

Print ISSN : 0972-8813
e-ISSN : 2582-2780

[Vol. 22(2) May-August 2024]

Pantnagar Journal of Research

(Formerly International Journal of Basic and
Applied Agricultural Research ISSN : 2349-8765)



G.B. Pant University of Agriculture & Technology, Pantnagar



CONTENTS

| | |
|---|-----|
| Impact of <i>Zea nicaraguensis</i> introgression on Kernel Trait Variability in maize lines | 231 |
| SENTHILKUMAR V., PRIYA GARKOTI., THOTLA NARESH, MAYANK TIWARI, ANIRUDH T. V. and NARENDRA KUMAR SINGH | |
| Improving <i>Brassica juncea</i> performance through hybrid breeding strategies: a focus on combining ability and heterosis analysis | 244 |
| ANU SINGH, USHA PANT, PREETI LOHANI, A. S. JEENA and ANIL KUMAR | |
| Study of Nano Urea application under graded n rates on growth, productivity and nitrogen use efficiency of transplanted rice (<i>Oryza sativa</i> L.) | 251 |
| S.K.YADAV , D.K.SINGH, PRATIMA ARYA and YUVRAJ SINGH | |
| Isolation, screening and characterization of Drought tolerant Plant Growth Promoting bacteria from Indian Himalayas | 261 |
| PRIYANKA KHATI, PANKAJ KUMAR MISHRA and LAKSHMI KANT | |
| Impact of Glomalin-Related Soil Proteins on <i>in vitro</i> Finger Millet (<i>Eleusine coracana</i> (L.) Gaertn.) seed germination | 272 |
| AMIT SINGH RANA, SUGANDHA PANT, ASHOK KUMAR VERMA and ASHUTOSH DUBEY | |
| Rating scale of pedological development in humid moisture regime of guava growing soils in north-east region of Haryana | 279 |
| DHARAM PAL and DINESH | |
| Coating micronized elemental sulphur powder on prilled urea: process and product evaluation | 286 |
| P. O. SURESH, N. R. PATEL, R. JAT, R. A. PANIA, A. K. MISHRA, P. B. VAISHNAV | |
| Multi-year temporal analysis of sheath blight incidence in rice using geostatistical technique | 297 |
| AMIT BIJLWAN, RAJEEV RANJAN, MANENDRA SINGH, RAJ KUMAR SINGH, RAJEEV KUMAR SRIVASTAVA, KRISHNA PRATAP SINGH and RAVINDRA KUMAR SINGH RAJPUT | |
| Efficiency assessment of classifiers for sugarcane area mapping: A machine learning approach with Google Earth Engine | 305 |
| POOJA YADAV, AJEET SINGH NAIN and SHIVANK DEVLİYAL | |
| Calibration and performance evaluation of the APSIM and CERES-Wheat model in the foot hills of Western Himalayas | 319 |
| NEHA PAREEK, A.S. NAIN, P. K. SINGH, HEMANT KUMAR, SHRUTI V. SINGH, MANJARI SINGH, PRIYANKA SWAMI and SANTOSH KUMAR | |
| Population dynamics of major insect pests of sesame and their correlation with meteorological factors | 330 |
| BHUMIKA RAWAT, M. S. KHAN, ASHUTOSH and DEEPIKA JEENGAR | |
| <i>In-vitro</i> screening of <i>Trichoderma</i> isolates for their antagonistic potential against <i>Rhizoctonia solani</i> causing aerial blight of Soybean | 335 |
| ARUNKUMAR, BHUPESH CHANDRA KABDWAL and ROOPALI SHARMA | |
| Physiological and biochemical responses of okra seed (<i>Abelmoschus esculentus</i> L.) to botanicals and containers during storage | 350 |
| SUNIL KUMAR, S. S. JAKHAR, ANIL KUMAR MALIK and AXAY BHUKER | |
| Effect of integrated weed management practices on growth parameters in vegetable pea (<i>Pisum sativum</i> L.) | 357 |
| NEELIMA RAWAT, MANOJ RAGHAV, DHIRENDRA SINGH, ALKA VERMA, NAVNEET PAREEK, HITAIISHI KURIYAL and IMAMUDDIN SHAH | |

