



Aspirin use and the role of PIK3CA RNA expression as a biomarker in head and neck cancers



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Abstract

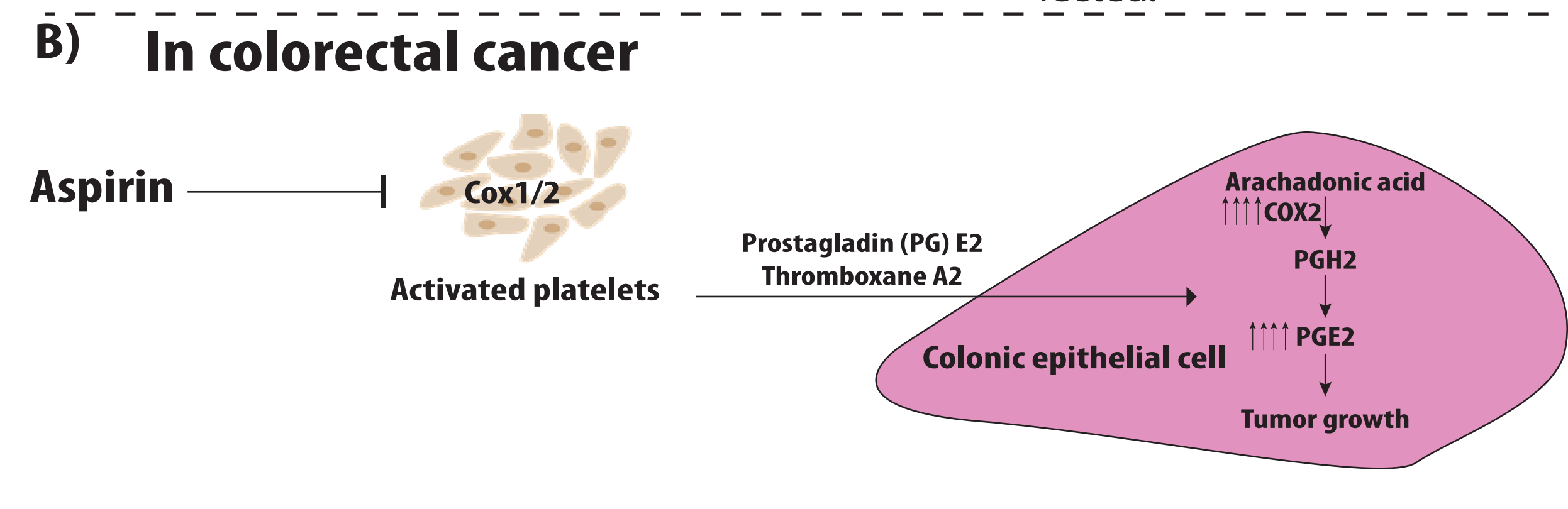
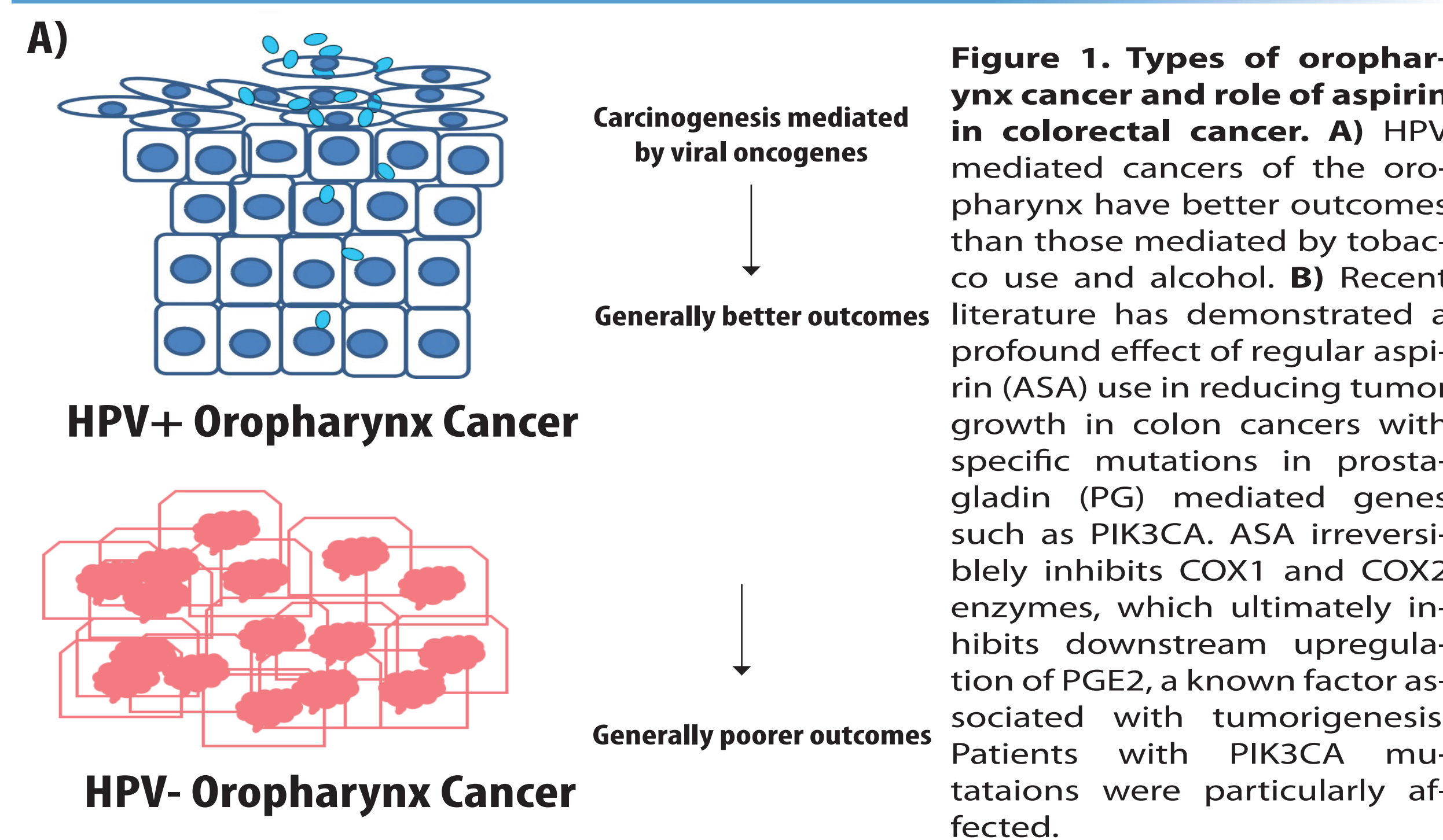
Objectives: Non-steroidal anti-inflammatory medications, particularly aspirin, are associated with a chemoprotective effect in various cancers. Regular aspirin use has been shown to have a survival benefit in patients with PIK3CA mutations in the setting of colon cancer and more recently certain head and neck malignancies. However, the potential survival benefit of aspirin during and after a diagnosis of head and neck squamous cell cancer (HNSCC) remains controversial. The objective of our study was to determine the association between aspirin (ASA) use, PIK3CA RNA expression and outcome determination in HNSCC.

