Data Article

Lipid quantitation and metabolomics data from vitamin E-deficient and -sufficient zebrafish embryos from 0 to 120 hours-post-fertilization

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The data herein is in support of our research article by McDougall et al. (2017) [1], in which we used our zebrafish model of embryonic vitamin E (VitE) deficiency to study the consequences of VitE deficiency during development. Adult 5D wild-type zebrafish (Danio rerio), fed defined diets without (E–) or with VitE (E+, 500 mg RRR-α-tocopheryl acetate/kg diet), were spawned to obtain E– and E+ embryos that we evaluated using metabolomics and specific lipid analyses (each measure at 24, 48, 72, 120 hours-post-fertilization, hpf), neurobehavioral development (locomotor responses at 96 hpf), and rescue strategies. Rescues were attempted using micro-injection into the yolksac using VitE (as a phospholipid emulsion containing d6-α-tocopherol at 0 hpf) or D-glucose (in saline at 24 hpf).

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### Value of the data

- Fatty acid quantification and peroxidation data during zebrafish embryonic development in E- vs. E+ zebrafish embryos may be used by other researchers to investigate antioxidant effects of VitE with respect to specific lipids.
- The metabolomics dataset may be utilized by other researchers to investigate the secondary metabolic effects of VitE deficiency.
- Rescue studies using microinjection into the yolksac may be compared to other methods of compound/nutrient delivery to developing zebrafish.

### 1. Data

**Fig. 1.** shows data from quantitative analyses of LA (linoleic acid, 18:2, omega-6); ARA (arachidonic acid, 20:4, omega-6); EPA (eicosapentaenoic acid, 20:5, omega-3); DHA (docosahexaenoic acid, 22:6, omega-3) in fatty acid extracts from samples with and without alcoholic saponification of E- and E+ embryos collected at 24, 48, 72, and 120 hpf. **Tables 1** and 2 provide detailed targeted metabolomics datasets for E- and E+ embryos collected at 24, 48, 72, and 120 hpf. Relative response intensity metabolomics data for choline and methylation pathway intermediates in E- and E+ embryos are shown in **Fig. 2.** Relative response intensities of antioxidant network components from metabolomic analyses, as well as quantification of α-tocopherol and ascorbic acid, in E- and E+ embryos (pmol/embryo) are shown in **Fig. 3.** Relative response intensities of glycolytic and tricarboxylic acid cycle intermediates in E- and E+ embryos are shown in **Fig. 4.** Relative response intensities of free saturated fatty acids and coenzyme A from metabolomics data in E- and E+ embryo are shown in **Fig. 5.** **Fig. 6** shows locomotor activity data from E- and E+ embryos micro-injected into the yolksac at 0 hpf.
with either saline or a VitE-emulsion. Fig. 7 shows locomotor activity data from E− and E+ embryos micro-injected into the yolksac at 24 hpf with either saline or D-glucose.

2. Experimental design, materials and methods

2.1. Study design

All experiments (i.e. lipid quantifications, targeted metabolomics analyses, and micro-injection rescue studies) were performed in duplicate and have been reported in detail [1].

2.2. Zebrafish husbandry and diets

The Institutional Animal Care and Use Committee of Oregon State University approved this protocol (ACUP Number: 4344). Tropical 5D strain zebrafish were housed in the Sinnhuber Aquatic Research Laboratory and complete details of the housing and husbandry have been reported [1].

2.3. Analyses

Diet and embryo α-tocopherol [2] and ascorbic acid [3] were determined using high-pressure liquid chromatography with electrochemical detection as reported [1].

Extraction and sample preparation for metabolomic analysis were performed following 24, 48, 72, and 120 hpf, embryos (n = 15 per replicate, n = 4 replicates per group), as described [1]. Chromatography was performed with a Shimadzu Nexera system (Shimadzu; Columbia, MD, USA) coupled to a high-resolution hybrid quadrupole–time–of-flight mass spectrometer (TripleTOF® 5600; SCIEX; Framingham, MA, USA). Two different LC analyses using reverse phase and HILIC columns were used, as described [1].

Analysis of total DHA, EPA, ARA, and LA were performed as described [2] with modifications, as described [1]. Chromatographic separations were carried out on 4.6 × 250 mm J’sphere ODS-H80 (4 µm, YMC Co, Kyoto, Japan) for negative ion analysis. TOF-MS and TOF-MS/MS were operated with
same parameters as for metabolomics, as described [1].