| | |
|---|------------|
| Maximizing Chrysanthemum (<i>Dendranthema gradiflora</i>T.) growth and yield: Unveiling the superiority of Black Polythene Mulch | 360 |
| HARSHITA BORA, MAMTA BOHRA and K. C. SINGH | |
| Utilization of ultrasonicated edible coating to prolong shelf life of fresh cut- onion | 368 |
| NEHA RAWAT, SATISH KUMAR SHARMA, ANIL KUMAR, NAVIN CHANDRA SHAHI, ASHUTOSH DUBEY, CHARU BISHT, ARCHANA GANGWAR | |
| Effect of cooperative societies on food security status of cassava farming households in delta state, Nigeria | 378 |
| IZEKOR, O.Band OKOROR O.T. | |
| Strategies for Improving Agricultural practices: A case study of tomato growers from Uttarakhand | 388 |
| TAMANNA JOSHI and ASHUTOSH SINGH | |
| Physico-functional and sensory qualities of instant custard powder incorporated with resistant starch from Grand Naine banana | 398 |
| SRUTHY. P. M., SHARON. C. L., SEEJA THOMACHAN PANJIKKARAN, A. N. JYOTHI, ANEENA E. R.and LAKSHMI P. S. | |
| Development and quality evaluation of rice-based meal replacer with chocolate flavour for adults | 404 |
| ATHIRA RAJ, SUMAN K.T., BEENA A. K., SEEJA THOMACHAN PANJIKKARAN, SHARON C. L., LAKSHMY P. S., DELGI JOSEPH C.and SREELAKSHMI A. S. | |
| Effect of bleaching on optical properties of <i>dhaincha</i> (<i>Sesbania aculeata</i>) pulp | 411 |
| SURABHI DAS, ANITA RANI, MANISHA GAHLOT, SAKSHI and NIDHI SISODIA | |
| Evaluation of genetic and non-genetic factors affecting first lactation traits in crossbred cattle | 421 |
| NAYLA FRAZ, B. N. SHAHI, R. S. BARWAL, C. V. SINGH and A. K. GHOSH | |
| Mushroom (<i>Agaricus bisporus</i>) waste as a replacement for deoiled rice bran and its impact on immunocompetence against Ranikhet (Newcastle) disease virus in Rhode Island Red Chicken | 426 |
| MANAS ARORA, R. KUMAR, A. TEWARI, A. KUMAR, J. PALOD and B.C MONDAL | |
| Effect of <i>Aloe vera</i> leaf extract on pathological lesions of <i>Escherichia coli</i> infected broiler chickens | 433 |
| MAMTA KUMARI, RAJENDAR P. GUPTA, DEEPIKA LATHER, PREETI BAGRI, RENU SINGH, SARVAN KUMARand KOMAL | |
| Effect of metronidazole on hematological parameters in Common Carp (<i>Cyprinus carpio</i>) | 443 |
| ANIKA SHARMA, MADHU SHARMA, TARANG SHAH and PRASANJIT DHAR | |
| Reproductive and productive performances of Japanese Quails (<i>Coturnix japonica</i>) under agro-climatic conditions of Assam | 449 |
| DEBAJIT DEKA, ARFAN ALI, ASHIM KUMAR SAIKIA, MRIDUL DEKA, UTPAL JYOTI SARMA, MANORANJAN NEOG and RANJIT KUMAR SAUD | |
| Performances of Turkey birds under backyard system in agro-climatic condition of Assam | 454 |
| DEBAJIT DEKA, ARFAN ALI, ASHIM KUMAR SAIKIA, MRIDUL DEKA, MANORANJAN NEOG, RANJIT KUMAR SAUD and UTPAL JYOTI SARMA | |
| Nutraceutical supplements for managing pain and inflammation: A special focus on palmitoylethanolamide and astaxanthin | 459 |
| AKHTER RASOOL, DIVYA CHAVAN, PULI VISHNUVARDHAN REDDY, JAN MOHD MUNEEB and IRTIQA MANZOOR | |
| Characterization and use of hydrochars from wheat straw, fruit peels, and sewage sludge: A potential biofuel source | 470 |
| KARAN SATHISH and SHWETA SARASWAT | |
| Battery assisted single wheel weeder for medicinal plants | 479 |
| SANDEEP KUMAR SAROJ and JAYANT SINGH | |
| Chat GPT: Perception of students towards AI tool | 486 |
| ARPITA SHARMA KANDPAL and POOJA GOSWAMI | |

