

# Peripubertal Nutritional Prevention of Cancer-Associated Gene Expression and Phenotypes

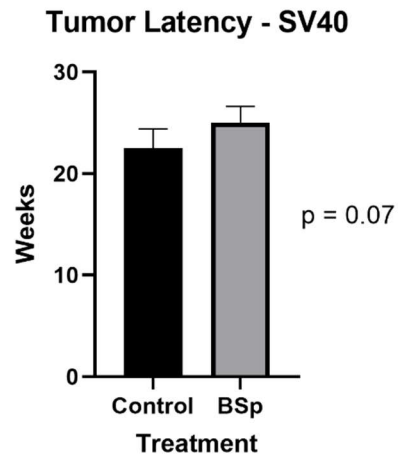
Andrew Brane, Itika Arora and Trygve O. Tollefsbol

**Table S1.** Primer sequences for RT-qPCR.

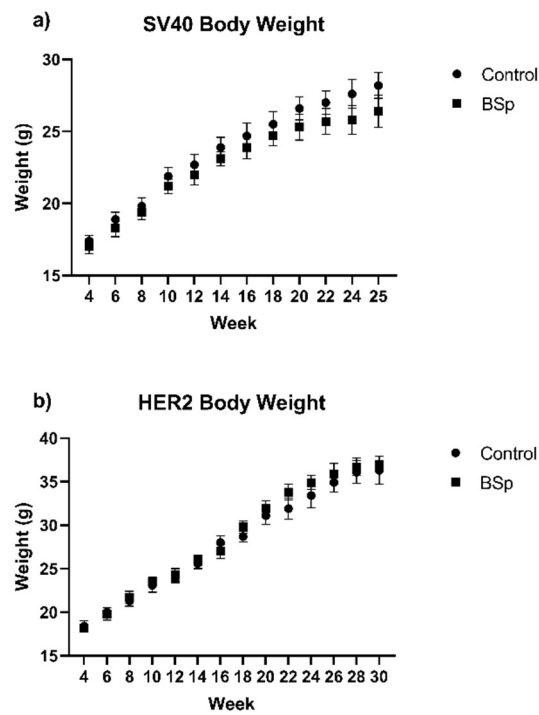
GAPDH	GTGGAGTCATACTGGAACATGTAG AATGGTGAAGGTCGGTGTG
P53	TGAAAATGTCTCCTGGCTCAG CTAGCATTGAGGCCCTCATC
P21	GAAGAGACAACGGCACACT CAGATCCACAGCGATATCCAG
BRCA2	TGTGTCATCCCTCTCCAGTATC GATGCCTAAACCCAGAAAGAGT
Erich4	CCCCATCTCCAGTCCCA TCAGGCCCTTATGTCTTCAG
Lman1I	ATCAAAGTAGATCCCGATGCC GAGATGCAGATGAGAGTGACTG
Clec4e	TGAGAGCTGCGATATGTTACG ATCCCACCACACAGAGAGA
Parp6	GCATACTTCATAATCTGGACAAGG GTAGAGGTGTTTGGCTATCCC
Chrdl2	TCTCAGTTGTCTGTCTTTGC AGTGTGTCCTGTGTAGCTGTA
Pcsk1	TCACAGTTATCTCCCTGACGA CCCAGAAGGCATTGAATATGG
Slc51b	GCTGCTTCTTTGATTCTGTT GCTTGGTATTTTCGTGCAGA