Methods: This was a retrospective cohort study of 170 patients with HNSCC treated at the University of North Carolina between 2007 and 2015 who had tissue available for processing. Quality RNA was isolated from 126/170 samples using a commercial nucleic acid extraction kit with on-column DNAase treatment to obtain the RNA expression level of PIK3CA. Charts were reviewed for pathologic and treatment data, overall survival (OS) and disease-free survival (DFS). All charts were queried for ASA use within 1 year of diagnosis or at any point during follow-up.

Results: A cohort of 126 patients with HNSCC had quality RNA after tissue processing. A total of 109/126 patients (86.4%) were HPV-positive. Unadjusted OS and DFS at 3 years were unchanged for ASA users vs non-users. However, when controlled for HPV status, HPV-negative ASA users have a marked survival benefit at 3 years. Interestingly, the expression of PIK3CA RNA was significantly higher in HPV-positive tumors ($p=0.02$) and the quantity of PIK3CA RNA had a negative correlation with both 3yr DFS ($p=0.0001$) and OS ($p=0.001$), regardless of ASA use.

Conclusions: ASA use has been shown to portend an improved disease specific and overall survival in other solid tumors in specific patient populations with certain tumor genetics, specifically those with PIK3CA mutations. Our data suggest that select HNSCC patients with higher tumor RNA expression levels of PIK3CA may have a reduction in OS and DFS that is aborted with NSAID use. These data could highlight a possible biomarker for selecting patients who are appropriate for ASA therapy following a diagnosis of HNSCC, specifically those that are HPV negative.