Effect of metronidazole on hematological parameters in Common Carp (*Cyprinus carpio*)

ANIKA SHARMA¹, MADHU SHARMA^{1*}, TARANG SHAH¹ and PRASANJIT DHAR²

¹Department of Fisheries, ²Department of Veterinary Microbiology, DGCN College of Veterinary and Animal Sciences, CSKHPKV, Palampur-176062 (H.P.)

*Corresponding author's email id: madhu.srma@gmail.com

ABSTRACT: The potential hemotoxic effect of the commonly utilized antibiotic metronidazole (MTZ) was investigated in the common carp (*Cyprinus carpio*). The fish were exposed to sublethal concentrations of MTZ, 5 mg/L and 10 mg/L in water for duration of 96 hours along with one control group. No macroscopic pathology or death of fish was observed at these concentrations. Various hematological parameters were evaluated, including Hemoglobin (Hb), Packed Cell Volume (PCV), Red blood cell (RBC), White blood cell (WBC), Mean Corpuscular Hemoglobin (MCH), Mean Corpuscular Volume (MCV), and Mean Corpuscular Hemoglobin Concentration (MCHC). Under the tested concentrations, MTZ- exposed fish at 5 mg/L dosage exhibited a gradual increase in Hb and RBC values, while a decrease in values of WBC, PCV, MCV, MCH and MCHC from control to 96 hours was observed. However, the fish exposed at 10mg/L dosage exhibited a gradual increase in RBC, MCHC values, while a variation in values in Hb, WBC, PCV, MCV and MCHC from control to 96 hours was seen. Moreover, hematological parameters have the potential to act as appropriate biomarkers for assessing fish health and can be utilized as bio-indicators in environmental bio-monitoring.

Key words: Metronidazole, Common Carp, Pharmaceuticals, Haematology

Antibiotics are a category of pharmaceutical compounds that are widely used and consumed on a worldwide. The worldwide usage of antibiotics is steadily on the rise, with a substantial 80% growth, mainly due to the escalating demand in low and middle-income countries. Aquatic contaminants originating from pharmaceuticals used in human health, agriculture, and aquaculture have garnered significant attention in recent years (Li *et al.*, 2011; Anoo *et al.*, 2024; Nwokediegwu *et al.*, 2024). Metronidazole (MTZ) is a widely used antibiotic as an antiparasitic and antibacterial compound in veterinary and medical use. Metronidazole (MTZ; 2-(2-methyl-5-nitro-1H-imidazol-1-yl) ethanol) classified under the 5-nitroimidazole class of drugs. It is widely utilized globally to treat various infections in humans and animals caused by protozoa or anaerobic bacteria (Rizzo *et al.*, 2010). Nevertheless, the utilization of this drug in the treatment of food fish has been prohibited in both Europe and the USA. This prohibition is primarily attributed to its potential to cause cancer, its ability to persist in the environment, and its harmful effects on aquatic organisms. Despite this, metronidazole

