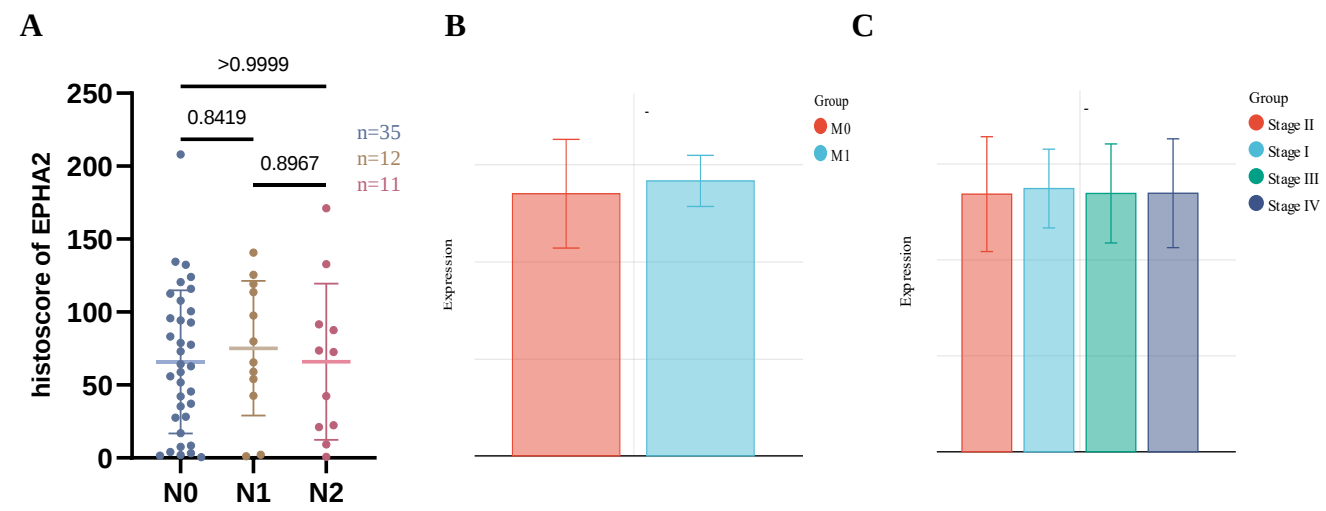
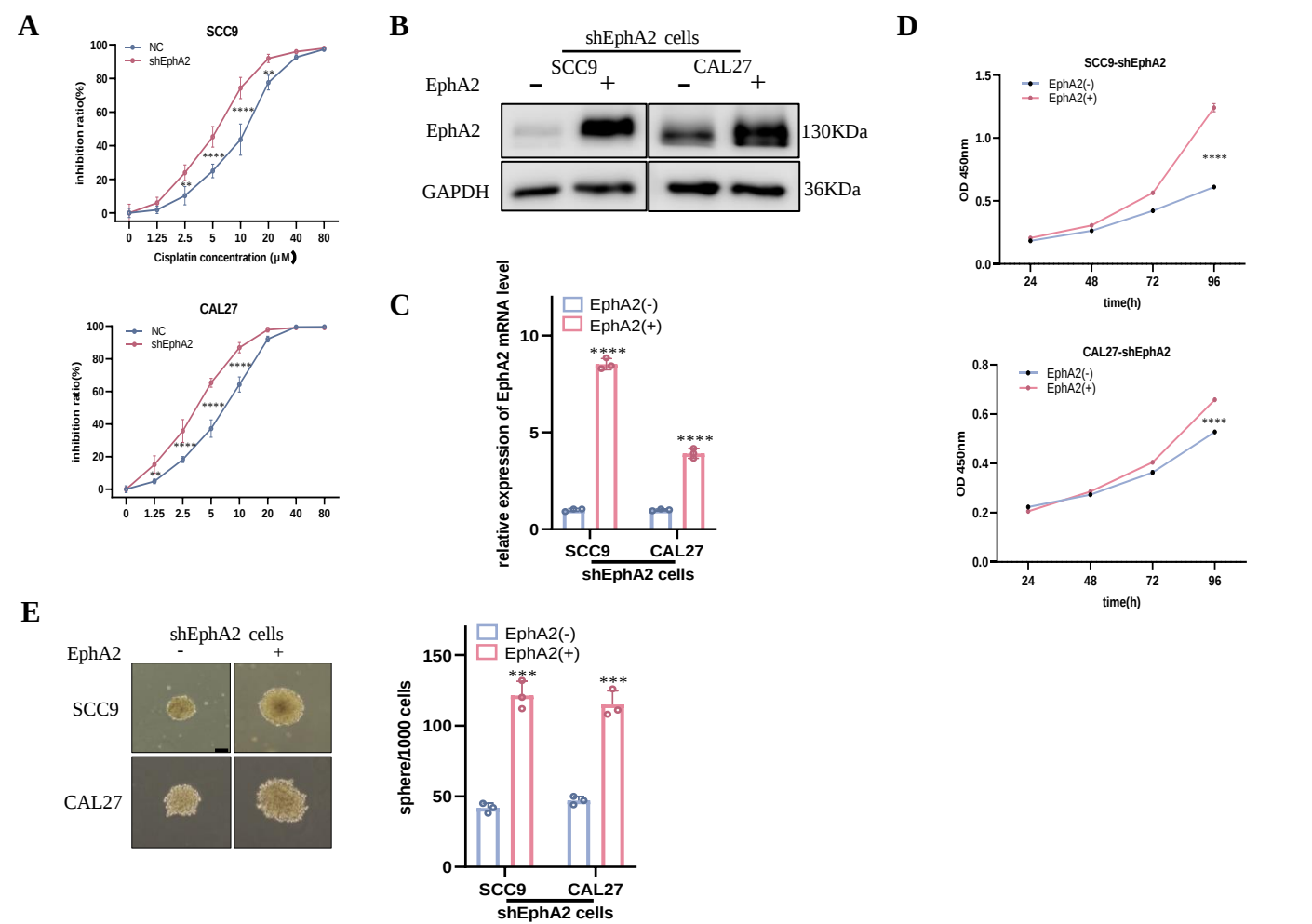


