The effect of silymarin and N-acetyl-L-systeine against microcystin LR-induced oxidative stress in visceral tissues of Balb/c mice.

By:
Anas Mohammad AL-Omari
B.Sc. Biological Sciences, Al-Albayt University

A Thesis Submitted in Partial Fulfillment of the Requirements for the degree of Master of Science in the Department of Biological Sciences, Yarmouk University, Irbid, Jordan

Approved by
Prof. Saad A. Al-Jassabi
Professor of Biochemistry, Yarmouk University.

Dr. Sayedda Mir
Associate Professor of Zoology / Fish Biology, Yarmouk University

Prof. Hanan I. Malkawi
Professor of Microbiology and Molecular Biology, Yarmouk University

Dr. Ahmed Maslat
Associate Professor of Molecular Genetics, Yarmouk University

Prof. Mohamad Nusier
Professor of Biochemistry and molecular Biology, Jordan University of Science and Technology, School of Medicine.
Abstract:

Microcystins (MCs) are hepatotoxic compounds produced mainly by the cyanobacteria *Microcystis* spp. which are distributed in water bodies worldwide. The exposure to MCs toxicity was reported worldwide in animals and in humans for over a century. There are about 70 known variants of MCs to date and the most toxic and widely distributed MC is Microcystin-LR (MCLR). Oxidative stress has been implicated as one of the possible mechanism of MC-LR induced toxicity.

The present investigation examined the toxicity effects of microcystin-LR isolated from *M. aeruginosa* present in King Talal Reservoir (KIR) on Balb/c mice tissue (small intestine, lung, heart, and spleen) after intraperitoneal (i.p) route of exposure to the toxin, which was measured by means of biochemical analysis. The study also investigated the antioxidant potential capacity for different antioxidants silymarin and N-acetyl-L-cysteine in the inhibition of toxicity by MCLR. The LD$_{50}$ dose was determined to be 19 mg toxin/kg mouse of body weight.

Mice were treated with 250 mg/kg silymarin and 10 mM N-acetyl-L-cysteine, orally and daily for 10 days, followed by intrapertoneal injection of the LD$_{50}$ dose. In toxin treated mice, visceral tissue toxicity was assessed by measuring the alkaline phosphatase activity (ALP), and increase serum
activity of alanine aminotransferase (ALT) and aspartate aminotransferase (AST). MC-LR treatment increased lipid peroxidation levels measured as thiobarbituric acid reactive substances (TBARS) concentrations as well as increased carbonyl content and methylglyoxal content levels along with depletion of glutathione (GSH) levels and reduction in alkaline phosphatase activity in small intestine and heart with different degrees compared to control group of toxin-treated when compared to control mice while the lung and spleen were not affected. In contrast, the antioxidant treated groups showed a decrease toxicity levels caused by LD₅₀ dose which indicated by lower levels of lipid peroxidation, methylglyoxal and carbonyl contents along with increase GSH concentration in visceral tissues. Higher ALP along with lower ALT and AST activities were detected in antioxidants treated groups. The results also showed that Silymarin has the highest antioxidant potential followed by the N-acetyl-L-cysteine in comparison to trolox standard.

The results indicate that silymarin and N-acetyl-L-cysteine have a potent protective effect against MC-LR induced toxicity and suggest that these antioxidants may find clinical applications against a variety of toxins where cellular damage is a consequence of reactive oxygen species.

Keywords: cyanobacteria, Microcystis aeruginosa, Microcystin-LR, Antioxidants, Silymarin, N-acetyl-L-cysteine.
ملخص الرسالة

"تأثير السيماين والاستيل مستئن على الإجهاد الأوكسيدي الناتج عن الميكروسيتيم في أنسجة أحشاء الفنار" 

إعداد

اسم محمد العمري

إشراف

أ.د. سعد عبد محمد الجسابي

מشرف شكر

د. سامح "غلام محمد " مير

تعد سموم الميكروسيتيم من البيبيتيدات الحلقيا سباعة الأحماض الأمينية حيث تنتجها طحالب الميكروسيتيم أروجينوزا بشكل أساسي الأكتر انتشارا ضمن الأنواع في المجمعات المائية، وتشكل هذه السموم خطورة وتهديد على حياة الإنسان والحيوان ويعتقد أن حدوث الإجهاد الأوكسيدي وراء سمية هذه المركبات.

تبش هذه الدراسة أثر سموم الميكروسيتيم المعزولة من طحالب الميكروسيتيم أروجينوزا من سد الملك طلال على الأحشاء الداخلية للفنان (الامتعاء الدقيقة، الرئتين، القلب، الطحال). بعد حقنها بالسم عن طريق التجوية البطنى، كما تبحث أيضا مدى فاعلية مضادات الأكسدة السييمارين والاستيل سيتيم في إبطاء سمية الميكروسيتيم. وقد تم تحديد الجرعة القاتلة لنصف مجموعة الفنان كانت 19مغ / كغم من وزن الفأر.