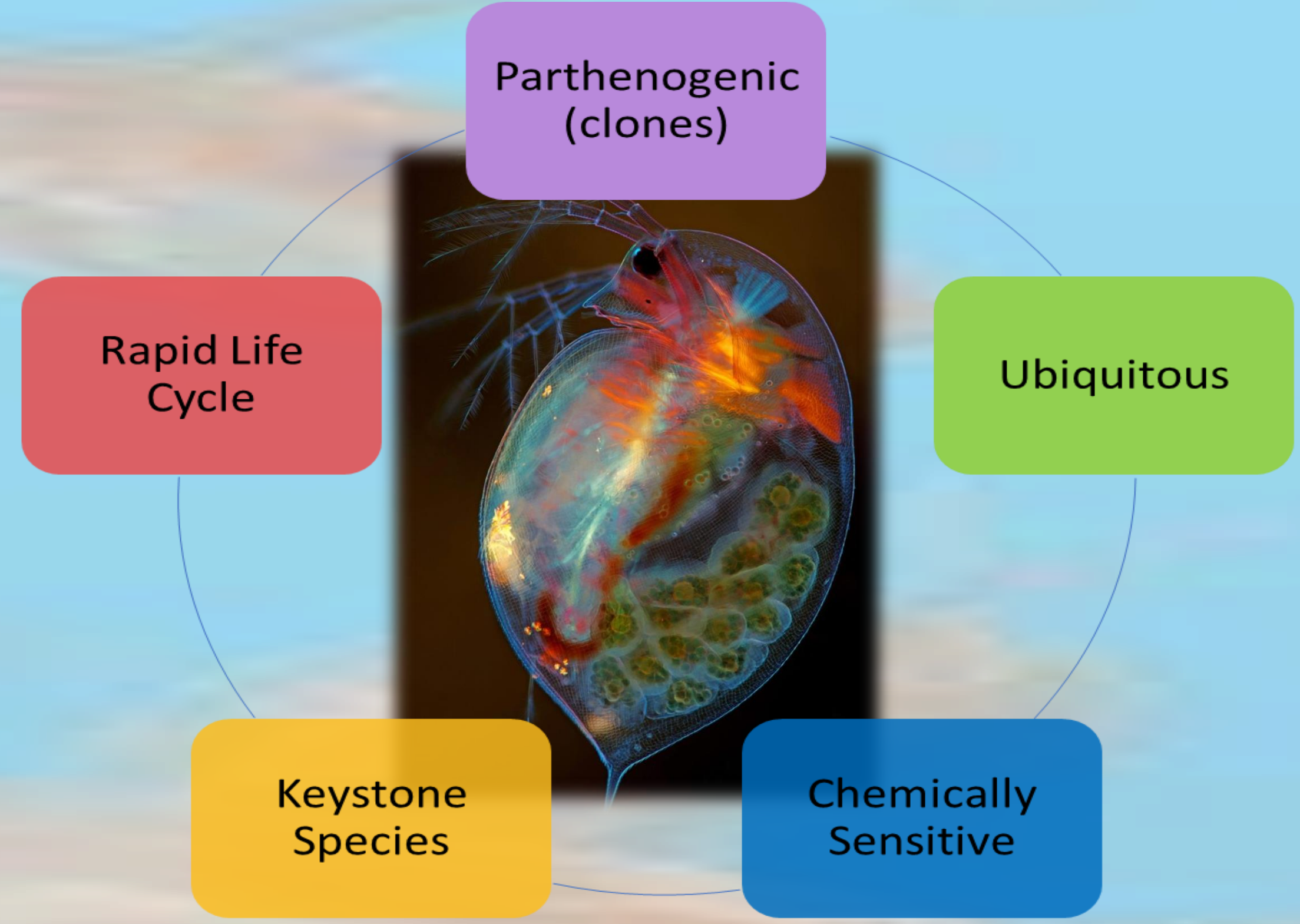


# Using Molecular Methods to Improve Metal Mixture Risk Assessment at Mining-Impacted Sites

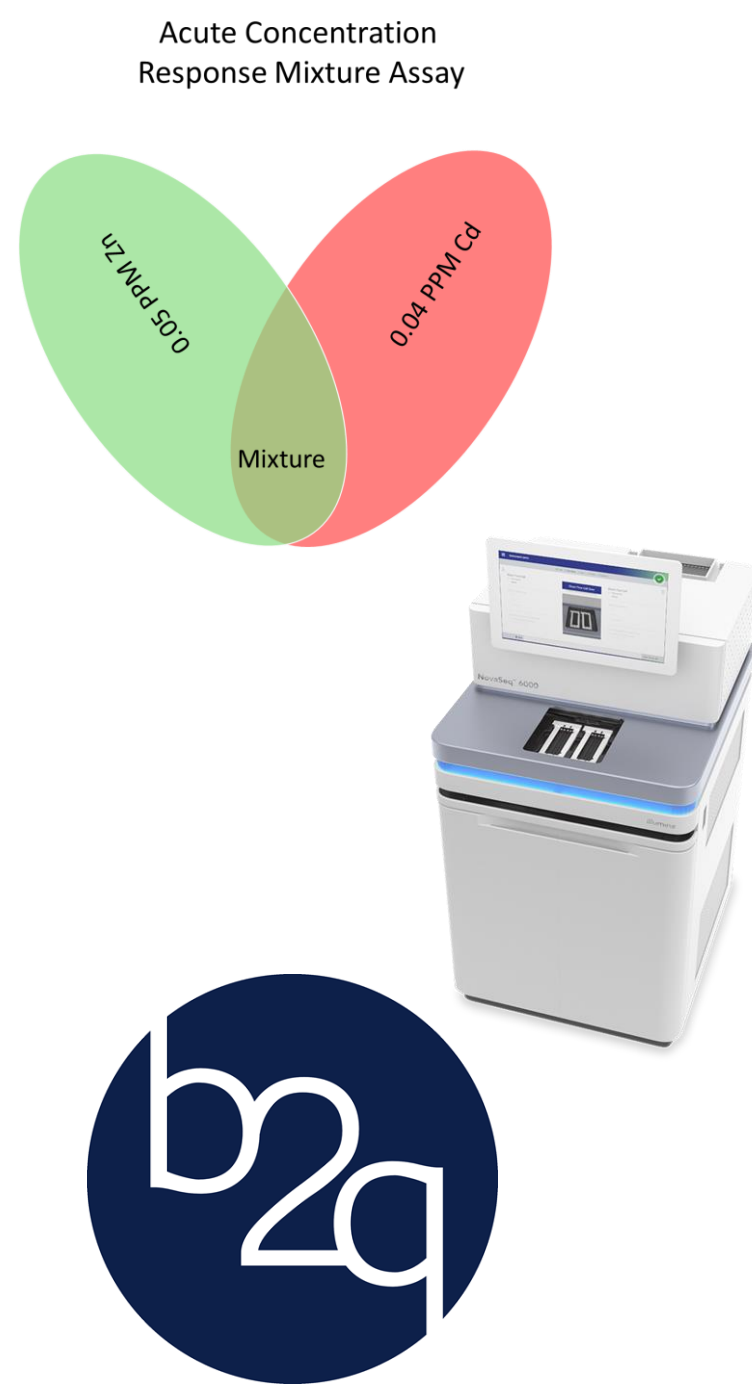
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