**Table S2.** Antibodies for Western blots (File S1).

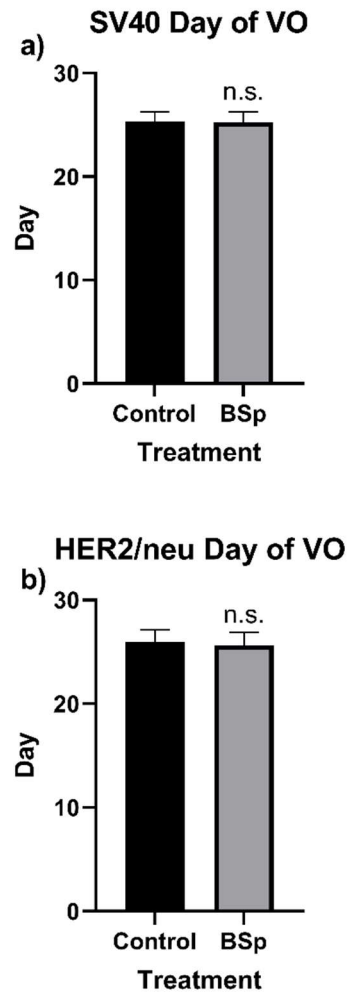
P53	Anti-p53 antibody (ab131442)
P21	Recombinant Anti-p21 antibody [EPR18021] (ab188224)
BRCA2	Anti-BRCA2 antibody (ab216972)
ActB	$\beta$ -Actin (13E5) Rabbit mAb #4970



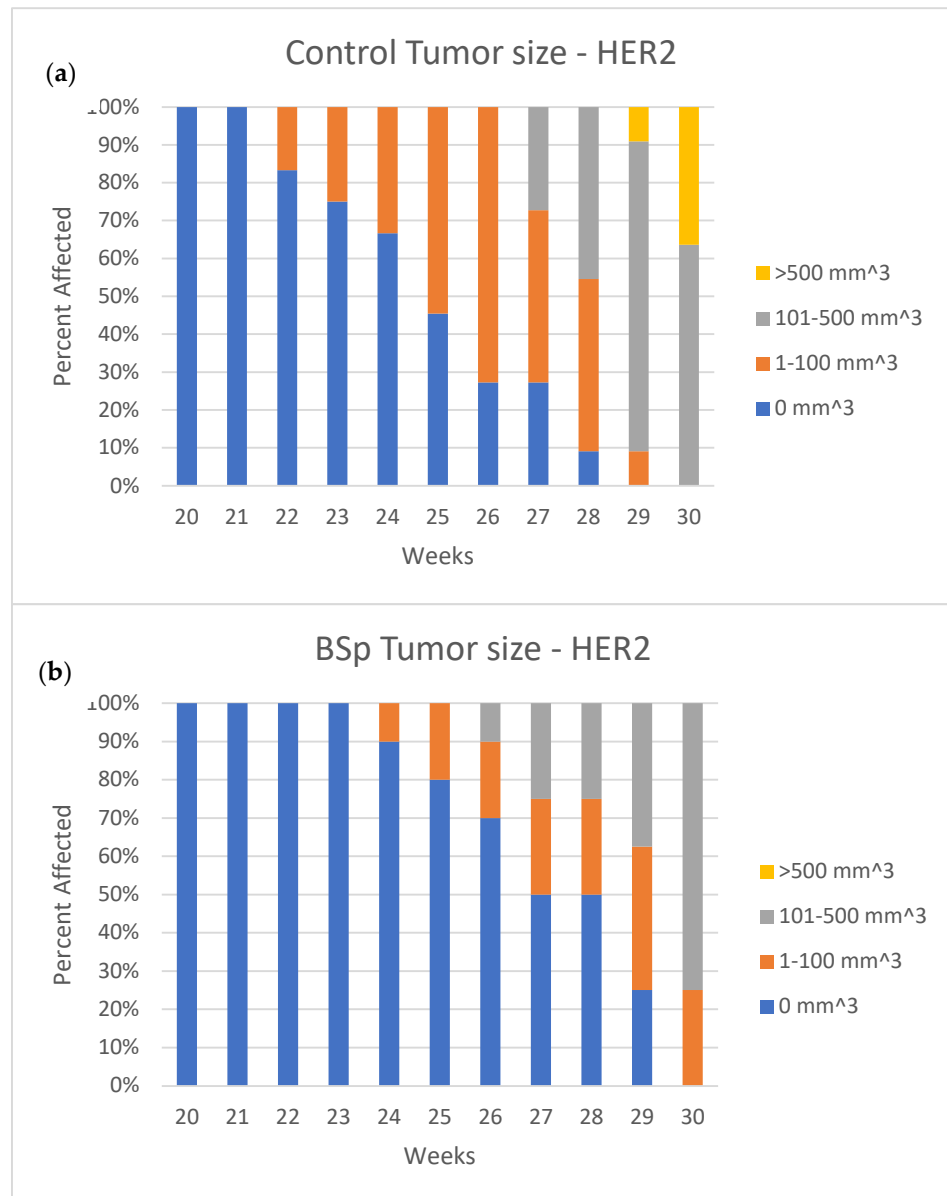
**Figure S1.** Average tumor latency observed in SV40 mice. BSp-treated mice developed tumors approximately 2 weeks later than controls in this mouse model.  $p = 0.07$  and  $n = 24$ .



