

## SUPPLEMENTAL DATA

# Sulfamethoxazole (SMX) Alters Immune and Apoptotic Endpoints in Developing Zebrafish (*Danio rerio*)

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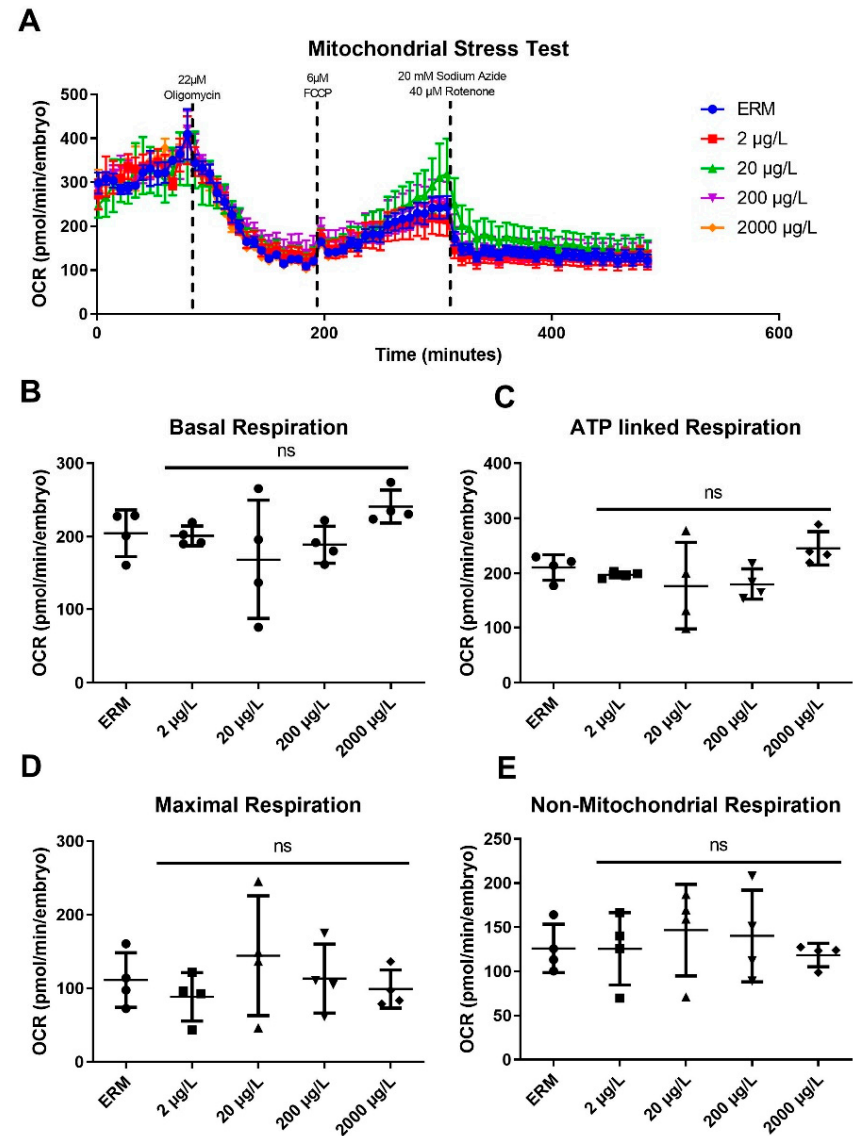
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**Supplemental Table S1.** Primers used for real-time PCR analysis.

