

Supplementary Data

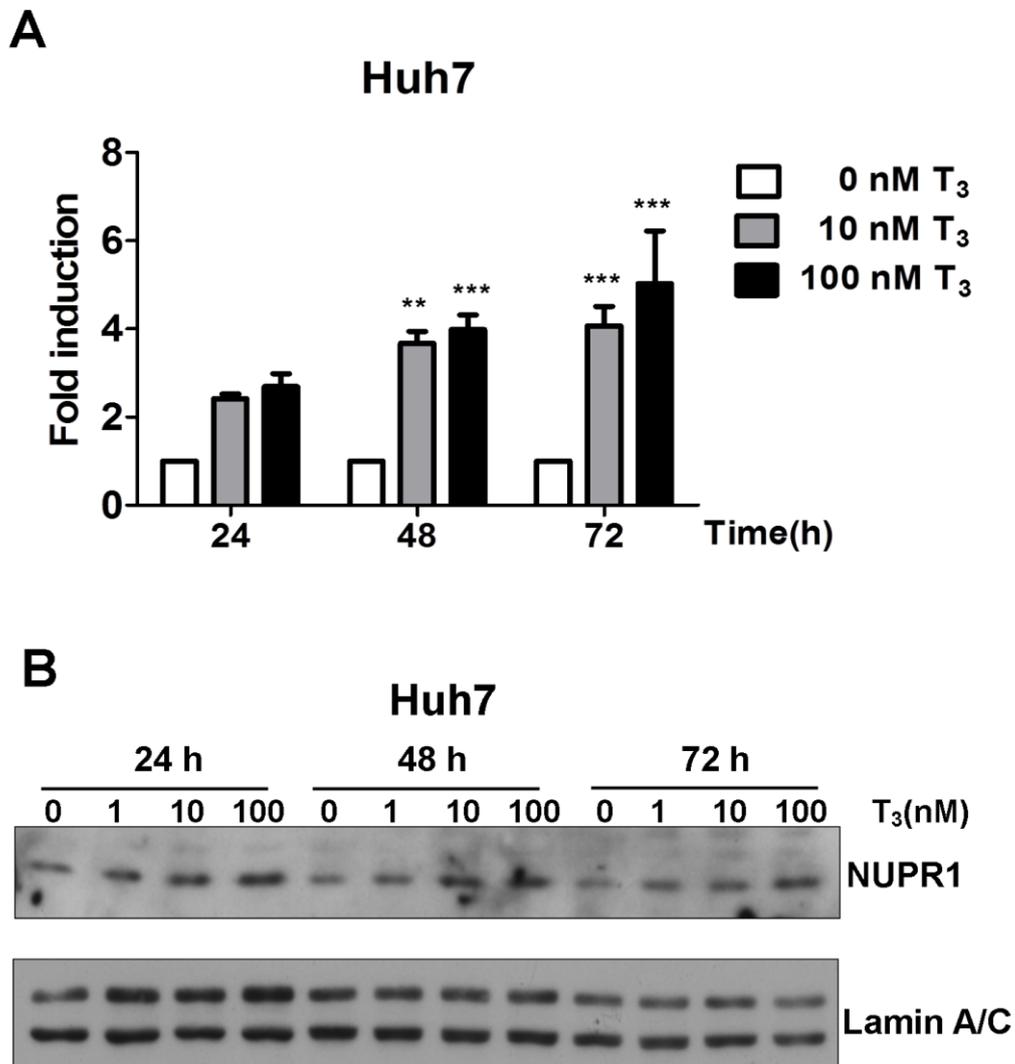


Figure S1: NUPR1 expression level in Huh7 hepatoma cells. (A) Huh7 cells were treated with T₃ (0–100 nM) for 24–72 h, and NUPR1 expression levels were measured using qRT-PCR. Expression of NUPR1 mRNA was normalized to that of 18s rRNA. (B) NUPR1 protein expression levels were determined in Huh7 cell lines treated for different times with different concentrations of T₃. Lamin A/C was used as an internal control.