is available for the treatment of ornamental fish through veterinary prescription, and is widely uses in some specific areas of South East Asia. MTZ is highly soluble in water (Wu and Fassihi, 2005), but it also exhibits hydrolytic and photostable properties (Sánchez-Polo *et al.*, 2012), potentially resulting in accumulation in aquatic ecosystems (Richardson and Bowron, 1985; Rediguieri *et al.*, 2011). Being soluble in water and not easily biodegradable, the discharged MTZ is assumed to be fairly persistent in water, where it may be taken up by aquatic animals (Rodriguez *et al.*, 2011). Moreover, this drug has been found to possess carcinogenicity, mutagenic and toxic properties. However, the effective removal of MTZ from waste water from sewage treatment plants still remains problematic. Hence, this drug has the potential to significantly impact non-target aquatic organisms and could lead to the creation of bacterial strains that are resistant to its effects (Johnson, 1993; Kümmerer, 2004). Jousimies-Somer *et al.* (1988) investigated and found that the metronidazole concentration in water before and after treatment at Spanish sewage plants was 164 ng/L and 167 ng/L, respectively. Through the

detection of the effluent from the sewage treatment plant, it was found that the concentration of MTZ had reached a level that could cause harmful effects towards humans and the ecological environment (Marta *et al.*, 2015). MTZ metabolites are carcinogenic and mutagenic in some animals because original nitromidonic ring is retained. This may pose the possibility for long-term effects upon these animals. MTZ used at standard dose has been shown to suppress the growth of bone marrow, which is the primary hematopoietic organ and central immune organ (Fararjeh *et al.*, 2008).

The analysis of fish hematological characteristics has become a crucial tool in gaining insights into both normal and pathological processes (Singh and Srivastava, 2010; Li *et al.*, 2011). Hematological analysis is a valuable tool for monitoring the health and condition of wild and cultured fish. Hematological indices vary based on factors such as fish species, age, sexual maturity cycle, and overall health status (Blaxhall, 1972). Furthermore, haematological assessments and serum component analyses have proven to be beneficial in identifying and diagnosing metabolic disruptions and diseases in fish (Aldrin and Messenger, 1982; Jamalzadeh *et al.*, 2009). It is known that blood represents 1.3–7% of fish body mass and represents one of the most active components that with hematopoietic organs contribute to constant metabolic processes ensuring gas and nutrient changes between animal body and environment. The value of standard hematological indices plays a crucial role in evaluating the overall physiological condition of fish, with a specific emphasis on their health status (Radu *et al.*, 2009). The Common carp (*Cyprinus carpio*) is categorized within the order Cypriniformes and the family Cyprinidae, which is recognized as the largest family of freshwater fish. It is commonly found in freshwater habitats like ponds, lakes, and rivers, and can also be occasionally found in brackish-water environments (Barus *et al.*, 2001). Common carp is considered to be a very important aquaculture species in many Asian and some European countries. It affects the aerobic decomposition of organic matter and nutrient availability in the water column via bioturbation of benthic sediment during feeding on

benthic organisms (Rahman, 2015). According to the FAO (2013), the common carp is the third most widely farmed and commercially valuable freshwater fish species worldwide. In Asia, common carp are normally cultured in various aquaculture systems but the most common is the semi-intensive pond polyculture system (FAO 2012).

Considering the importance of fish as a source of food for humans, as well as their value in reflecting the quality of the aquatic environment (Kollner *et al.*, 2002), it is useful to further assess the effects of MTZ on fish health. The aim of this study was to evaluate the effects of MTZ on the hematological parameters in Common Carp.