Background



Methods

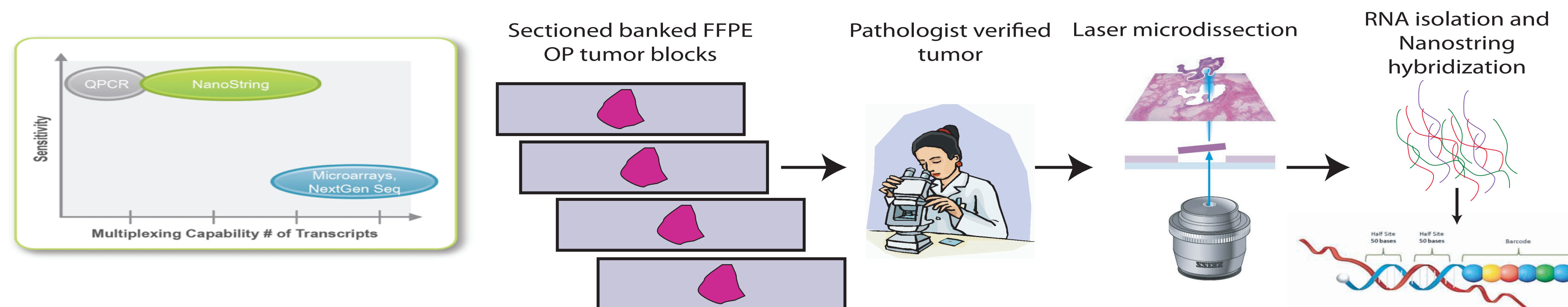


Figure 2. Using NanoString RNA assays to capture gene expression data. NanoString RNA analysis utilizes high sensitivity (similar to qPCR) detection to count RNA transcript quantities, while doing these counts in parallel with up to 770 targets (giving microarray-like throughput). In our study, we isolated FFPE-based tumors, had pathologic verification of tumor on sectioned slides, had known tumor dissected and then RNA isolated from harvested tissue. We then performed NanoString based capture of the RNA to determine levels of many transcripts, including PIK3CA/B

Results

Table 1. General Patient Demographics

Variable	N	Mean	Lower 95%CI	Upper 95%CI
Age	104	61.86	59.94	63.79
Race (% Non-white)	104	8.7	3.2	14.1
Sex (% Female)	104	7.7	2.5	12.9
Age at Dx	104	56.79	54.99	58.59
OS from dx to censure (months)	104	70.93	61.48	80.39
DFS time from dx (months)	104	63.05	53.74	72.36
% ASA started within 1 year	104	29.8	20.9	38.8
% ASA started anytime	104	39.4	29.9	49.0
% Other NSAID use	104	8.7	3.2	14.2

