Figure S1. Characterization of plasma-derived EVs isolated from healthy controls and HNSCC patients. Western blotting revealed the expression of the transmembrane protein (Flotillin-1, CD81), cytosolic protein (TSG101) and negative marker (Cytochrome c), the specific and negative markers for EVs in isolated EVs of healthy controls and HNSCC patients.
**Figure S2.** Receiver-operator characteristic (ROC) curve of miR-27b-3p, miR-491-5p, miR-1910-5p, miR-630 and 2-miRNAs-panel models in HNSCC patients (N=73) and healthy controls (N=20). The sensitivity, specificity, and AUC (areas under the curve) values are shown.
**Figure S3.** The correlation between proposed miRNAs models and clinical characteristics using Mann-Whitney and Kruskal-Wallis test. (A-F) The correlation between each miRNA model with clinical parameters, clinical stages, age, sex, smoking status, P16 status, and primary tumor sites. (A) miRNA-491-5p; (B) miRNA-27b-3p; (C) miRNA-1910-5p; (D) miR-630; (E) 2miR-panel.
Figure S4. Receiver-operator characteristic (ROC) curve of ΔmiR-27b-3p, ΔmiR-491-5p, ΔmiR1910-5p, ΔmiR-630, and Δ2-miRNAs-panel. The AUC, sensitivity, and specificity represent the prognostic performance of ΔmiRNAs to discriminate between patients with tumor recurrence and no sign of disease in 1 year.
**Figure S5.** The expression level of miR-491-5p in tissue samples from TCGA HNSCC patients and normal. Y-axis is Log2 transformed and represented as reads per million miRNAs mapped (RPM) value. Significance was determined with the Mann-Whitney U-test.