MATERIALS AND METHODS

Experimental organisms

The present study was conducted on Common carp (*Cyprinus carpio*) obtained from the Department of Fisheries farm at CSK Himachal Pradesh Agricultural University, Palampur (India). The removal of dermal infection was done with 0.2% KMnO₄ for 2 minutes in a stock tank. The fish were transferred to the laboratory and kept in three different aquaria of 200 liters capacity half filled with dechlorinated water for acclimatization. The fish were fed on the farm-made pelleted feed once in a day. The trial was conducted for duration of 96 hrs. Fresh dechlorinated water was regularly introduced on a daily basis to replace the existing water throughout the duration of the trial. The total number of 24 fish were divided into three different groups, *i.e.*, Treatment groups (Group I & II) and control group with 8 fish in each treatment group and in control. The treatment groups were subjected to addition of MTZ at the rate 5mg/L and 10mg/L concentrations in respective treatment group. The average length and weight of the fish was 19±0.5 cm and 103.9±6.5 g respectively.

Drug and dosage

The drug MTZ was purchased from a local pharmacy manufactured by Denis Chem Lab Ltd. The liquid formulation had a concentration of 500 mg/100ml. The sublethal concentrations of MTZ were decided as 5mg/L and 10mg/L of water for the exposure

according to the value given by Cavas and Ergene (2005).

Collection of samples

The blood samples were collected from the caudal vein with heparinized syringe storage vials at 24, 48, 72 and 96 hours intervals from treatment as well as the control groups. The samples were transported in an ice box to the laboratory and were analyzed immediately.

Hematological analysis

The blood samples were collected and analyzed for Haemoglobin, Packed Cell Volume, Red Blood Cell (RBC) White Blood Cell (WBC), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH) and Mean Corpuscular Hemoglobin Concentration (MCHC). The haemoglobin (Hb) concentration was determined using the Cyan Methemoglobin method (Sahli's Hemoglobinometer method) (Jain, 1986). The red blood cells (RBC) and white blood cells (WBC) counts were determined by the Haemocytometer (Neubauer Chamber) method. The packed cell volume (PCV) was determined by the Micro-haematocrit method. MCV, MCH and MCHC were computed according to formulas'

$$\begin{aligned} \text{MCV (fl)} &= \text{PCV} \times 10/\text{RBC} \\ \text{MCH (pg)} &= \text{Hb} \times 10/\text{RBC} \\ \text{MCHC (g/dL)} &= \text{Hb} \times 100/\text{PCV} \end{aligned}$$

Statistical Analysis

The results obtained in the study are expressed as mean \pm S.E and to study the significance of difference in values of different haematological parameters between treated and control groups a one-way analysis of variance (ANOVA) followed by Post Hoc Tukey's Test using statistical software 'Graph Pad' was applied.

RESULTS AND DISCUSSION

The values of all the hematological parameters are given in Table 1. Significant variations ($p < 0.05$) were observed between various blood parameters with different concentrations of MTZ. The data

shows that change in hematological indices of fish may be due to MTZ and are determined both by concentration of MTZ and time of exposure. The hemoglobin (g %) of fish treated with MTZ show an increase at both the concentrations however a greater increase in values was seen at 10mg/L concentration.

In comparison to the control group, a decline in values of PCV (%) was observed at 5 mg/L concentration while an increase was observed at 10mg/L concentration. In comparison to the control group, an increase was seen in the values of RBC count at both concentrations. However, a greater increase was observed at 10 mg/L concentration.

The WBC count (10^3 cells/ μ L) of fish treated with MTZ results in decline in values at both the concentrations. In comparison to the control group, a major decline in values of MCV was observed at both concentrations. The MCH values of fish treated with MTZ showed a decrease in the value at both the concentration. The MCHC values were observed to increase at both concentrations as compared to the control group. However, the increase was more significant at 10 mg /L.

MTZ is an antibiotic used for parasitic infections in a number of species. Its interaction with fish of enormous value makes this drug particularly important. In the present study, the influences of MTZ on some humoral parameters of common carp were investigated. MTZ is used as an antibacterial and antiparasitic antibiotic, there is little information about its potential influence on the immune system cellularity and function.