Figure 4. 3- and 5-year overall survival in ASA users

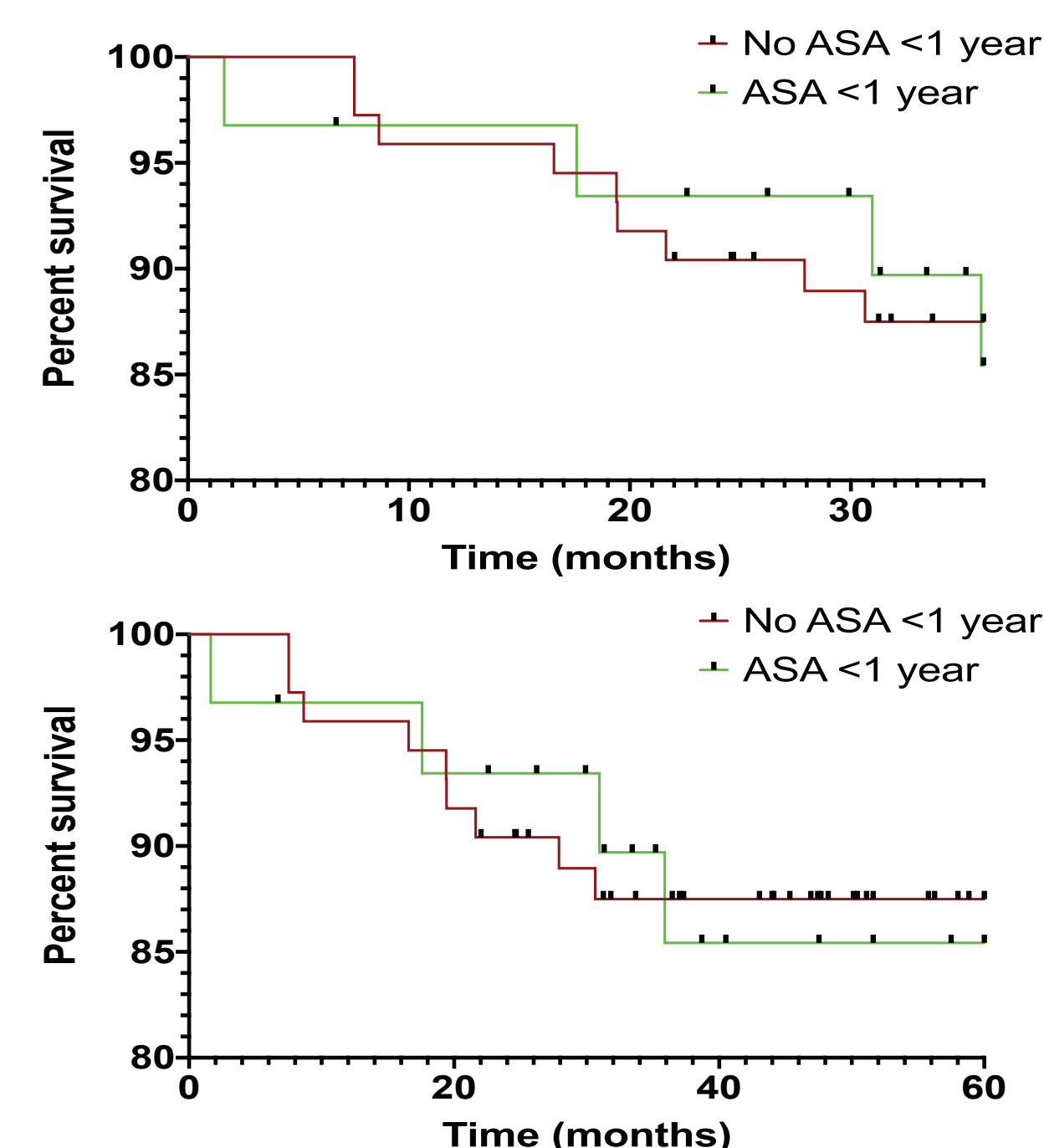


Figure 3. Relative RNA