Supplementary Fig. 1



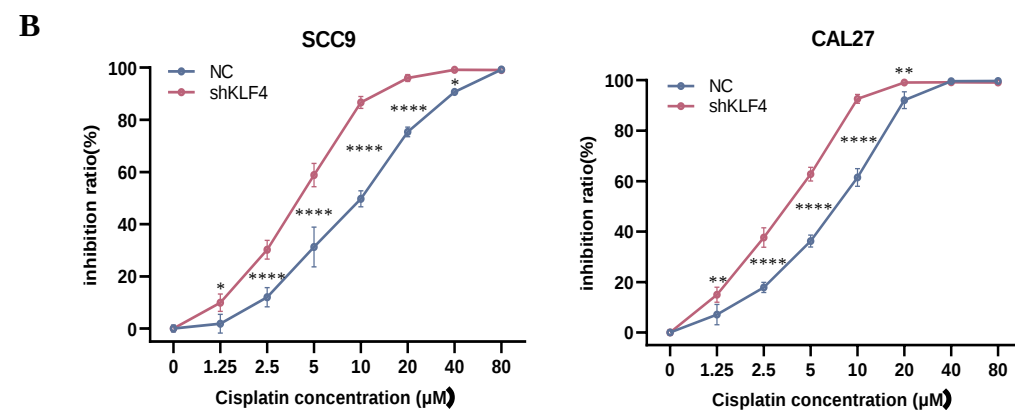
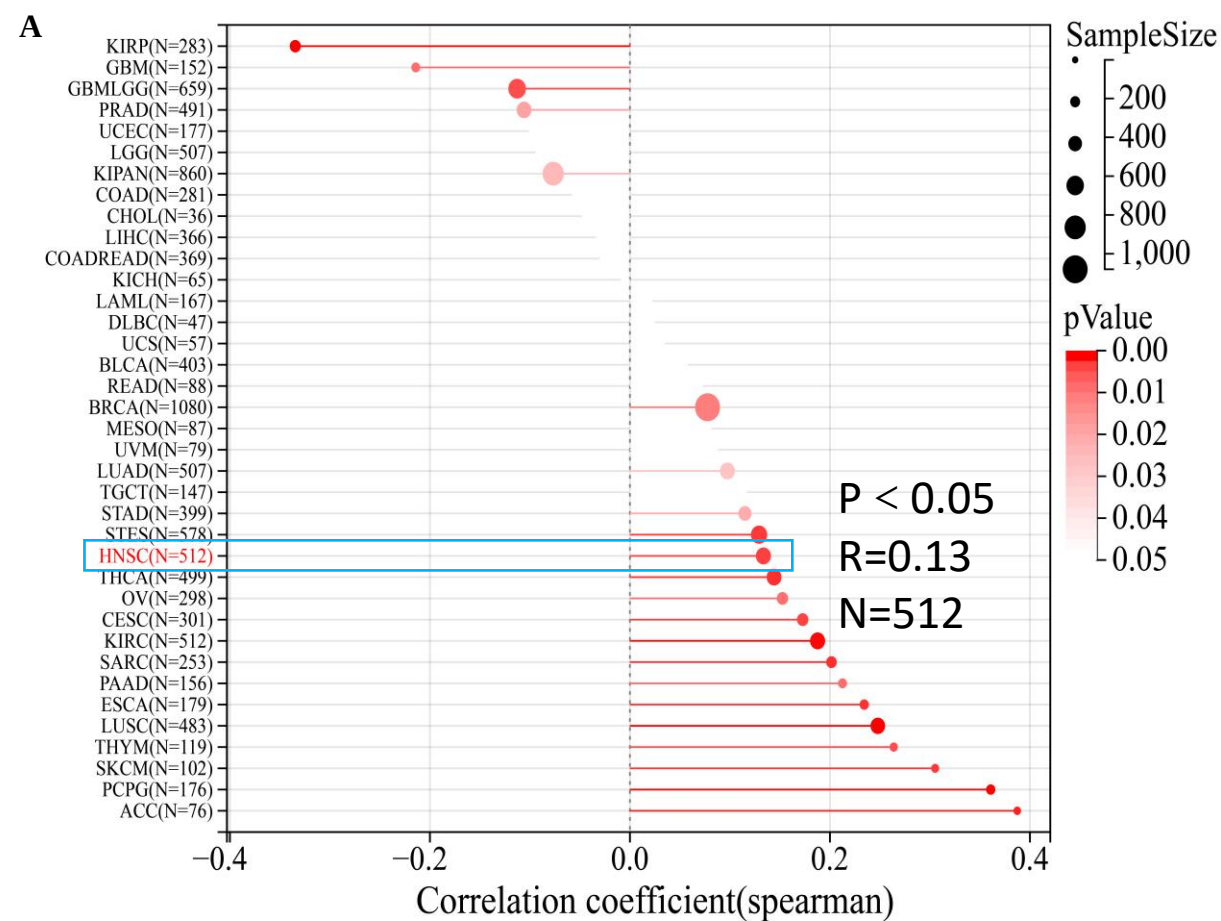
**Supplementary Fig. 1** A EphA2 was not associated with OSCC lymph node metastasis acquired from our tissue microarrays. Statistical analyses: one-way ANOVA. Data are presented as means  $\pm$  SDs. **B,C** EphA2 was not associated with HNSCC distant metastasis (B) and stage (C) acquired from SangerBox.