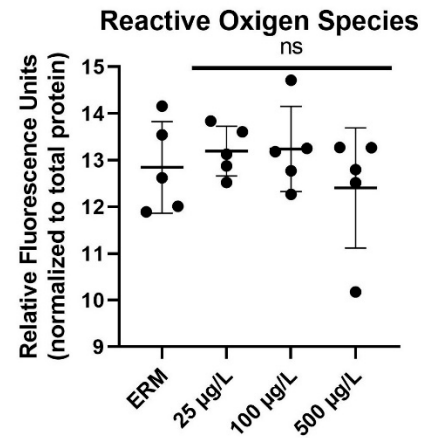
Gene name	Gene Symbol	Forward (5' to 3')	Reverse (5' to 3')	Reference
Acetylcholinesterase	<i>ache</i>	GCTAATGAGCAAAAGCATGTGGGC	TATCTGTGATGTTAAGCAGACGAGGCA	NM_131846.2
BCL2 Apoptosis Regulator	<i>bcl-2</i>	TCACTCGTTCAGACCCTCAT	ACGCTTTCCACGCACAT	Deng et al. 2009
BCL2 Associated Agonist Of Cell Death	<i>bad</i>	CAAGCCTGGATAAACAC	GGCAGATTGAAAGAAAG	Lu et al. 2011
BCL2 Associated X, Apoptosis Regulator	<i>bax</i>	GGCTATTTCAACCAGGGTTCC	TGCGAATCACCAATGCTGT	Liu et al., 2021
Beta-actin	<i>bactin</i>	CGAGCAGGAGATGGGAACC	CAACGGAAACGCTCATTGC	Wang et al. 2018
Caspase 3	<i>casp 3</i>	CCGCTGCCCATCACTA	ATCCTTTTCACGACCATCT	Deng et al. 2009
Caspase 7	<i>casp 7</i>	TTCGAGTCCTGGTCGGAAGA	CCTTGCTGCCATCCTGTAA	MG957999.1
Caspase 9	<i>casp 9</i>	ACCTCAATGGCCAGAACTGTC	TCCTCCAGCACACGATCAAG	Deng et al. 2009
Catalase	<i>cat</i>	CTCCTGATGTGGCCCGATAC	TCAGATGCCC GGCCATATTC	Sarkar et al., 2014
chemokine (C-X-C motif) ligand 18b	<i>Cxcl-C1c</i>	GGCATTACACCCAAAGCG	GCGAGCACGATTCACGAGAG	Stockhammer et al., 2009
Glutathione peroxidase 1a	<i>alias gpx1a</i>	CACCCTCTGTTTGCGTTCC	CTCTTTAATATCAGCATCA	Wu et al., 2016
Heat shock protein 70	<i>hsp70</i>	GAAGACGGCATCTTTGAGGTGA	GGGCCCTCTTGTTCTGACTGAT	Hahn et al., 2014
Interferon Gamma	<i>ifng</i>	GAATGGCTTGGCCGATACAGGATA	TCCTCCACCTTTGACTTGTCATC	Jin et al., 2010
Interleukin 17	<i>il-17a</i>	CGAGAGCCTGTATCCTAC	CGTAATCCTGGACCTCAA	Zhang et al., 2014

Interleukin 1b	<i>IL-1β</i>	TGGACTTCGCAGCACAAAATG	GTTCACTTCACGCTCTTGGATG	Watzke et al., 2007
Ribosomal 18s	<i>rps18</i>	TCGCTAGTTGGCATCGTTTATG	CGGAGGTTCTGAAGACGATCA	Wang et al. 2018
Superoxide dismutase 1	<i>sod1 (Cu/Zn SOD)</i>	CAACACAAACGGCTGCATCA	TTTGCAACACCACTGGCATC	Sarkar et al., 2014
Superoxide dismutase 2	<i>sod2 (Mn SOD)</i>	AGCGTGACTTTGGCTCATTT	ATGAGACCTGTGGTCCCTTG	Sarkar et al., 2014
Glutathione S-transferase	<i>gst</i>	CTATACATGCGGCGAAGCT	GGCATTGCTCTGGACGAT	Mukhopadhyay et al., 2014
Nuclear factor erythroid 2-related factor 2	<i>nrf2a</i>	CCAGATCCAATCGTGGAGTTT	GAAGGATCCGTCTTCGGTTATG	Awoyemi et al., 2019



**Supplemental Figure S1.** Mitochondrial bioenergetics in 54 hpf zebrafish embryos. (A) Oxygen consumption rate (OCR); (B) basal respiration;

(C) ATP-linked respiration; (D) maximal respiration; (E) non-mitochondrial respiration. Data are reported as mean  $\pm$  SD (one-way ANOVA followed by Dunnett's multiple comparisons test,  $p > 0.05$ ).



**Supplemental Figure S2.** Reactive oxygen species (ROS) in zebrafish embryos exposed to sulfamethoxazole for 7 days expressed as relative fluorescence units ( $\mu\text{g}/\text{mL}$  protein). Horizontal line represents mean value of the group ( $\pm\text{SD}$ ) (ANOVA followed by a Dunnett's test,  $n=5$  per treatment).

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