levels of PIK3CA to PIK3CB

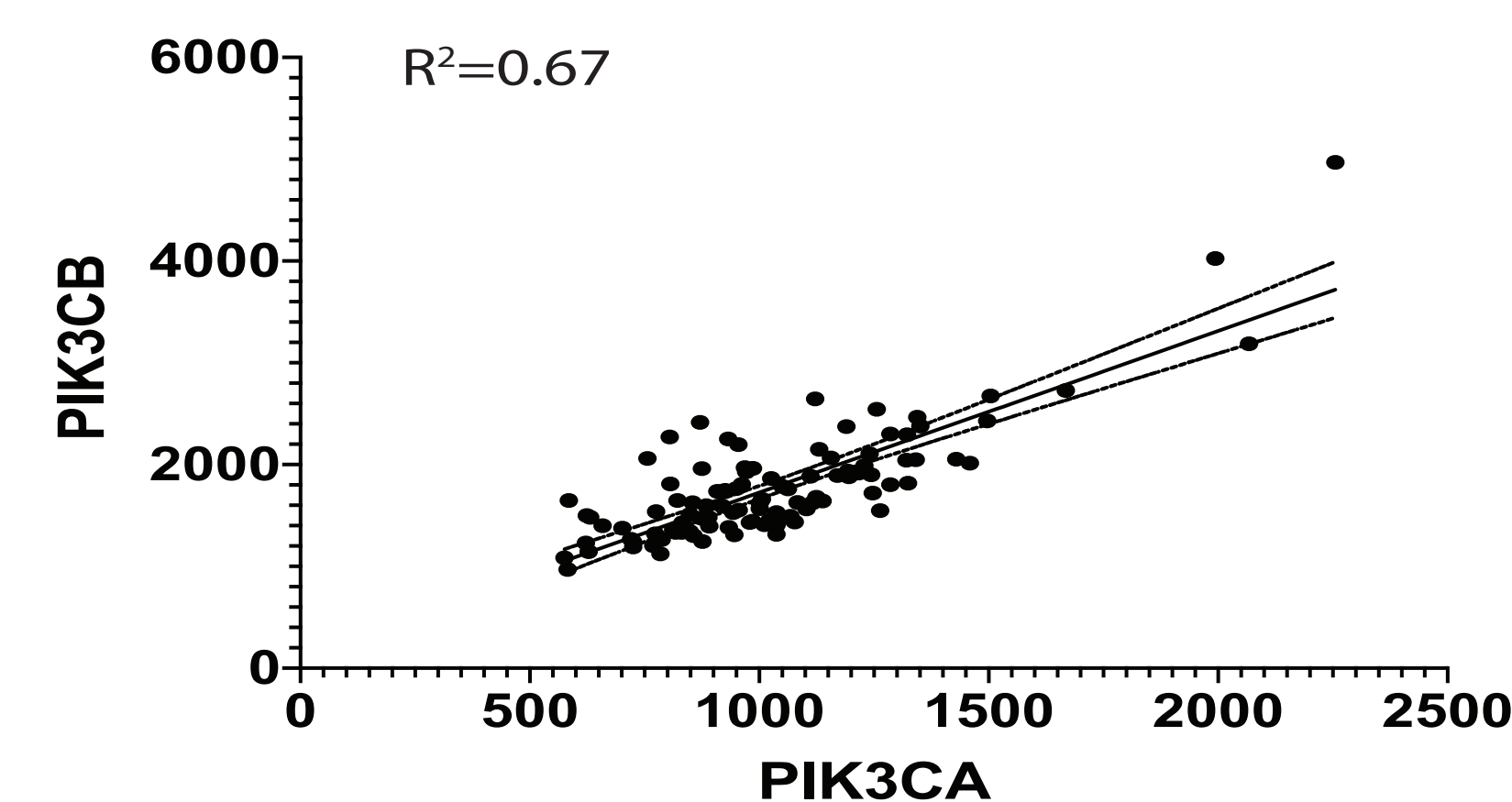


Figure 5. 3- and 5-year DFS in ASA users