# Supplementary Fig. 2

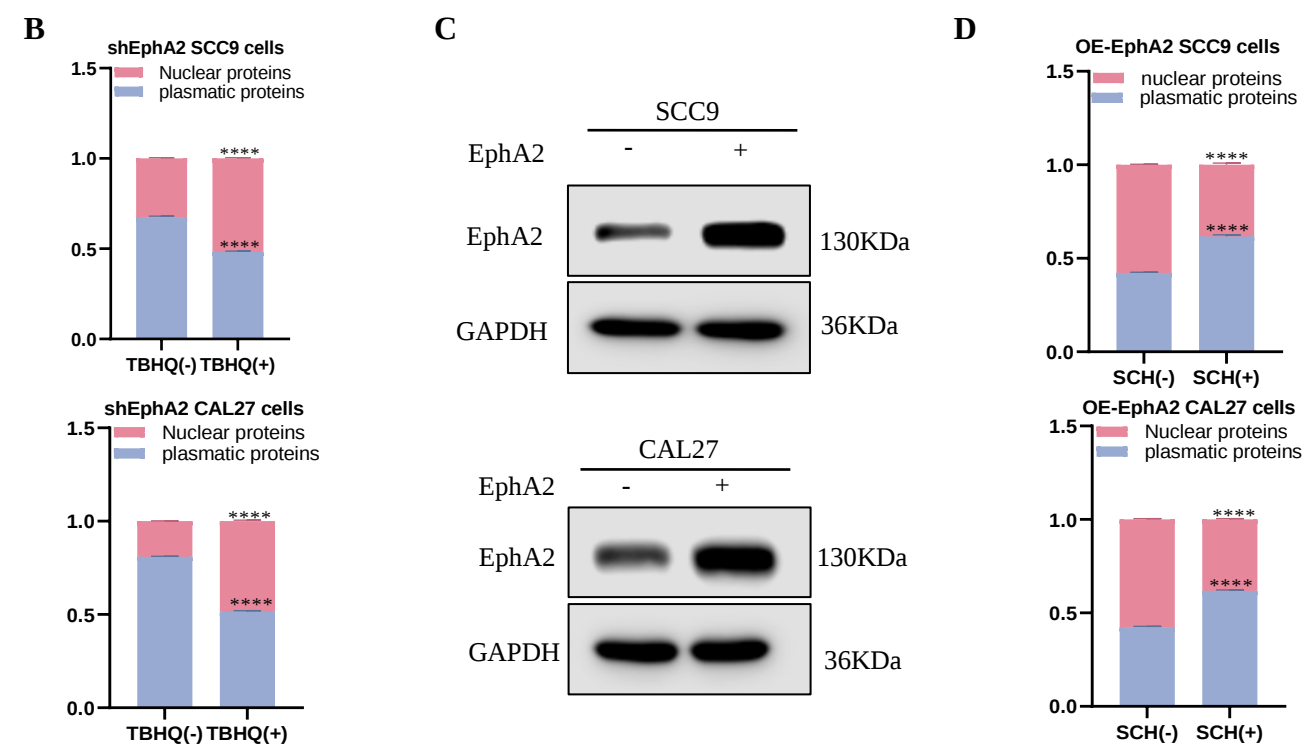
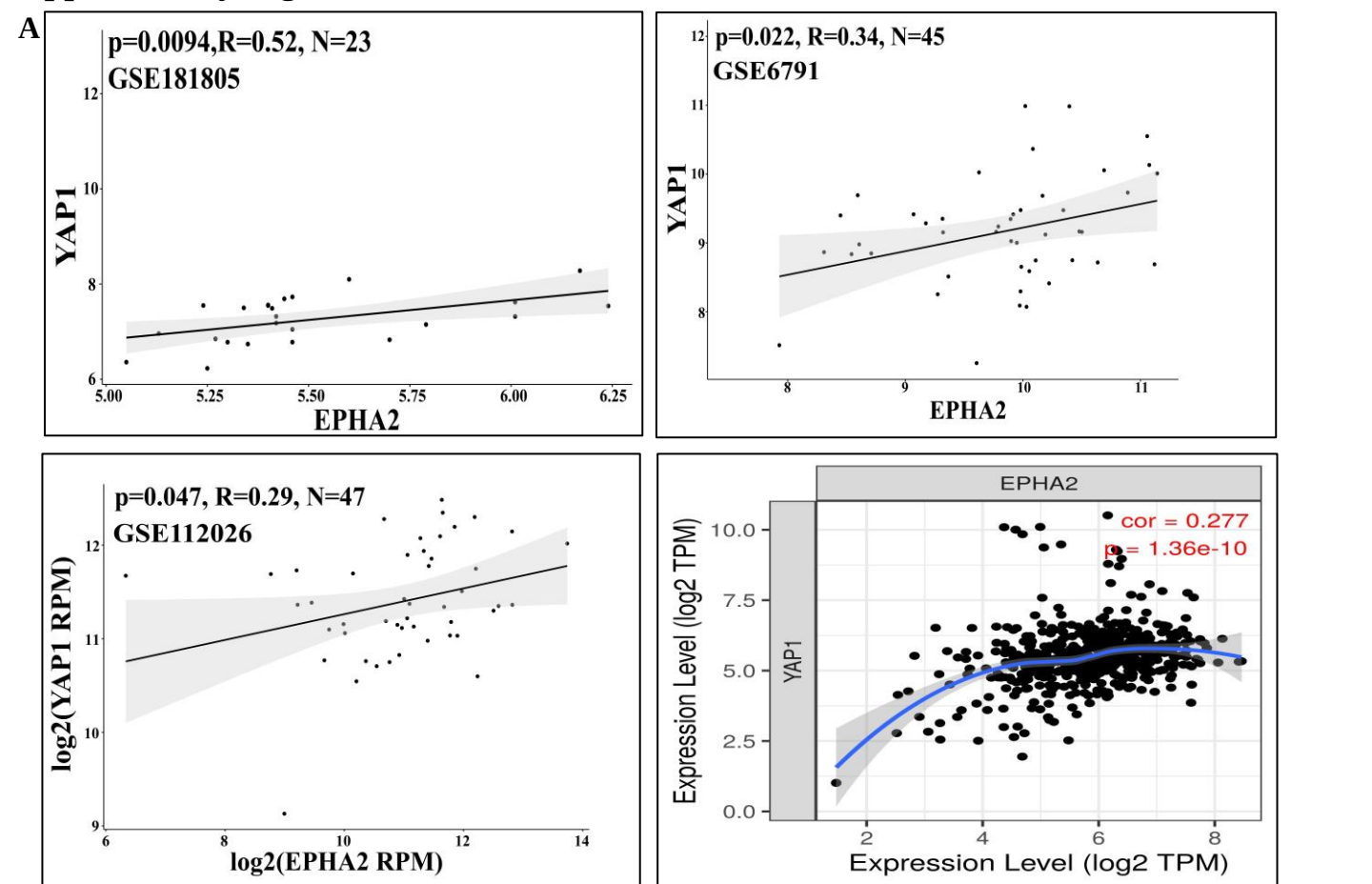


**Supplementary Fig. 2. A** sensitivity to cisplatin was increased in EphA2 knockdown cells. **B, C** The protein (B) and mRNA (C) expression of overexpression EphA2 in shEphA2-OSCC cells. **D** Overexpression of EphA2 enhanced the proliferation ability of OSCC cells. **E** Overexpression of EphA2 enhanced the sphere formation ability of OSCC cells. Scale bars: 100 $\mu\text{m}$ .