This study has revealed that MTZ caused significant increase in the RBC, PCV, Hb values. The probable reason for this can be that MTZ has the potential to stimulate erythropoietin release from the kidneys with a resultant increase in fish RBC production (erythropoiesis) which could ultimately induce polycythemia. Increased oxygen demand at tissue due to stress may lead to increase in RBC and Hb as they are directly involved in oxygen transportation. WBCs play a pivotal role in fish immunology they constitute the cellular component of innate

Table 1: Value of different hematological Parameters in fish after treatment with 5mg/l and 10mg/l concentration of MTZ and control group (Mean±S.E.)

| Hematological Parameters | MTZ | Control | 24 hours | 48 hours | 72 hours | 96 hours |
|--------------------------|---------|-----------|--------------|----------------|---------------|---------------|
| Hb | 5mg/l | 6.55±0.03 | 8.05 ± 0.38* | 8.15±0.14* | 9.35±0.70*** | 9.7±0.05*** |
| | 10 mg/l | 6.35±0.03 | 9.5± 0.05*** | 8.4±0.11*** | 12.75±0.40*** | 10.50±0.30*** |
| PCV | 5mg/l | 33.5±0.3 | 29 ±0.5 | 25±2.8* | 31.5±3.7 | 28±1.10 |
| | 10 mg/l | 28.5±0.3 | 39 ±0.44*** | 31.5±1.10 | 40.60±0.40*** | 30.6±0.30 |
| RBCs Count | 5mg/l | 1.8±0.03 | 2.2 ±0.10 | 4.1±0.20*** | 4.15±0.03*** | 6.8±0.10*** |
| | 10 mg/l | 2.2±0.03 | 2.45 ±0.03 | 3.70±0.11*** | 4.60±0.40*** | 7.25±0.03*** |
| WBCs count | 5mg/l | 23.7±0.3 | 21.5 ±0.30 | 22.6±1.7 | 19.60±0.30 | 17.40±0.50** |
| | 10 mg/l | 21.8±0.3 | 23.80± 0.50 | 23.2±1.20 | 18.50±0.30** | 17.1±0.20*** |
| MCV | 5mg/l | 187.5±0.2 | 133.30 ±9.60 | 62.70±10.50*** | 74.70±8.80*** | 41.10±1.00*** |
| | 10 mg/l | 126.5±0.3 | 156.00 ±2.30 | 85.70±6.5*** | 89±7.80*** | 41.3±0.20*** |
| MCH | 5mg/l | 35.6±0.24 | 36.60 ±0.50 | 19.95±0.80*** | 22.20±1.60*** | 14.2±0.2*** |
| | 10 mg/l | 28.1±0.3 | 38.00 ±0.20* | 23.40±0.03*** | 28.10±1.60 | 13.70±0.03*** |
| MCHC | 5mg/l | 18.6±0.4 | 27.80 ±1.60 | 34±4.50* | 37.40±2.60** | 38.70±1.20* |
| | 10 mg/l | 22±0.3 | 24.30 ±0.20 | 27.00±1.70** | 31.90±1.10*** | 33.80±0.30*** |

*-($p \leq 0.05$); **-($p \leq 0.01$); ***-($p \leq 0.001$)

immunity. MTZ has been proven to elicit suppression in the primary lymphoid organ bone marrow. The hemotoxic effects obtained in the current study can be explained on the basis that, MTZ when used in the higher dose may lead to the suppression of the bone marrow (El-Nahas and El Ashmawy, 2004). Further Tomaszewski (1997) have observed that change in hematopoietic organ hinder granulopoiesis and lymphopoiesis. In a study, the lymphocytes proliferation assay revealed that MTZ has been reported to impart inhibitory effects on blood lymphocytes proliferation. (Fararjeh *et al.*, 2008). WBC count is a cellular innate immunity parameter of common carp and is an indicator of cellular defense. The MTZ acts as antibacterial and antiparasitic agent, thus leads to lesser infection load which is probably in agreement with declined WBC count in this study. The decrease in white blood cell (WBC) count observed in the treatment groups could potentially be attributed to the secretion of epinephrine in response to stress. Epinephrine has the ability to induce spleen contraction and subsequently lead to a reduction in the number of leukocytes. This decline in leukocyte count may ultimately compromise the effectiveness of the immune system (Svoboda, 2001; Witesta, 2003). This supports the finding of our study w.r.t WBC count, which showed a decline trend towards 96 hours of drug administration. Similar decrease is also observed by Han *et al.* (2014) in MTZ-treated fish

