

Supporting Information for

Benzalkonium chloride disinfectants induce apoptosis, inhibit proliferation, and activate the integrated stress response in a 3-D *in vitro* model of neurodevelopment

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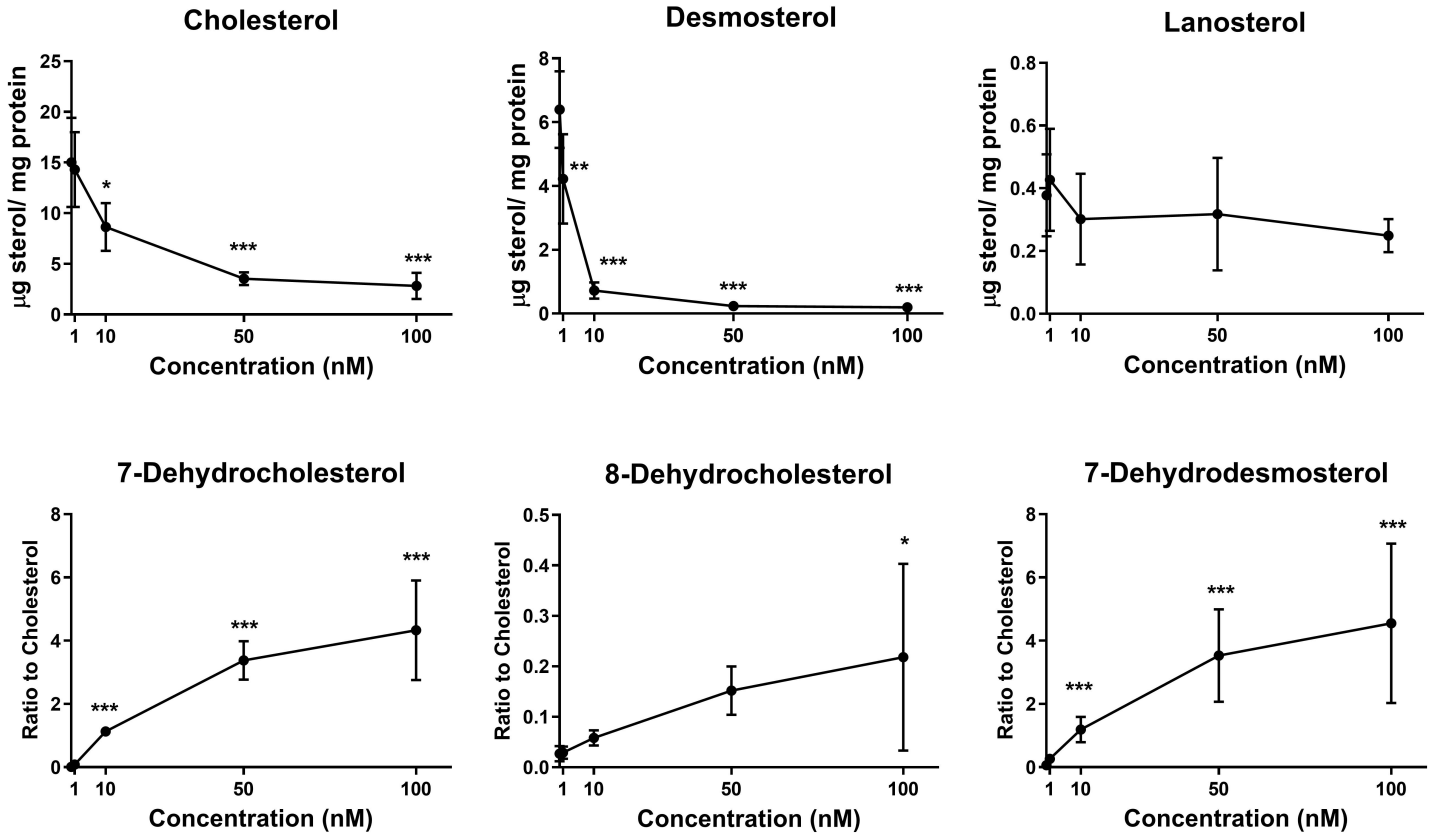


Figure S1. Inhibition of cholesterol biosynthesis by AY9944 in neurospheres. Neurospheres exposed to vehicle control (0 nM) or AY9944 at 1, 10, 50, and 100 nM from DIV 4 to DIV 7 show alterations in levels of sterols in the post-squalene cholesterol biosynthetic pathway. N = 4 biological replicates per condition. Adjusted *P* value: *, *P* < 0.05; **, *P* < 0.01; ***, *P* < 0.001 All adjusted *P*-value < 0.05.