**Supplementary Fig. 3**

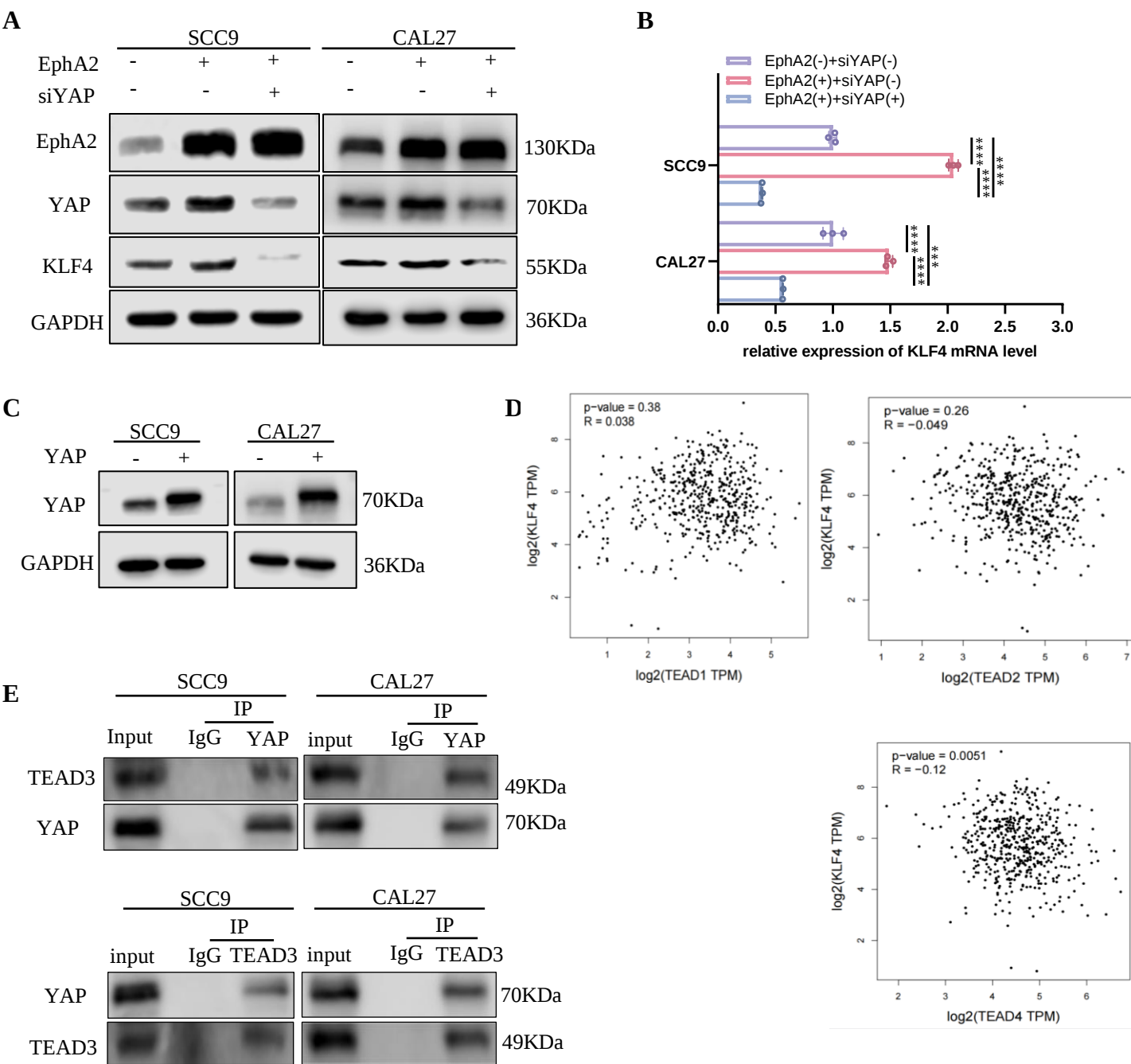


**Supplementary Fig. 3. A** The correlation between KLF4 and stemness scores in head and neck squamous cell carcinoma acquired from SangerBox website. **B** Sensitivity to cisplatin was increased in KLF4 knockdown cells.