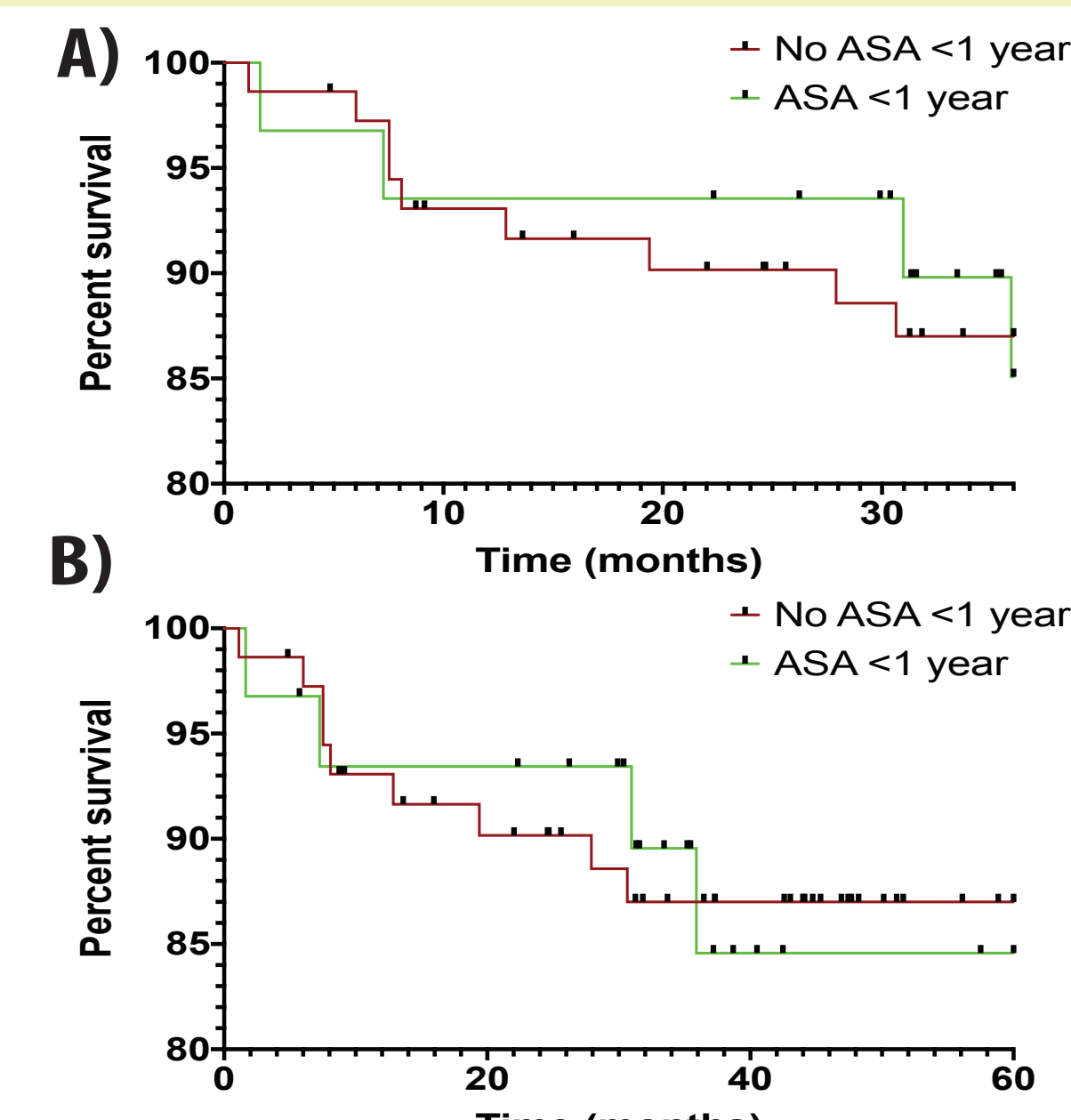


Figure 6. HPV-negative OPSCC has increased overall survival at 36 months with ASA use