2.4. Microinjection rescue studies

Embryos were microinjected as described and criteria used to assess supplementation tolerance of zebrafish embryos using ZAAP at 24, 48, and 120 hpf, as described [1].
Fig. 3. Relative response intensities of antioxidant network components from metabolomics and quantification of α-tocopherol and ascorbic acid. A. E− and E+ embryo (n=15/sample; 4 samples/group) relative response data was normalized against QC sample intensities (n=4) for each individual metabolite. B. Quantified levels of α-tocopherol and ascorbic acid, according to established protocols (31) and (33), respectively. Statistical significance (p < 0.05) was calculated using 2-way ANOVA with Sidak’s post-test for multiple comparisons of normalized and natural log-transformed intensity values. Shown are means ± SEM; p-Values are for VitE x Age interactions, unless indicated otherwise. Paired comparisons p-values are indicated as *p < 0.05, **p < 0.005, ***p < 0.001, ****p < 0.0001. C. Antioxidant network scheme showing interaction of antioxidants with lipid radicals and consumption or NADPH.
Fig. 4. Relative response intensities of glycolytic and tricarboxylic acid cycle intermediates. E− and E+ embryo (n=15/sample; 4 samples/group) data were normalized against QC sample intensities (n=4) for each individual metabolite. Statistical significance (p<0.05) was calculated using 2-way ANOVA with Sidak’s post-test for multiple comparisons of normalized and natural log-transformed intensity values. Shown are means ± SEM; p-values are for VitE x Age interactions. Paired comparisons p-values are indicated as *p<0.05, **p<0.005, ***p<0.001, ****p<0.0001.
Fig. 5. Relative response intensities of free saturated fatty acids and coenzyme A from metabolomic analyses. E- and E+ embryo (n=15/sample; 4 samples/group) data were normalized against QC sample intensities (n=4) for each individual metabolite. Statistical significance (p < 0.05) was calculated using 2-way ANOVA with Sidak’s post-test for multiple comparisons of normalized and natural log-transformed intensity values. Shown are means ± SEM; p-values are for VitE x Age interactions. Paired comparison p-values are indicated as * < 0.05, ** < 0.005, *** < 0.001, **** < 0.0001.
Fig. 6. E- compared with E+ embryos have impaired behavior when injected with saline (upper panel), but restored responses when injected with VitE (lower panel). A. Embryos were analyzed in 96-well plates (128 embryos per group). Locomotor activities following a series of light stimuli (a stimulus every 6 for 24 min) were measured as distance moved (mm) over time (seconds). At 96 hpf, E- (red) embryos treated with saline (upper panel) were 47% less responsive to light than were E+ embryos (E- area-under-curve, AUC: 2040 ± 178; E+ AUC: 3877 ± 228; p < 0.0001). Embryos with morphological defects were not included in data analysis. E- behavior was restored using VitE injection into the yolk at the 1 cell stage (lower panel E- AUC: 2970 ± 280; E+ AUC: 3340 ± 226, not significantly different). B. Bar chart comparisons of respective time-course data. VitE (tocopherol)-injected E- and E+ embryo locomotor activities were not significantly different.
Fig. 7. Locomotor response assay activity data showing neurobehavioral impairment. E− and E+ embryos (96 hpf) were analyzed in 96-well plates (128 embryos per group). Locomotor activities following a series of light stimuli (every 6 for 24 min) were measured as distance moved (mm) over time (seconds). E− (red) embryos treated with saline (upper panel) were 84% less responsive to light than were E+ (blue) embryos (E− area-under-curve, AUC: 572 ± 72 E+ AUC: 3580 ± 387; p < 0.0001). Embryos with morphological defects were not included in data analysis. E− behavior was partially restored by approximately 50% following glucose injection into the yolk at 24 hpf (lower panel; E− AUC: 2502 ± 150; E+ AUC: 3734 ± 359; p < 0.0001). Statistical significance was determined using a Kolmogorov–Smirnov test (p < 0.01).
2.5. Behavioral assessments

Locomotor activity was measured in a total of \( n = 128 \) embryos per group using Viewpoint Zebrabox [4,5], as described [1].

2.6. Data processing and statistical analyses

All data processing and statistical analyses were performed as described in [4–6], with modification made as reported [1].

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Transparency document. Supporting information

Transparency data associated with this article can be found in the online version at http://dx.doi.org/10.1016/j.dib.2017.02.046.

Appendix A. Supplementary material

Supplementary data associated with this article can be found in the online version at http://dx.doi.org/10.1016/j.dib.2017.02.046.

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