**Supplementary Fig. 4**

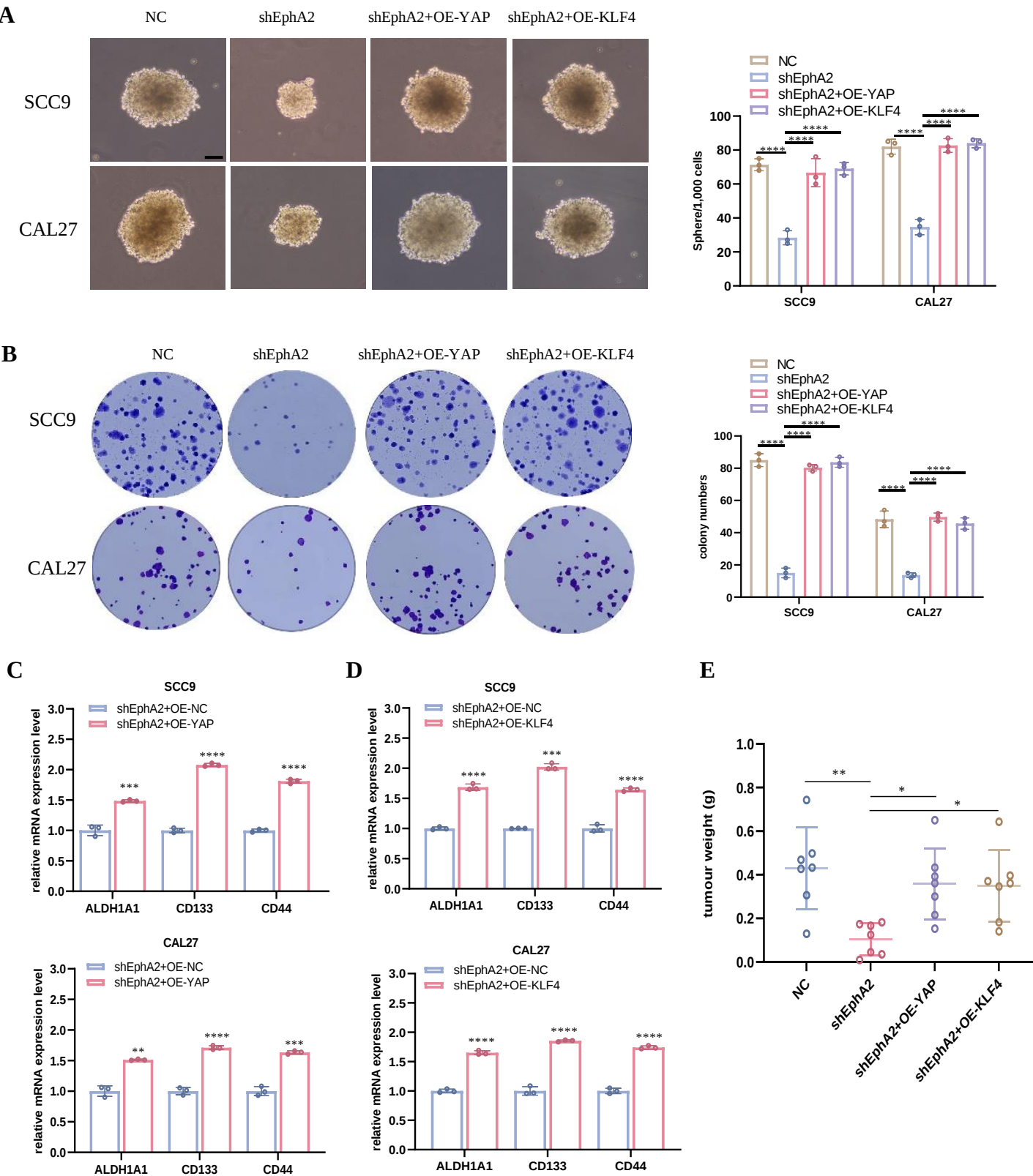
**Supplementary Fig. 4** **A** Different datasets show positive correlation between YAP and EphA2. From left to right, GSE181805 ( $r=0.52$ ,  $p=0.0094$ ), GSE6791 ( $r=0.34$ ,  $p=0.022$ ), GSE112026 ( $r=0.29$ ,  $p=0.047$ ) and the web tool of TIMER (<https://cistrome.shinyapps.io/timer/>). **B** The percentage of YAP in the nucleus increased after the use of ERK agonists (TBHQ, 5  $\mu$ M). **C** The effect of EphA2 overexpression. **D** Decreased YAP occupancy in the nucleus following the use of ERK inhibitors (SCH772984 (SCH), 10  $\mu$ M).

# Supplementary Fig. 5



**Supplementary Fig. 5. A, B** Knockdown of YAP reverses the promotion of KLF4 by OE-EphA2. **C** The effect of YAP overexpression. **D** The correlation among TEAD1, TEAD2 and TEAD4 with KLF4, which was acquired from GEPIA2. **E** The result of coimmunoprecipitation between YAP and TEAD3 in OSCC cells.

Supplementary Fig. 6



**Supplementary Fig. 6 A-D** Overexpression of YAP or KLF4 reversed the inhibitory effect of shEphA2 on the sphere-forming (A), and colony-forming abilities (B) and the expression of CSC-related markers (C, D). **E** Weights of tumors excised from the nude mice. n=7.