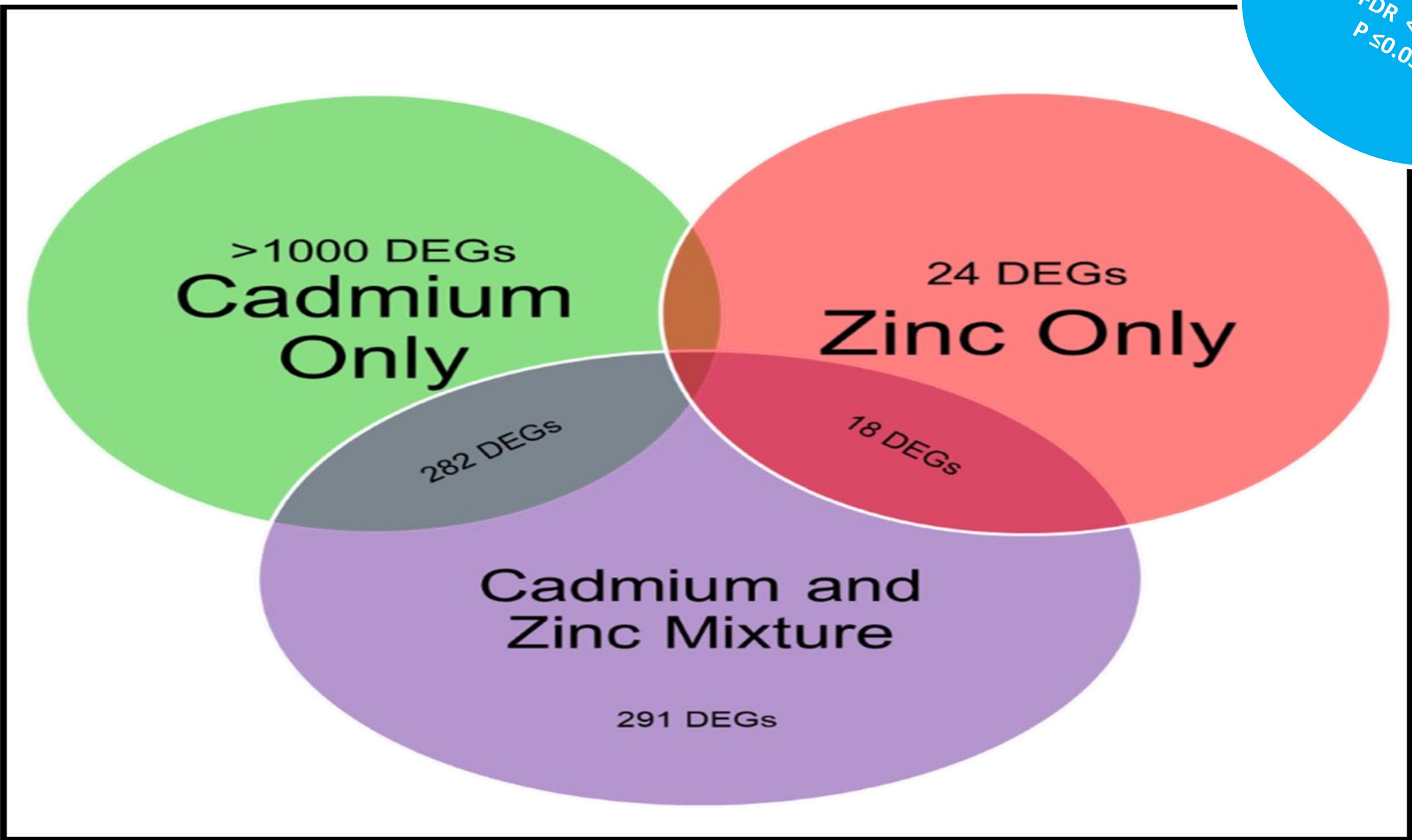
- ### Hypotheses
- The Cd only exposed group and the Zn-Cd mixture exposed group will have DEGs in common indicative of bioavailable Cd in the presence of Zn
  - Transcriptional analysis will reveal key physiological mechanisms indicative of toxicity in the Cd only exposed group that will be nullified in the Zn-Cd mixture exposed group of *D. magna*.