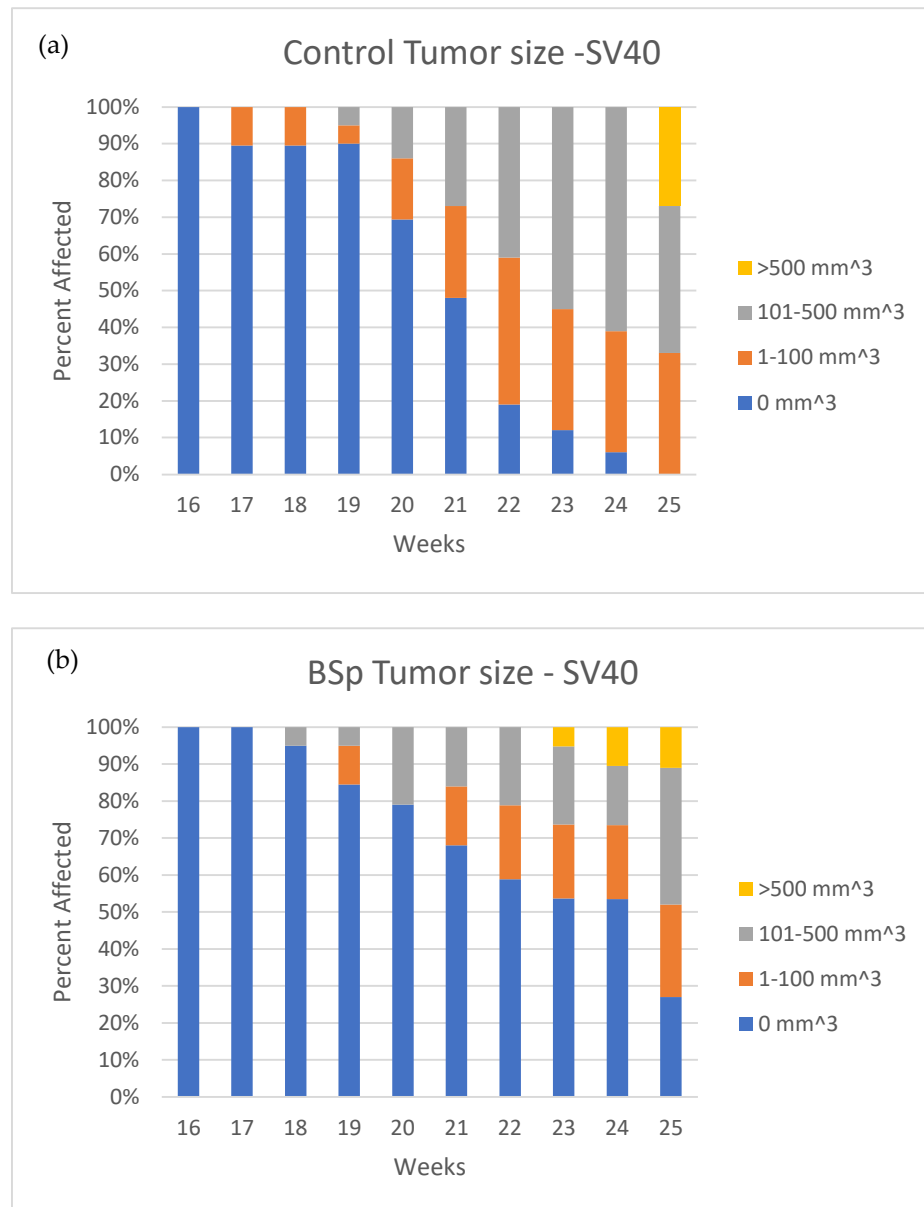
**Figure S2.** Mouse body weight for SV40 (a) and HER2/neu (b) mice. Treatment did not have a significant effect on total body weight in either mouse strain.  $n = 24$  for both strains.



