

Single microcystin exposure impairs the hypothalamic-pituitary-gonadal axis at different levels in female rats

Flavia C. F. Dos Santos¹, Gabriela F. C. Lima¹, Eduardo Merlo¹, Cidalia de F. Januario¹, Leandro Miranda-Alves², Rosiane A. Miranda³, Patrícia C. Lisboa³, Jones B. Graceli¹.

¹Department of Morphology, Federal University of Espírito Santo, 290440-090, Vitória, Brazil.

²Experimental Endocrinology Research, Development and Innovation Group, Institute of Biomedical Sciences, Federal University of Rio de Janeiro, 21941-904, Ilha do Governador, Brazil

³Laboratory of Endocrine Physiology, Biology Institute, State University of Rio de Janeiro, RJ, Brazil.

Microcystin (MC) is the most common cyanobacterial toxin. Few studies have evaluated the MC effects on the hypothalamic-pituitary-gonadal (HPG) axis function. In this study, we assessed whether MC exposure results in HPG axis changes. Female rats were exposed to a single dose of MC at environmentally relevant levels 5, 20 and 40 µg/kg (MC5, 20 and 40 respectively, n=15). After 24 hours, we evaluated reproductive parameters for 15 days. Improper functioning of the HPG axis was observed in MC rats. Specifically, MC reduced hypothalamic gonadotropin-releasing hormone (GnRH) protein expression in MC40 rats and increased the pituitary protein expression of gonadotropin-releasing hormone receptor (GnRHr) in MC20 rats compared to control rats ($p < 0.05$). A borderline increase in pituitary lutropin β was observed in MC20 rats compared to control rats ($p: 0.09$). In addition, MC exposure reduced serum luteinizing hormone (LH) levels in MC40 rats and increased follicle-stimulating hormone (FSH) levels in MC20 and MC40 rats compared to control rats ($p < 0.05$). MC exposure increased pituitary IL-6 and ED1 protein expression (inflammatory markers) in MC5 rats compared to control ($p < 0.05$). Additionally, MC exposure borderline increased hypothalamic inducible nitric oxide synthase (iNOS) expression in MC5 compared to control rats ($p: 0.06$). A negative linear correlation between hypothalamus GnRH and iNOS protein expression was observed ($p < 0.05$). MC exposure reduced the primary follicles in MC5 and MC40 rats and increased the corpora lutea number in MC20 rats compared to control rats ($p < 0.05$). An increase in anti-Müllerian hormone (AMH) and progesterone levels were found in MC20 rats, and a decrease in estrogen levels were found in MC5 and MC40 rats ($p < 0.05$). MC exposure increased ovarian vascular endothelial growth factor receptor (VEGFr), LH receptor (LHr), AMH, ED1, IL-6 and Gp91-phox (oxidative stress marker) protein expression in MC20 rats compared to control rats ($p < 0.05$). MC increased uterine area and reduced endometrial gland number in MC5 and MC40 rats ($p < 0.05$). Blunted estrogen-negative feedback was observed in MC40 rats after ovariectomy, with no changes in LH levels compared to intact MC rats ($p > 0.05$). Therefore, these data suggest that a MC leads to abnormal HPG axis function in female rats.