

Supplemental table and figure

Table S1: Clinical characterization of the clinical samples for IHC analysis.

		TSCC (n=77)	TSCC with follow-up (n=52)	Leukoplakia (n=22)	Normal (n=12)
Age	Mean(y, Range)	49.5(21-77)	46.2(21-73)	51 (34-82)	49.9(33-71)
Gender	Male: n (%)	42(54.5%)	24(46.2%)	13(59.1%)	8(66.7%)
	Female: n (%)	35(45.5%)	28(53.8%)	9(40.9%)	4(33.3%)
Pathological T Stage	pT 4: n (%)	4(5.2%)	8(15.4%)		
	pT 3: n (%)	12(15.6%)	8(15.4%)		
Pathological N Stage	pT 2: n (%)	36(46.7%)	26(50.0%)		
	pT 1: n (%)	23(29.9%)	10(19.2%)		
Pathological pN+ : n (%)		31(40.3%)	24(46.2%)		
N Stage	pN- : n (%)	46(59.7%)	28(53.8%)		
Clinical stage	III-IV: n(%)	39(50.6%)	30(57.7%)		
	I-II: n(%)	38(49.4%)	22(42.3%)		
Grade	Well: n(%)	46(59.7%)	38(73.1%)		
	Moderate: n(%)	19(24.7%)	10(19.2%)		
	poor: n(%)	12(15.6%)	4(7.7%)		
Grade	light: n(%)			11(50.0%)	
	Moderate: n(%)			7(31.8%)	
	severe: n(%)			4(18.2%)	

Table S2: siRNA sequences used for transfection.

	Gene No.	sense (5'-3')
Bmi1	NM_005180	CCAGACCACUACUGAAUAUTT
C-myc	NM_002467	ACAGCAGCUAUCUGCAAGATT
Control		UUCUCCGAACGUGUCACGUTT

Table S3: The actual P values for the comparison of Bmi1 staining intensities.

	normal tongue tissue	LP	L LP	M-S LP	TSCC	Pathological N Stage(pN ⁺)	Clinical stage(III+IV)
normal tongue tissue		0.014	0.042	0.002	0.036		
LP					0.281		
L LP				0.037	0.610		
M-S LP					0.057		
TSCC							
Pathological N Stage(pN ⁻)						0.006	
Clinical stage (I+II)							0.004

*: L LP: light LP, M-S LP: moderate and severe LP, TSCC: tongue squamous cell carcinoma, N:

lymph node.

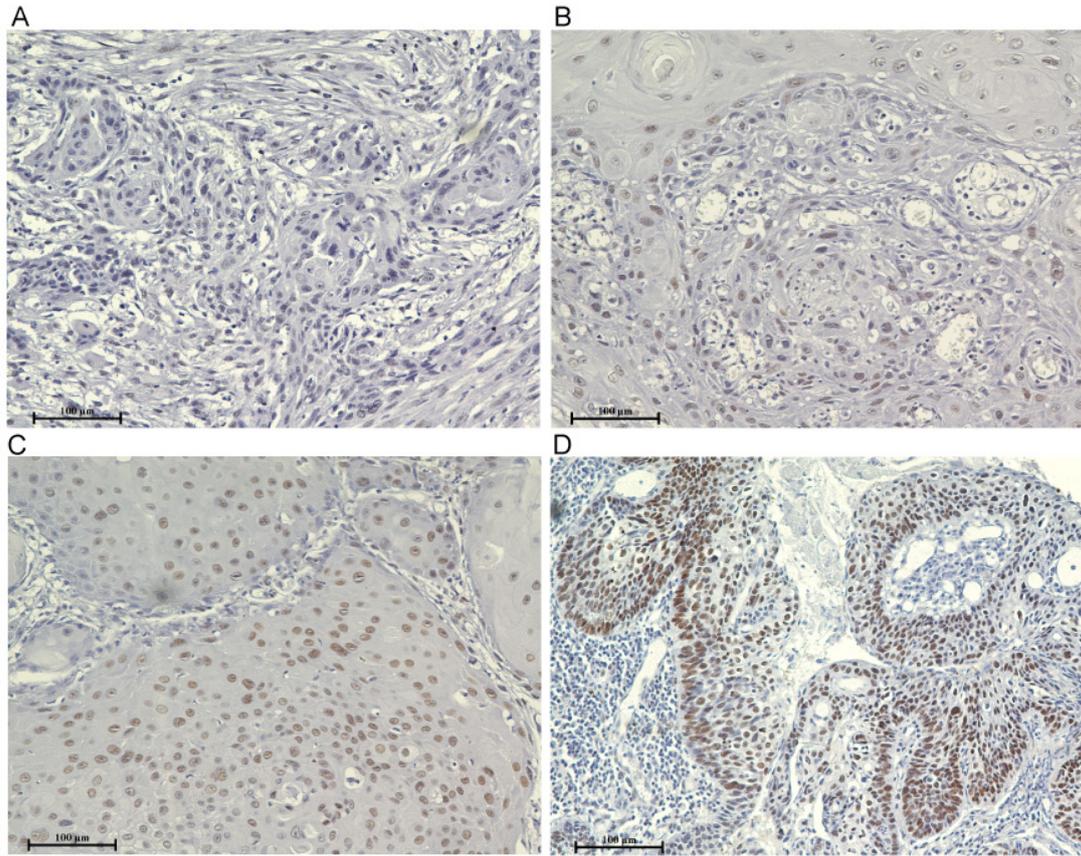


Figure S1: The representative images of Bmi1 staining intensities for each score (0-3).

Immunohistochemistry analyses for Bmi1 were performed as described in Material and Methods. **A:** score 0, **B:** score 1, **C:** score 2, **D:** score 3. (Scale bar: 100 µm).

