Downregulation of miR-146b-5p via iodine involvement repressed papillary thyroid carcinoma cell proliferation

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Supplementary Figure 1 The expression of NIS is higher in high iodine areas than in adequate areas. (A) H&E showing the features of thyroid follicular cells and papillary thyroid carcinoma (20×); (B) RT-PCR was performed to determine the expression of miR-146b-5p in tissues. Relative levels were normalised to those of U6. #p < 0.05 vs Adequate iodine. (C) Expression of NIS in adequate iodine and high iodine areas (Immunohistochemistry, 20×).

Supplementary Figure 2 10^{-3} M NaI promotes Smad4 expression via miR-146b-5p repression in PTC cells. Luciferase assay showed that miR-146b-5p could target Smad4 with or without 10^{-3} M NaI treatment. n = 3, #p < 0.05.

Supplementary Figure 3 10^{-3} M NaI reduces viability and proliferation in PTC cells. (A) MTT assay, (B) Cell counting tests determined cell proliferation and viability at 24, 48, 72, and 96 h. n = 3. Data are presented as the mean ± SD of three independent experiments. *p < 0.0167 vs. con.

Supplementary Figure 4 10^{-3} M NaI promotes p21 and p57 expression in PTC cells. Western blot analysis for p21 and p57 in cells treated with 10^{-3} M NaI. Data are presented as means ± SD of three independent experiments. #p < 0.05 vs con.

Supplementary Figure 5 10^{-3} M NaI promotes p21, p27, p57 and cyclin D1 expression in PTC tissues. RT-PCR was performed to determine the expression of p21, p27, p57 and cyclin D1. GAPDH was used as an internal control. Data are presented as means ± SD of three independent experiments. #p < 0.05 vs Adequate iodine.
**Supplementary Figure 6** $10^{-3}$ M NaI upregulates thyroid-specific genes in PTC cells. (A and C) The mRNA and protein expression of TG in TPC-1 cells. No detectable levels of NIS, TSHR and TPO were observed in TPC-1 cells. (B and D) The mRNA and protein expression of TG and TSHR in BCPAP cells. No detectable levels of NIS and TPO were observed in BCPAP cells. Data are presented as means ± SD of three independent experiments. # $p < 0.05$ vs Control.

**Supplementary Figure 7** $10^{-3}$ M NaI inhibits proliferation in PTC cells via miR-146b-5p. MTT assays determined cell proliferation at 24, 48, and 72 h. n = 3. Data are presented as the mean ± SD of three independent experiments. # $p < 0.05$ vs con, ## $p < 0.05$ vs NaI.