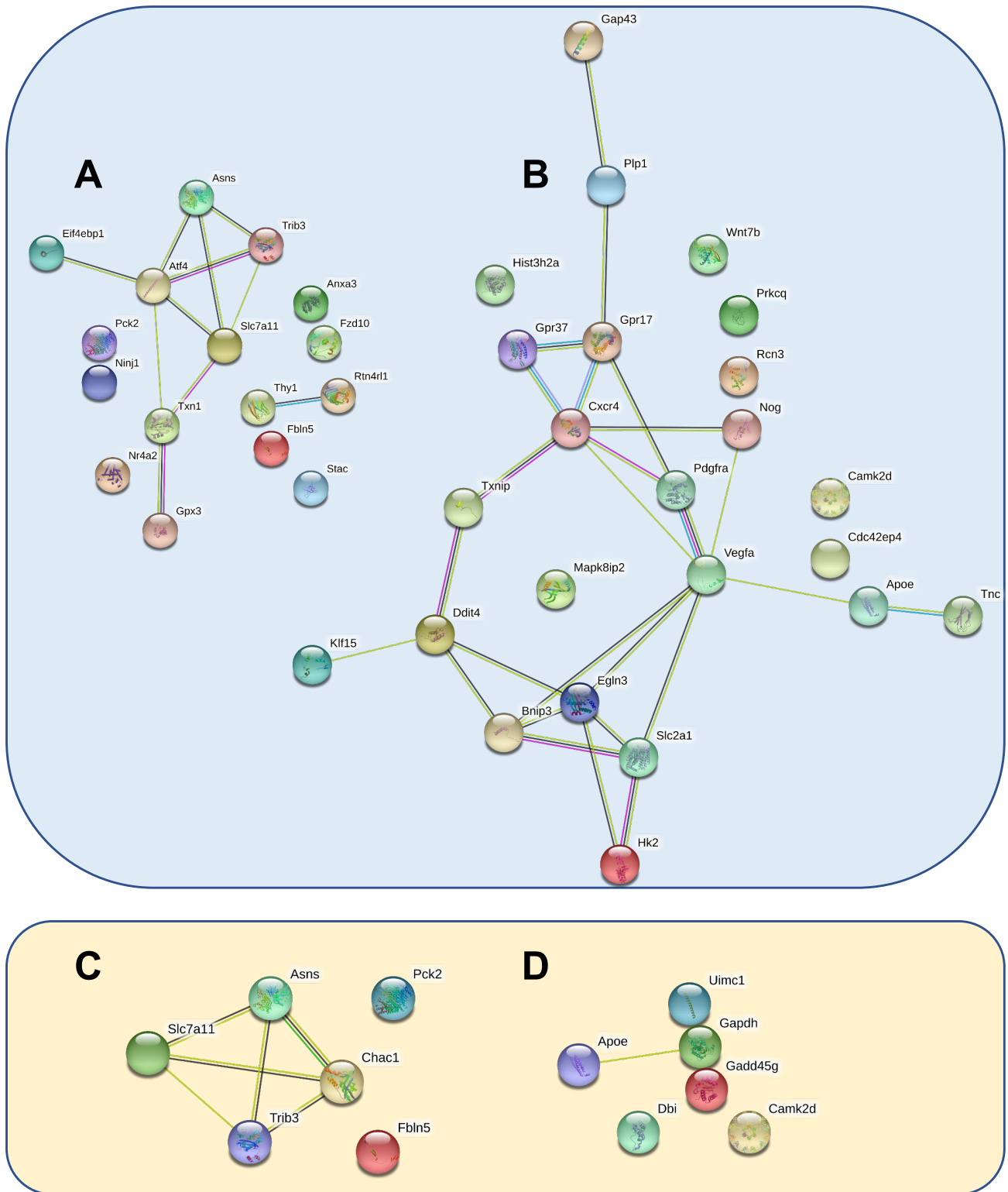


Figure S2. STRING networks of DEGs involved in the integrated stress response. Top: STRING networks of DEGs upregulated (A) or downregulated (B) in response to BAC C12 exposure. Bottom: STRING networks of DEGs upregulated (C) or downregulated (D) in response to BAC C16 exposure. n=3 biological replicates per condition. All adjusted P-value < 0.05.

