RNA-seq characterization of blood in the Broad-snouted caiman (*Caiman latirostris*) after pesticide exposure and immunological challenge

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<u>Abstract:</u> The analysis of expressed genes in a tissue at the mRNA level (transcriptomics) provides much increased insight into biological processes at the molecular level. The aim of this study was to analyze gene expression profile of blood samples obtained from juvenile *C. latirostris* after acute exposure to a cypermethrin-based pesticide formulation and immune challenge with *Escherichia coli*.

Eight juvenile caiman were separated in two experimental groups: a negative control (C) and a group exposed (E) to a cypermethrin formulation (25% CYP). Animals were maintained under controlled conditions into plastics containers and the exposure was performed by voluntary immersion in water, at a concentration of 2 μg/l during 15 days. After exposure, all animals were immunologically challenged by an injection with *E. coli* suspension (0.5 A) at al dose of 0.1 ml/kg during 96 hs. Blood samples were taken to all animals at the beginning and at the end of the experiment and immediately preserved. TRIzol reagent was used for RNA extraction following protocols previously adapted for the species. Libraries were prepared (DNA 0.1-5.0 μg) and DNA was sequenced using *Illumina* sequencing Technology (NGS) through a Hiseq3000.

We identified 1266 differentially expressed genes in caiman challenged by *E. coli*: 489 (38.6%) downregulated (DR) and 777 (61.4%) upregulated (UR). When we analyze animals previously exposed to CYP, the number of genes differentially expressed decreased to 592, being 248 (41.9%) DR and 344 (58.1%) UR. These results showed that the previous exposure of CYP seem to cause a decrease in the immune response of the animals. In order to go further in the understanding of these results, we performed a Functional Annotation analysis using *DAVID Bioinformatic database*, identifying different enriched metabolic pathways in each condition, including lipid and carbohydrates metabolism, immune response and cell adhesion function.

Keywords: differentially expressed genes, transcriptome, toxicity, immune response

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