POD 130). Size bar: 50 μ m.



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Supplementary. Figure 4. Stage-wise qRT-PCR analysis of endocrine/exocrine marker expression. hNEPT were allowed to attach to tissue culture-treated plates for 48h. T=0 represents the analysis conducted on attached cells at 48h. hNEPT were subsequently exposed to BMP-7 in the presence of serum for 4-6d (stage 2). As before, qRT-PCR analyses were done at the end of the stage. In stage 3, cells were cultured for 3-4d in serum-free medium without BMP-7. qRT-PCR was done, again, at the end of the stage. Results are normalized against t=0 (100% of expression). Insulin and other pancreatic/epithelial cell markers were down-regulated in stage 2 (BMP-7 treatment), only to be up-regulated several-fold in the last stage of culture (serum-free, no BMP-7). Of note, fold-increases vs. t=0 are normalized to relevant endogenous controls. However, since there is a ~5-fold increase in DNA content throughout culture (denoting proliferation, see Results), the absolute increase of endocrine/exocrine marker expression at the end of the treatment is actually much higher. Abbreviations: INS, insulin; AMY: amylase; KRT19, cytokeratin 19; GCG: glucagon.

