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Stocking density and copper effects on the antioxidant defenses and hematological parameters of pacu

Fernanda Garcia Sampaio[*], Cheila de Lima Boijink[1], Eliane Tie Oba[2], Laila Romagueira Bichara dos Santos[3], Ana Lúcia Kalinin[4], Marcos Eliseu Losekann[5], Alfredo José Barreto Luiz[5] and Francisco Tadeu Rantin[4]
[*] Pesquisadora, Empresa Brasileira de Pesquisa Agropecuária – Embrapa; Rod. SP 340, km 127,5; cep: 13820-000; Jaguariúna, SP; fesampaio@cnpma.embrapa.br; [1] Embrapa, Manaus, AM; [2]Embrapa - Macapá, AP; [3]Departamento de Fisiologia e Biofísica; Instituto de Ciências Bioquímicas; Universidade de São Paulo, SP; [4]Laboratório de Zoofisiologia e Bioquímica Comparativa, Departamento de Ciências Fisiológicas, Universidade Federal de São Carlos, São Carlos, SP - Instituto Nacional de Ciência e Tecnologia – Fisiologia Comparada; [5]Embrapa - Jaguariúna, SP.

The use of copper products, such as $CuSO_4$, in the aquatic environment is an emerging issue in environmental chemistry. In this study the objectives were to determine the acute toxicity of copper in pacu and the influence of fish stocking densities to copper toxicity. Pacu were exposed to increased water copper concentration ($[Cu]_W$) at different fish stocking density: low density 1.0 g L^{-1} (Ld) and high density 2.5 g L^{-1} (Hd). Mean lethal concentrations (LC_{50}) were determined for Ld and Hd. Fish (n = 5 and n = 12; Wt = 36.8 ± 4.3 g; 2 - 4 replicates) were distributed in 180 L glass aquaria (static system) for 48 h and submitted to increased copper concentrations. Fish (n = 5 to 12; wt = 38.0 ± 3.2 g) were randomly distributed in the experimental glass aquaria (180 L; static system, 2 - 3 replicates for each group) in order to verify effects of copper and stocking densities on physiological parameters of pacu. At Ld, fish were exposed to control (copper free, 0CuLd), 0.5 mg Cu^{2+} L^{-1} (0.5CuLd) and 1.0 mg Cu^{2+} L^{-1} (1.0CuLd). At Hd, fish were exposed to control (copper free, 0CuHd), 2.0 mg Cu^{2+} L^{-1} (2.0CuHd) and 4.0 mg Cu^{2+} L^{-1} (4.0CuHd), for 48 hours. The LC_{50} was 3.5-fold higher with an increase of 2.5 times in the stocking density. Fish of both Ld and Hd were very sensitive to increases in water copper concentrations, displaying time and dose-dependent curves. The hepatic lipid hydroperoxide (HP) level increased in fish exposed to 0.5CuLd and 1.0CuLd when compared with control ones. Fish exposed to increased copper concentration, at Hd, the HP formation decreased when exposed to 2.0CuHd and increased when exposed to 4.0CuHd. Addition of copper to the Ld group caused increase in hepatic superoxide dismutase (SOD) activity. However, when copper was added to the Hd group the liver SOD activity remained unchanged, showing that fish stocking density directly influences the SOD response to copper exposure. Glutathione peroxidase (GSH-Px) increased proportionally to $[Cu]_W$ at Ld. However, when copper concentration increased from 0.5CuLd to 1.0CuLd, no differences were detected. In Hd, the increase in $[Cu]_W$ did not change the GSH-Px activity in liver, suggesting that higher fish stocking density changes the copper effects in fish liver GSH-Px activity. Catalase (CAT) activity of fish exposed to 0.5CuLd and 1.0CuLd, increased when compared to 0CuLd. The same increase was observed in CAT activity of fish exposed to 4.0CuHd while 2.0CuHd was similar to control values. Comparing the red blood cell count (RBC) values at different stocking densities, fish exposed to 0.5CuLd presented RBC higher than in 2.0CuHd. The hemoglobin concentration (Hb) of fish exposed to increased $[Cu]_W$ augmented in both stocking densities. The increase of Hb in the Ld was more pronounced, showing that at a low density, the copper effect was more intensified. Increasing fish stock density unchanged biochemical parameters and antioxidant status. Our results indicate that fish sensitivity to copper decreases as fish stocking density increases. The biomarkers analyzed seem to be sensitive as antioxidant status and hematological parameters as each density presented the same tendency in response to copper increases, and are more significant in the Ld when compared to the Hd. These results point out that fish stock density has to be taken into consideration when estimating copper toxicity in fish culture.

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