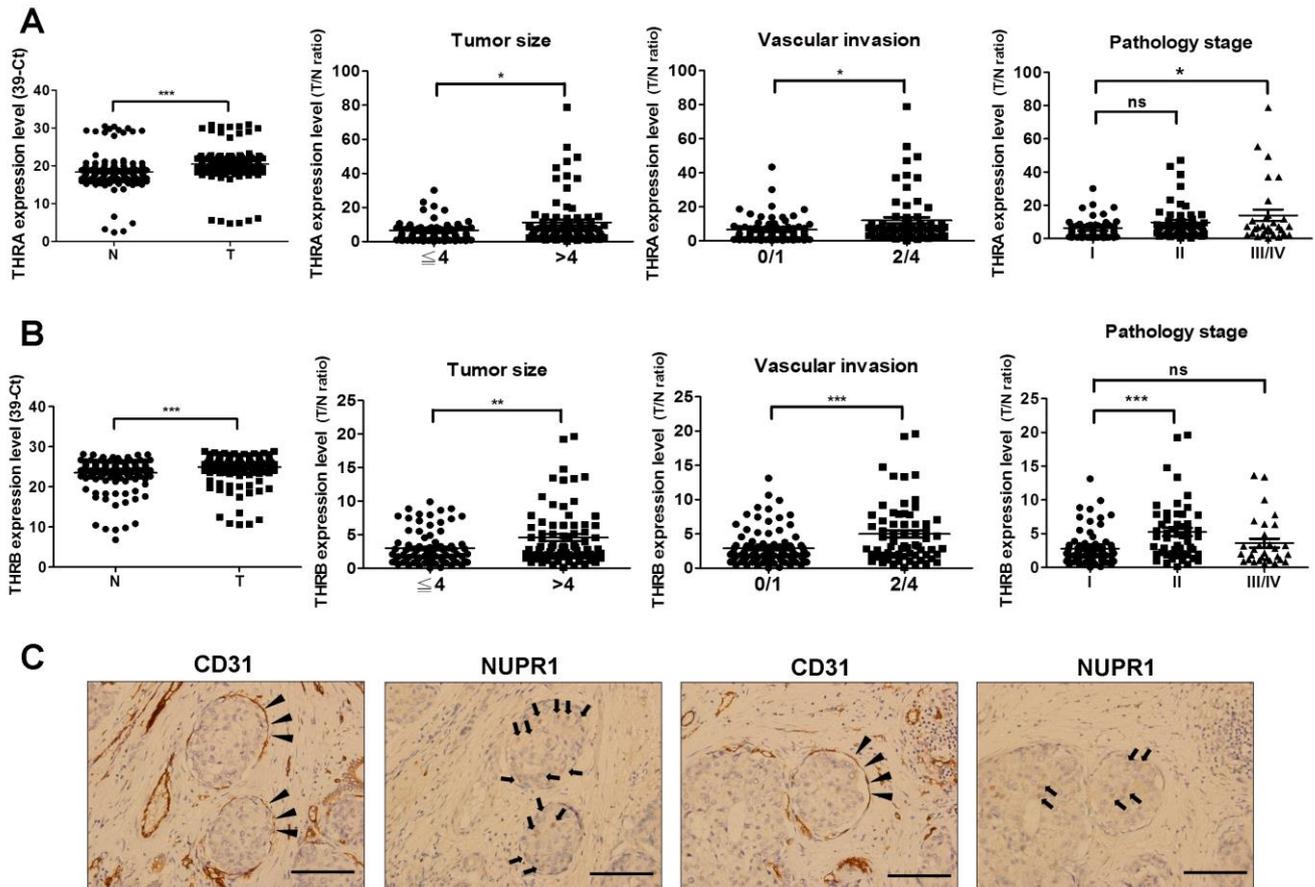


Figure S2: High expression level of thyroid hormone receptors in clinical HCC samples. (A-B) Expression of TR α and TR β in 158 paired human HCC (T) and adjacent normal (N) tissues were determined by qRT-PCR; expression of TR mRNA was normalized to that of 18s rRNA (left panel). TR expression levels were highly correlated with tumor size, vascular invasion and pathology stage. Differences were analyzed using one-way ANOVA (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$). (C) Immunohistochemical study for CD31 and NUPR1 highlighted a vascular channel (arrowheads) with a tumor thrombus expressed NUPR1 (arrows). Scale bar: 100 μm .

