

## DIFFERENTIAL PATHWAY ENRICHMENT OF UPREGULATED AND DOWNREGULATED GENES IN TAMBAQUI *Colossoma macropomum* MUSCLE UNDER HOUSEFLY LARVAE MEAL

**Vito Antonio Mastrochirico-Filho** (Embrapa Fisheries and Aquaculture - CNPASA)  
**Romulo Veiga Paixão** (Embrapa Fisheries and Aquaculture - CNPASA)  
**Geice Ribeiro da Silva** (Embrapa Fisheries and Aquaculture - CNPASA)  
**Cherlle Kally Lima de Almeida** (Embrapa Fisheries and Aquaculture - CNPASA)  
**Ana Paula Oeda Rodrigues** (Embrapa Fisheries and Aquaculture - CNPASA)  
**Rodrigo Fortes da Silva** (Embrapa Fisheries and Aquaculture - CNPASA)  
**Eduardo Sousa Varela** (Embrapa Fisheries and Aquaculture - CNPASA)

Tambaqui (*Colossoma macropomum*) is the most economically important native species in South American aquaculture. To reduce reliance on fishmeal, sustainable protein alternatives such as insect larvae are being explored, with housefly (*Musca domestica*) larvae meal as one promising option. Here, we examine gene expression changes in white muscle of tambaqui fed a diet containing 32% housefly larvae meal, focusing on functional enrichment of differentially expressed genes (DEGs). Tambaqui were fed a diet containing 32% housefly larvae meal, and white muscle samples underwent transcriptomic (RNA-seq) analysis. Differential expression analysis identified 51 significantly upregulated and 148 downregulated genes in the larvae meal group relative to a control diet. Upregulated genes were enriched in metabolic processes and stress-response pathways. In GO, terms related to carbohydrate and glucose metabolism (e.g., regulation of glucose metabolic process) were prominent, alongside stress-activated MAPK signaling (JNK cascade). KEGG analysis indicated increased lipid catabolism, with “Fatty acid degradation” as the top enriched pathway. Autophagy and one-carbon metabolism pathways were also represented, suggesting greater energy mobilization under the larvae meal diet. Downregulated genes were mainly associated with muscle structure and contraction. GO terms for muscle contraction, muscle cell development, and myofibril assembly were among the most enriched, reflecting broad downregulation of myofibrillar protein genes (e.g., tropomyosins, troponins, myosins). Similarly, KEGG analysis showed enrichment of muscle contraction-related pathways among downregulated genes. Many of these genes encode muscle cytoskeletal components, consistent with overrepresentation of a muscle cytoskeleton module and the “cardiac muscle contraction” pathway. This pattern confirms the suppression of genes involved in the contractile apparatus. In summary, the housefly larvae meal diet elicited upregulation of muscle metabolic and stress-response pathways, while downregulating genes for muscle fiber structure and contraction. These shifts suggest a potential muscle remodeling or mild catabolic state. Optimizing insect meal inclusion levels and balancing essential nutrients (e.g., amino acids) may be crucial to support healthy muscle development.

Funding: This study was supported by FAPEMIG APQ-00684-22, CNPq 443875/2024-3, FAPT/SEAGRO 2023.