# Supplementary Table 1

| Patients | Type | Sex    | Age | Location                  | TNM     |
|----------|------|--------|-----|---------------------------|---------|
| 1        | OSCC | Male   | 53  | Buccal mucosa             | T2N0M0  |
| 2        | OSCC | Male   | 45  | Palate                    | T2N0M0  |
| 3        | OSCC | Male   | 72  | Buccal mucosa             | T2N0M0  |
| 4        | OSCC | Female | 57  | Tongue                    | T2N0M0  |
| 5        | OSCC | Male   | 55  | Tongue                    | T2N1M0  |
| 6        | OSCC | Male   | 52  | Tongue and Mouth floor    | T3N0M0  |
| 7        | OSCC | Female | 66  | Tongue                    | T2N0M0  |
| 8        | OSCC | Male   | 66  | Tongue and Mouth floor    | T4N2bM0 |
| 9        | OSCC | Male   | 43  | Tongue                    | T1N0M0  |
| 10       | OSCC | Female | 52  | Tongue                    | T2N0M0  |
| 11       | OSCC | Male   | 87  | Palate                    | T1N0M0  |
| 12       | OSCC | Male   | 47  | Tongue                    | T2N2M0  |
| 13       | OSCC | Female | 60  | Tongue                    | T1N0M0  |
| 14       | OSCC | Male   | 81  | Tongue                    | T2N2bM0 |
| 15       | OSCC | Male   | 51  | Tongue and Mouth floor    | T4N2cM0 |
| 16       | OSCC | Female | 41  | Buccal mucosa             | T4N0M0  |
| 17       | OSCC | Male   | 59  | Tongue                    | T3N1M0  |
| 18       | OSCC | Male   | 53  | Mouth floor               | T2N0M0  |
| 19       | OSCC | Female | 67  | Tongue                    | T2N2M0  |
| 20       | OSCC | Male   | 71  | Soft Palate               | T2N1M0  |
| 21       | OSCC | Female | 68  | Buccal mucosa             | T2N2M0  |
| 22       | OSCC | Male   | 54  | Tongue                    | T2N1M0  |
| 23       | OSCC | Male   | 54  | Lip                       | T2N1M0  |
| 24       | OSCC | Male   | 56  | Gingiva and Mouth floor   | T3N0M0  |
| 25       | OSCC | Male   | 80  | Gingiva                   | T4N0M0  |
| 26       | OSCC | Male   | 61  | Gingiva and Mouth floor   | T1N0M0  |
| 27       | OSCC | Male   | 60  | Mouth floor               | T4N2bM0 |
| 28       | OSCC | Male   | 56  | Tongue                    | T2N0M0  |
| 29       | OSCC | Female | 77  | Buccal mucosa and Gingiva | T4N2M0  |
| 30       | OSCC | Male   | 50  | Tongue                    | T1N0M0  |
| 31       | OSCC | Male   | 60  | Buccal mucosa             | T1N2M0  |
| 32       | OSCC | Female | 80  | Buccal mucosa             | T2N0M0  |
| 33       | OSCC | Male   | 53  | Tongue and Gingiva        | T3N1M0  |
| 34       | OSCC | Female | 80  | Gingiva                   | T2N0M0  |
| 35       | OSCC | Male   | 68  | Tongue                    | T2N0M0  |

| Patients | Type   | Sex    | Age | Location                  | TNM     |
|----------|--------|--------|-----|---------------------------|---------|
| 36       | OSCC   | Male   | 71  | Soft Palate               | T4N0M0  |
| 37       | OSCC   | Female | 54  | Tongue                    | T3N1M0  |
| 38       | OSCC   | Male   | 44  | Tongue                    | T4N2M0  |
| 39       | OSCC   | Female | 48  | Tongue                    | T2N1M0  |
| 40       | OSCC   | Male   | 47  | Tongue                    | T3N0M0  |
| 41       | OSCC   | Female | 71  | Buccal mucosa             | T2N1M0  |
| 42       | OSCC   | Male   | 45  | Palate                    | T4N0M0  |
| 43       | OSCC   | Male   | 64  | Tongue                    | T2N0M0  |
| 44       | OSCC   | Female | 70  | Gingiva                   | T2N0M0  |
| 45       | OSCC   | Male   | 60  | Buccal mucosa             | T3N2bM0 |
| 46       | OSCC   | Male   | 56  | Mouth floor               | T4N1M0  |
| 47       | OSCC   | Male   | 56  | Gingiva                   | T2N0M0  |
| 48       | OSCC   | Female | 55  | Lip                       | T2N0M0  |
| 49       | OSCC   | Male   | 69  | Soft Palate               | T2N0M0  |
| 50       | OSCC   | Male   | 62  | Gingiva                   | T4N0M0  |
| 51       | OSCC   | Male   | 55  | Tongue and Mouth floor    | T2N1M0  |
| 52       | OSCC   | Male   | 36  | Tongue                    | T2N0M0  |
| 53       | OSCC   | Female | 58  | Gingiva                   | T2N0M0  |
| 54       | OSCC   | Male   | 63  | Tongue                    | T2N0M0  |
| 55       | OSCC   | Male   | 76  | Tongue                    | T2N2cM0 |
| 56       | OSCC   | Female | 55  | Buccal mucosa and Gingiva | T3N0M0  |
| 57       | OSCC   | Male   | 52  | Tongue                    | T1N0M0  |
| 58       | OSCC   | Female | 34  | Tongue                    | T2N0M0  |
| 59       | Normal | Female | 59  | Tongue                    |         |
| 60       | Normal | Female | 43  | Tongue                    |         |
| 61       | Normal | Male   | 66  | Tongue                    |         |
| 62       | Normal | Male   | 77  | Buccal mucosa             |         |
| 63       | Normal | Male   | 67  | Tongue                    |         |
| 64       | Normal | Male   | 62  | Buccal mucosa             |         |
| 65       | Normal | Male   | 49  | Palate                    |         |
| 66       | Normal | Female | 85  | Tongue                    |         |
| 67       | Normal | Female | 59  | Tongue                    |         |
| 68       | Normal | Male   | 63  | Tongue                    |         |
| 69       | Normal | Male   | 62  | Gingiva                   |         |
| 70       | Normal | Male   | 76  | Tongue                    |         |

Supplementary Table 1. Patient information of tissue microarrays.