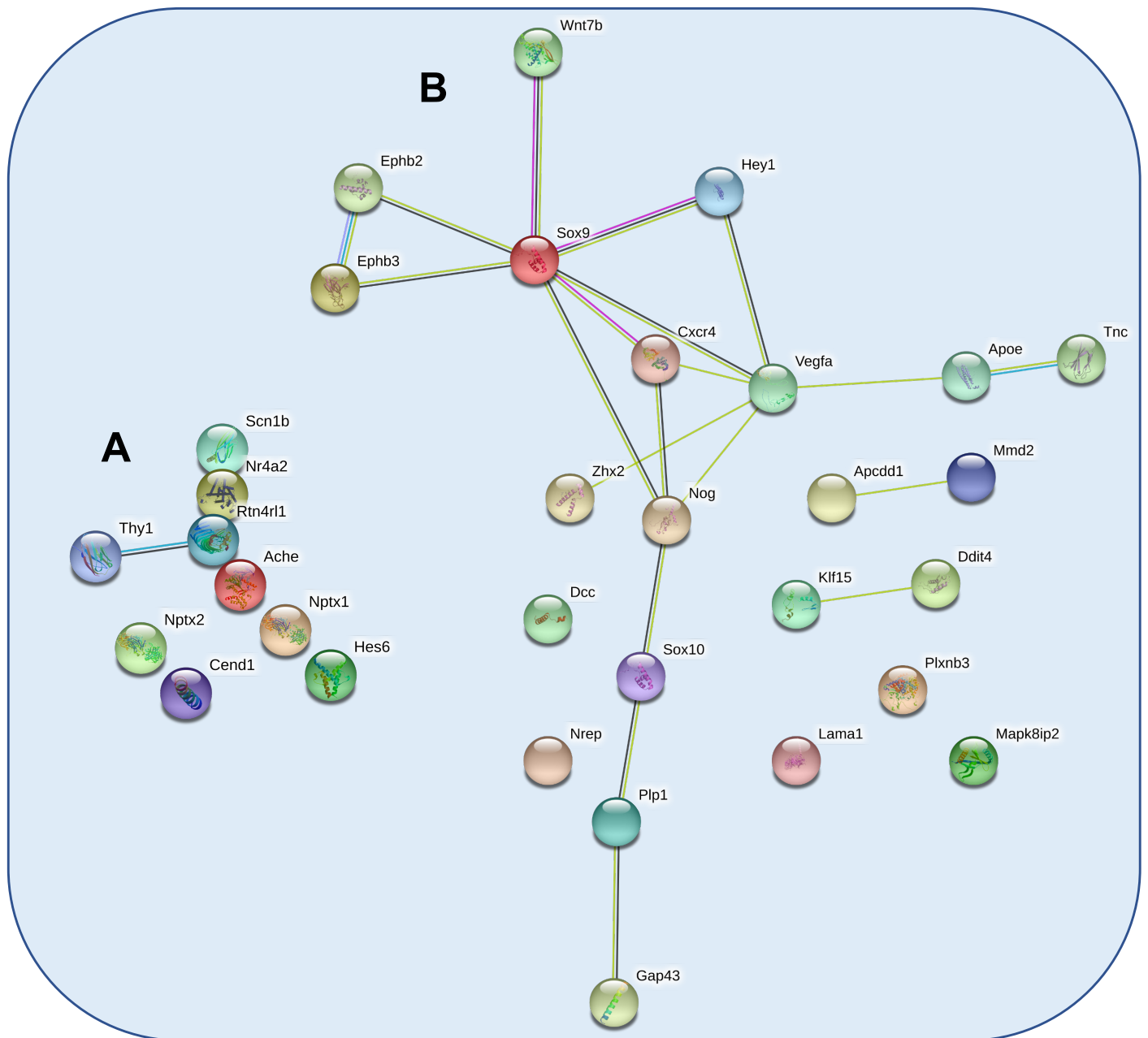


Figure S3. DEGs from BAC C12-exposed neurospheres involved in neurogenesis. (A) DEGs involved in the GO biological process of neurogenesis that are upregulated in response to BAC C12 exposure. (B) DEGs involved in the GO biological process of neurogenesis that are downregulated in response to BAC C12 exposure. n=3 biological replicates per condition. All adjusted P-values < 0.05.

Table S1. Top 10 biological processes altered by both BACs.

GO Biological Process	BAC C12 vs. Control			BAC C16 vs. Control		
	# DEGs	# Total	-log <i>P</i> -value	# DEGs	# Total	-log <i>P</i> -value
response to stress	41	2214	15.84	12	2205	6.44
phosphate-containing compound metabolic process	18	674	13.29	5	665	4.84
Phosphorylation	15	508	13.29	4	499	4.38
cell death	26	1523	8.29	8	1508	4.34
programmed cell death	25	1453	8.16	8	1446	4.65
apoptotic process	21	1216	7.06	7	1201	4.54
regulation of transport	31	2084	6.67	10	2066	4.39
small molecule metabolic process	22	1337	6.55	8	1331	5.28
chemical homeostasis	21	1308	5.94	8	1301	5.46
MAPK cascade	19	1193	5.37	11	1178	12.29