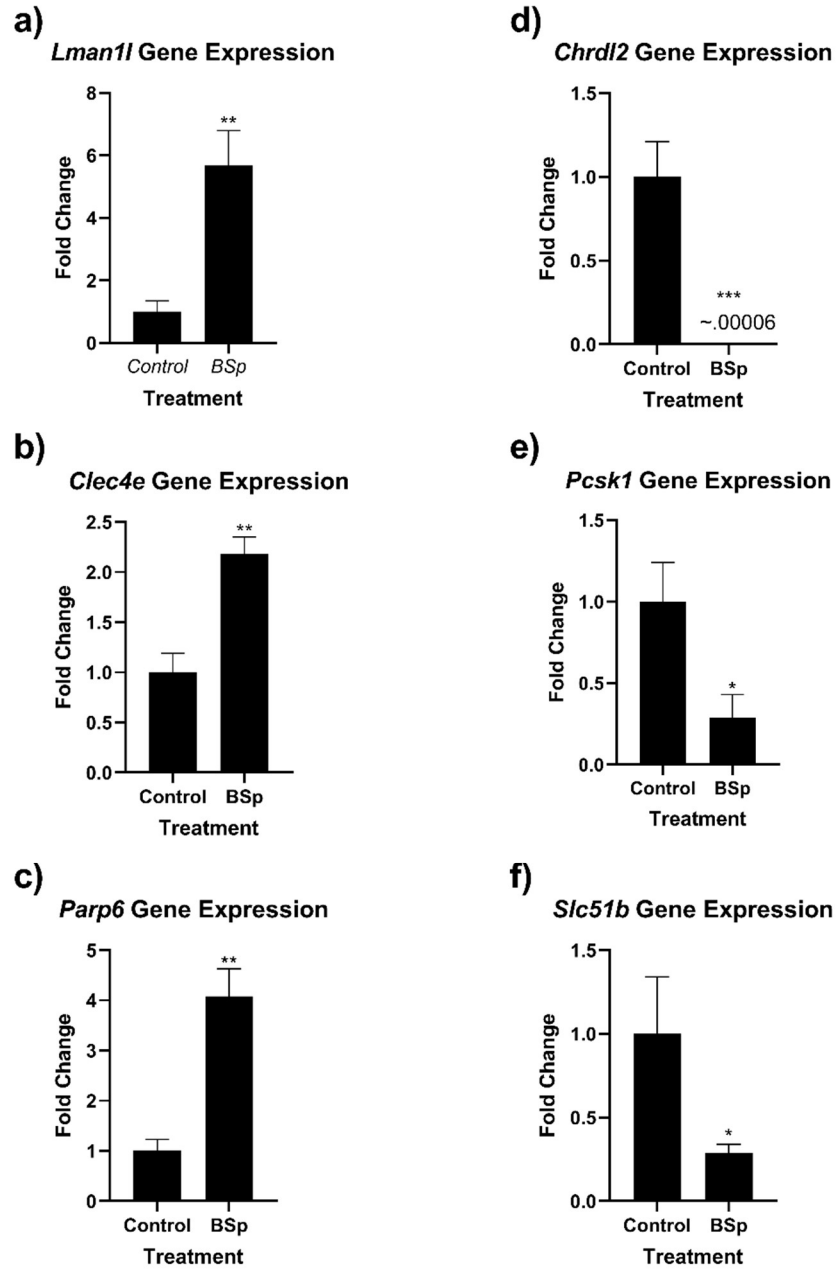
**Figure S3.** Average day of vaginal opening (VO) for SV40 (**a**) and HER2/neu (**b**) mice. Treatment did not have a significant effect on VO in either mouse strain.  $n = 24$  for both strains. n.s., not significant.