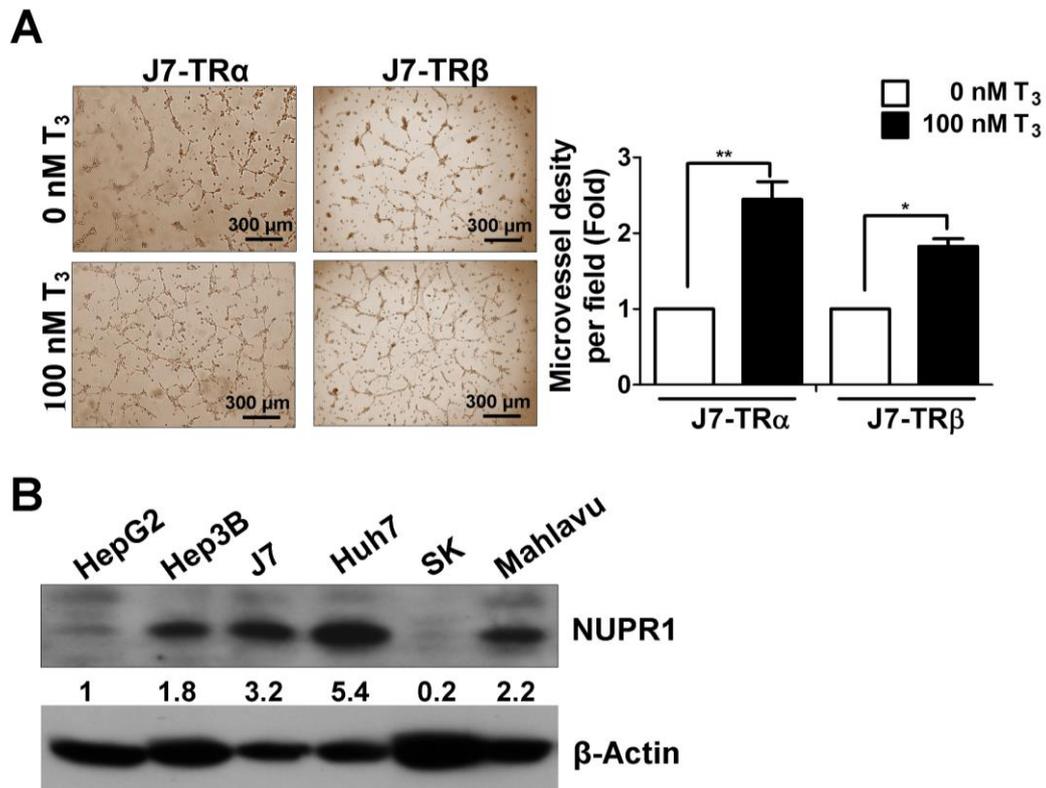


Figure S3: Thyroid hormone induces HUVECs tube formation in TR-overexpression J7 cells. (A) J7-TR α and J7-TR β cells were treated with T₃-depleted serum (0 nM T₃) or 100 nM T₃ for 24 h and CM was harvested. HUVECs were seeded onto matrigel-coated dishes and incubated with CM for 16 h. Representative images are shown (*p < 0.05, **p < 0.01). (B) Endogenous NUPR1 expression level in HepG2, Hep3B, J7, Huh7, SK and Mahlavu hepatoma cell lines were analyzed by western blot. β -actin was used as a loading control.

Table S1. Univariate and multivariate analyses of overall survival in HCC patients by Cox regression analysis

Variables	Univariate Analysis		Multivariate analysis	
	HR (95% CI)	<i>P</i>	HR (95% CI)	<i>P</i>
Gender	1.186 (0.707-1.988)	0.502		
Age	0.735 (0.437-1.237)	0.227		
Cirrhosis	0.947 (0.557-1.609)	0.834		
AFP	1.037 (0.513-2.098)	0.508		
Tumor type	1.479 (0.821-2.663)	0.173		
Tumor size	2.352 (1.368-4.045)	0.001	1.536 (0.832-2.835)	0.171
Vascular invasion	3.232 (1.833-5.699)	0.000	1.517 (1.138-2.021)	0.004
Pathological stage	2.745 (1.558-4.837)	0.000	0.917 (0.365-2.304)	0.854
Grading	0.217 (0.009-5.480)	0.147		
NUPR1	1.561 (0.925-2.633)	0.081		
THRA	1.098 (0.656-1.838)	0.712		
THRB	1.643 (0.973-2.774)	0.051		

Abbreviations: HR: hazard ratio; CI: confidence interval