- ### INTRO
- Mining waste impacts human and environmental health
  - Metal mixtures are common in mining effluent
  - Metals behave differently in mixtures
  - One behavior observed is a less-than-additive (protective) effect of Zn for Cd toxicity in *D. magna* at some doses
  - Physiological response plays a role in this protective effect

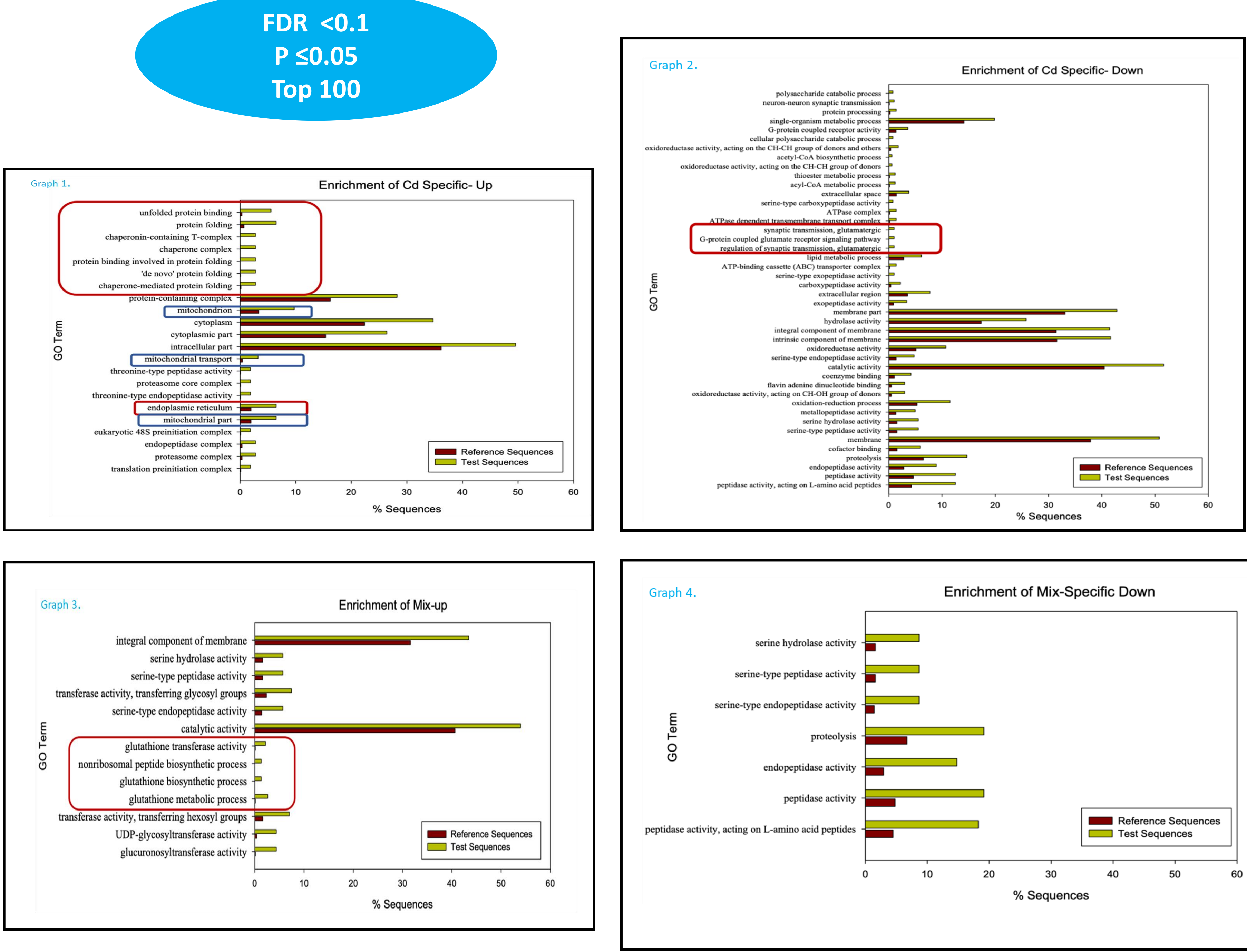
- ### METHODS
1. Exposure: Acute Zn, Cd, and Zn-Cd mixtures using *D. magna*
  2. RNA extraction: TRIzol/Chloroform with a Qiagen RNEasy Mini Kit, Converted to cDNA
  3. Library Prep and QC: Novagene, Inc.
  4. Gene expression: using an Illumina Novaseq 6000
  5. Physiological Effects: Differentially Expressed Gene (DEG) counts and gene ontology analysis using Blast2Go



- ### RESULTS
- *D. magna* exposed to toxic doses of Cd alone differentially expressed over 1000 genes (DEGs)
  - In the Cd-Zn mixture at the same Cd concentration, the number of DEGs decreased to 291. 282 of those DEGs were shared between the two groups.
  - Exposure to Zn alone at the same Zn concentration as in the mixture resulted in only 24 DEGs.



- ### DISCUSSION
- These findings are consistent with a model in which Cd is still bioavailable in the mixture, but Zn may prevent deleterious physiological effects
  - Overall, this work indicates an important role for an internal physiological interaction in the presence of metal mixtures. More work is required to better understand that role.



Key differentially expressed genes as gene functional groups for each treatment group of *D. magna*. Graph 1 shows the DEGs for upregulated genes in Cd only treated *D. magna*. 2 shows the DEGs for downregulated genes in Cd only treated *D. magna*. Graph 3 shows the DEGs for upregulated genes in Cd and Zn mixture treated *D. magna*. Graph 4 shows the DEGs for downregulated genes in Cd and Zn mixture treated *D. magna*.