**Figure S4.** Approximate tumor size for HER2/neu mice in both control (a) and BSp (b) fed mice.  $n = 24$  for both groups.

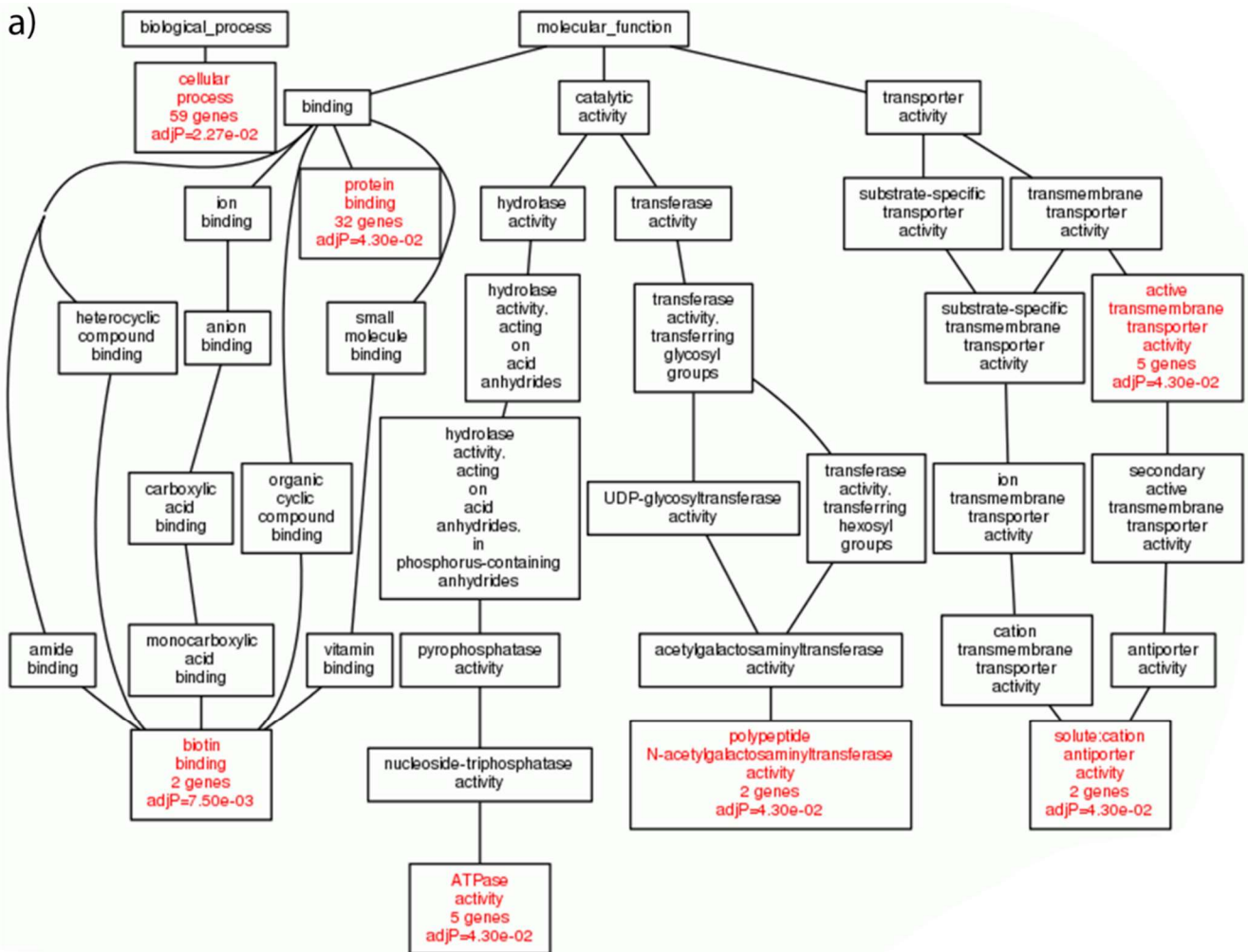


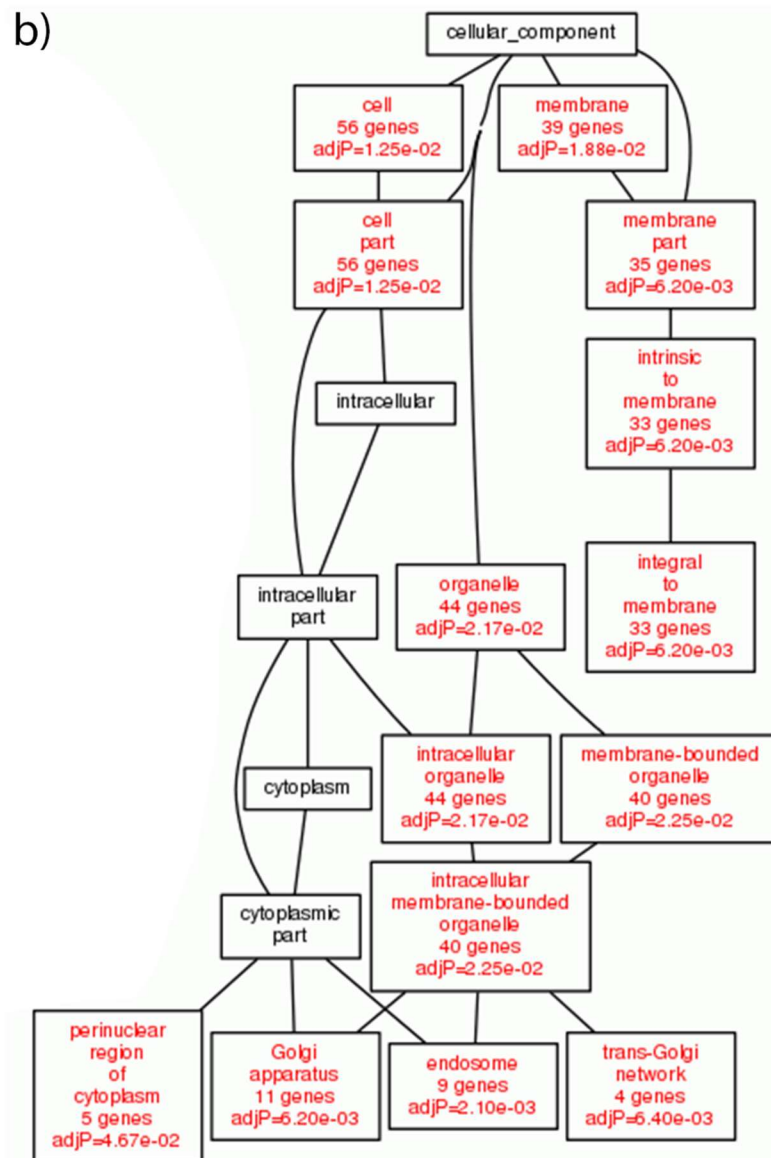
**Figure S5.** Approximate tumor size for SV40 mice in both control (a) and BSp (b) fed mice.  $n = 24$  for both groups.



**Figure S6.** PCR verification of tumor suppressors *Lman1l* (a), *Clec4e* (b), *Parp6* (c), as well as oncogenes *Chrdl2* (d), *Pcsk1* (e), and *Slc51b* (f).  $n = 10$  for all RT-qPCR. \* =  $p < 0.05$ , \*\* =  $p < 0.01$ , \*\*\* =  $p < 0.001$ .

a)





**Figure S7.** Gene ontology analysis for downregulated genes identified by RNA sequencing. Overall, there were 21 significantly upregulated groups of genes within the overarching groups of biological processes (a), molecular function (a), and cellular components (b). Significant groupings of genes are identified in red. P-value cutoff for gene ontology analysis was  $p < 0.05$ .



a)