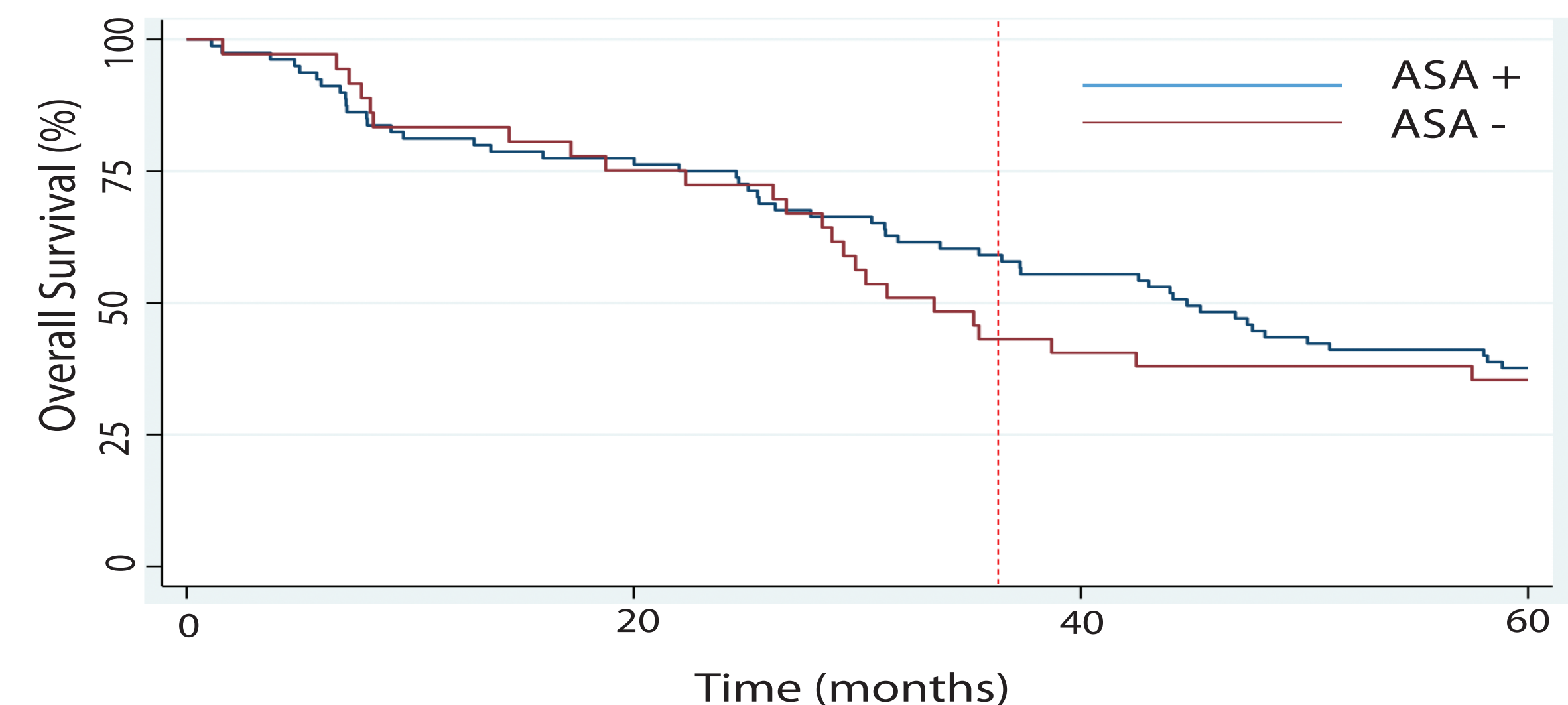
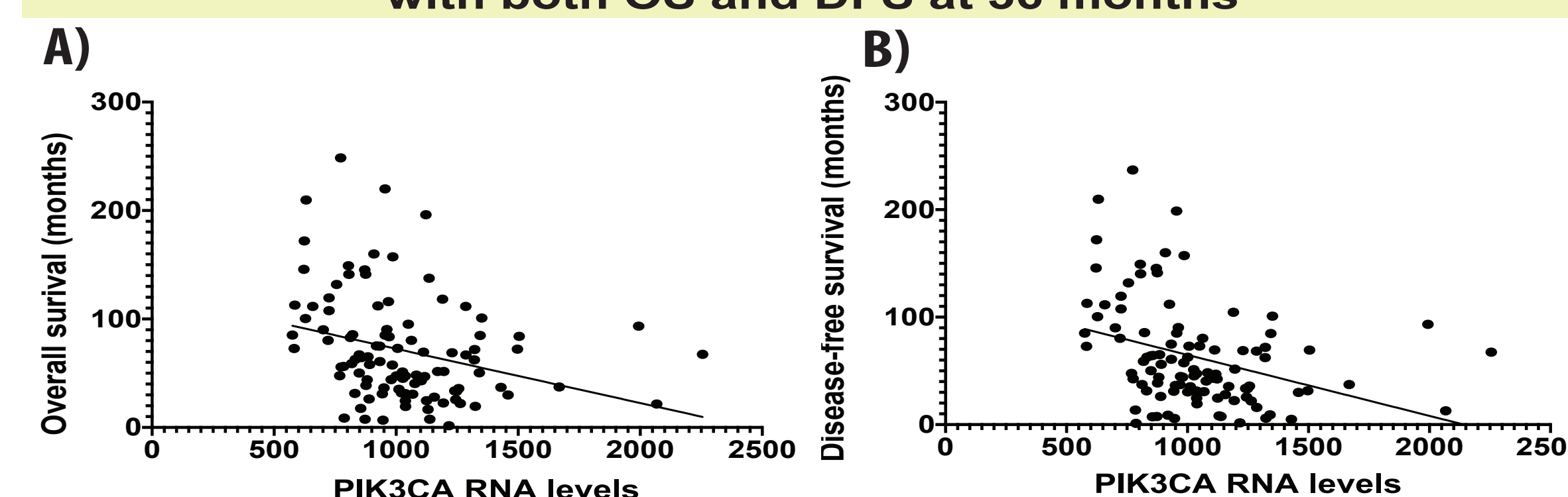


Figure 7. Increasing PIK3CA expression has negative correlation with both OS and DFS at 36 months



Conclusions

- 1.) Multiple isoforms of PIK3C are expressed in OPSCC.
- 2.) ASA use has no apparent effect in patients with HPV+ OPSCC, and given these are the majority of tumors, the positive effect of ASA use could be missed in HPV-negative tumors.
- 3.) When controlled for HPV status, HPV-negative OPSCC have a significantly increased OS/DFS at 36 months compared to HPV-positive OPSCC.
- 4.) Higher PIK3CA levels are correlated with decreased overall survival and decreased disease free survival.