# Supplementary Table 2

| Patients | Type | Sex   | Age | Location               |
|----------|------|-------|-----|------------------------|
| 1        | OSCC | male  | 73  | Tongue                 |
| 2        | OSCC | male  | 70  | Tongue                 |
| 3        | OSCC | male  | 75  | Buccal mucosa          |
| 4        | OSCC | male  | 65  | Tongue                 |
| 5        | OSCC | male  | 53  | Tongue                 |
| 6        | OSCC | male  | 69  | Tongue and Mouth floor |
| 7        | OSCC | male  | 53  | Tongue                 |
| 8        | OSCC | male  | 50  | Mouth floor            |
| 9        | OSCC | femal | 57  | Buccal mucosa          |
| 10       | OSCC | male  | 48  | Buccal mucosa          |
| 11       | OSCC | male  | 70  | Tongue                 |
| 12       | OSCC | male  | 39  | Tongue                 |
| 13       | OSCC | femal | 76  | Gingiva                |
| 14       | OSCC | femal | 72  | Tongue                 |
| 15       | OSCC | male  | 55  | Tongue and Mouth floor |
| 16       | OSCC | male  | 82  | Tongue                 |

Supplementary Table 2. Patient information of Fig.1a (patient 1-8) and Fig.1b (patient 9-16)

Supplementary Table 3

|         | Forward (5'-3')         | Reverse (5'-3')         |
|---------|-------------------------|-------------------------|
| EphA2   | TGGCTCACACCCGTATG       | GTCGCCAGACATCACGTTG     |
| YAP     | TAGCCCTGCGTAGCCAGTTA    | TCATGCTTAGTCCACTGTCTGT  |
| KLF4    | ACCTACACAAAGAGTTCCCATC  | TGTGTTTACGGTAGTGCCTG    |
| SOX2    | GACCAGCTCGCAGACCTACAT   | ATGGAGCCAAGAGCCATGC     |
| OCT4    | ACATGTGTAAGCTGCGGCC     | GTTGTGCATAGTCGCTGCTTG   |
| c-MYC   | CCTGGTGCTCCATGAGGAGAC   | CAGACTCTGACCTTTTGCCAGG  |
| BM11    | CGTGTATTGTTTCGTTACCTGGA | TTCAGTAGTGGTCTGGTCTTGT  |
| NAONG   | GAAATACCTCAGCCTCCAGC    | GCGTCACACCATGTCTATTC    |
| CD133   | AGTCGGAAACTGGCAGATAGC   | GGTAGTGTGTACTGGGCCAAT   |
| ALDH1A1 | TGTTAGCTGATGCCGACTTG    | TTCTTAGCCCCTCAACACT     |
| CD44    | CTGCCGCTTTGCAGGTGTA     | CATTGTGGGCAAGGTGCTATT   |
| TEAD3   | ACAGGTGTCCAGCCACATAC    | CATGGCCTTGATGCCAACC     |
| GAPDH   | GGAGCGAGATCCCTCCAAAAT   | GGCTGTTGTCATACTTCTCATGG |

Supplementary Table 3. The primer sequence of RT-qPCR

Supplementary Table 4

|            | Forward (5'-3')        | Reverse (5'-3')        |
|------------|------------------------|------------------------|
| YAP-siNC   | UUCUCCGAACGUGUCACGUTT  | ACGUGACACGUUCGGAGAATT  |
| YAP-si1    | GCUCAGAUCCUUUCCUUAATT  | UUAAGGAAAGGAUCUGAGCTT  |
| YAP-si2    | CCUCACAGCAGAACCGUUUTT  | AAACGGUUCUGCUGUGAGGTT  |
| YAP-si3    | CCAAGUCUGCAGGAAGCUUTT  | AAGCUUCCUGCAGACUUGGTT  |
| TEAD3-siNC | UUCUCCGAACGUGUCACGUTT  | ACGUGACACGUUCGGAGAATT  |
| TEAD3-si1  | GCAAACAGGUGGUAGAGAATT  | UUCUCUACCACCUUGUUUGCTT |
| TEAD3-si2  | CCAGUGUCCUGCAGAACAAATT | UUGUUCUGCAGGACACUGGTT  |
| TEAD3-si3  | GGAAGACUCGGACGAGAAATT  | UUUCUCGUCCGAGUCUUCCTT  |

Supplementary Table 4. The sequence of RNA interference assay.

Supplementary Table 5

| site      | Forward (5'-3')           | Reverse (5'-3')           |
|-----------|---------------------------|---------------------------|
| 270~277   | CTAGGTTGACACCAGCCTAAGC    | GGTGTTIACTTTGGATTCCGGC    |
| 405~412   | AAAGTGATACCGAACACCACATTCA | CTCTCCCCTTGGTTTGTGATCAGTA |
| 1049~1056 | TGCCACTGCATTCCAGCTTG      | TTTCAACCAGCCATCTCGAAGC    |
| 1286~1293 | ATGCCTCTGAGTAAATCTTGAGCC  | GAGAAGGCCAGAGGAGTGTTC     |
| 1981~1988 | TIAGCTGCCATAGCAACGATG     | CGCGTTCCTTACTTATAACTTCCT  |

Supplementary Table 5. The primer sequence in ChIP-qPCR