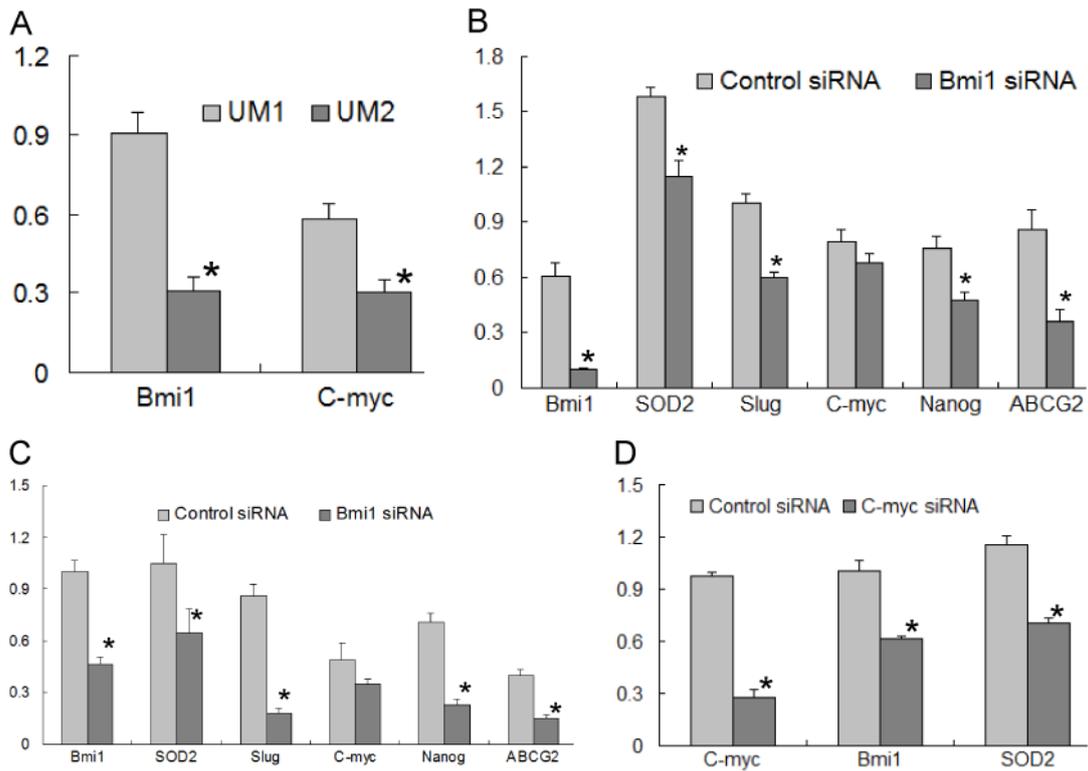


Figure S2: The results obtained from western blot (Figure 3-5) were quantified by Quantity One software.

A: The expression of Bmi1 and C-myc in UM1 and UM2 (quantified of Figure 3A). **B:** The expression of Bmi1, SOD2, Slug, and stem cell markers (ABCG2, Nanog) was significantly decreased in UM1 cells transfected with Bmi1 siRNA (quantified of Figure 3A, 3G). **C:** The expression of Bmi1, SOD2, Slug, and stem cell markers (ABCG2, Nanog) was significantly inhibited in SP cells transfected with Bmi1 siRNA (quantified of Figure 4A). **D:** The expression of C-myc, Bmi1, and SOD2 was significantly inhibited in SP cells transfected with C-myc siRNA (quantified of Figure 5E). *: $P < 0.05$.

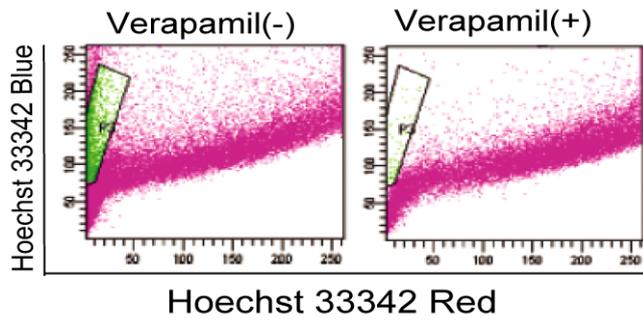


Figure S3: SP cells are sorted by Hoechst 33342 staining and identified by verapamil.

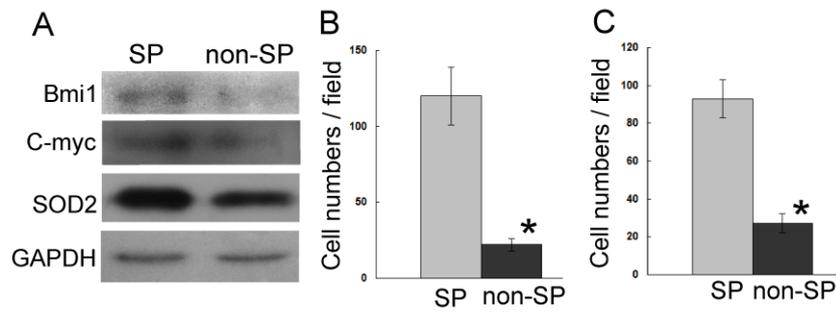


Figure S4: SP cells demonstrate increased SOD2 expression and migration and invasion abilities in TSCC.

(A) The expression of C-myc, Bmi1, and SOD2 was detected using western blotting, with GAPDH as a loading control. (B, C) The migration (B) and invasion (C) abilities were significantly higher for SP cells compared to non-SP cells. *: $P < 0.05$.