in a concentration-dependent manner compared to the control. Decrease in the value of MCV has been observed which may be due to defective hemoglobin synthesis which results in micro cystic anemia. Increased value of MCHC may be due membrane loss and consequent spherical shape attained by the cell.

These consequences indicate immunosuppressive effects subsequently increase fish susceptibility to bacterial, viral and fungal diseases. Finally, the current investigation demonstrates that MTZ inhibits both humoral and cell-mediated immune responses. The current study's findings suggest that MTZ modifies immune function since it caused alterations in these parameters. Additionally, the study's findings reveal the hemotoxicity of MTZ in fish *C. carpio* when exposed to concentrations of 5mg/L and 10mg/L. Significant dose dependent changes in the values of the assessed haematological parameters are observed. However, the changes are more severe at 10 mg/L concentration.

CONCLUSION

The current investigation results show that MTZ exhibits inhibitory properties on both humoral and cell-mediated immune responses. Findings of the present study would be useful to demonstrate the adverse effects of MTZ use, emphasizing the

importance of the effect on fish which could be very important concerning public health.

ACKNOWLEDGMENTS

The authors are thankful to Dean COVAS, CSKHPKV, Palampur, H.P., India for providing laboratory facility.

REFERENCES

- Aldrin, J.F., Laurenciu Messenger, F.B. (1982). La Biochimie Clinique en Aquaculture, Interet et Perspective CNEXO, Actes Colloq nr., 291-326.
- Anoob, F., Arachchi, S., Azamathulla, H., Najib Almahbashi and Rathnayake, U. (2024). Nanoadsorbents as an effective wastewater treatment candidate for pharmaceutical contaminants; towards sustainable policy development. *Case Studies in Chemical and Environmental Engineering*, 9, 100639.
- Barus, V., Peaz, M. and Kohlmann, K. (2001). *Cyprinus carpio* (Linnaeus, 1758). In Banarescu PM, Paepke HJ, editor. The freshwater fishes of Europe, v. 5/III; Cyprinidae 2/III, and Gasterosteidae. Germany: AULA-G GmbH Wiebelsheim, Pp. 85–179.
- Blaxhall, P.C. (1972). The haematological assessment of the health of freshwater fish. *Journal of Fish Biology*, 4: 593-604.
- Cavas, T. and Ergene, S. (2005). Genotoxicity evaluation of metronidazole using the piscine micronucleus test by acridine orange fluorescent staining. *Environmental Toxicology and Pharmacology*, 19(1): 107-111.
- El-Nahas, A.F. and El-Ashmawy, I.M. (2004). Reproductive and cytogenetic toxicity of metronidazole in male mice. *Pharmacology and Toxicology*, 5: 226–31.
- FAO (2012). Fishstate plus: Universal software for fishery statistical time series (available at: www.fao.org/fi/statist/fisoft/fishplus.asp).
- FAO (2013). Fishstate plus: Universal software for fishery statistical time series (available at: www.fao.org/fi/statist/fisoft/fishplus.asp).
- Fararjeh, M., Mohammad, M.K., Bustanji, Y., AlKhatib, H. and Abdalla, S. (2008). Evaluation of immunosuppression induced by metronidazole in Balb/c mice and human peripheral blood lymphocytes. *International Immunopharmacology*, 8: 341–350.
- Han, J., Zhang, L., Yang, S., Wang, J. and Tan, D. (2014). Detrimental effects of metronidazole on selected innate immunological indicators in common carp (*Cyprinus carpio* L.). *Bulletin of Environmental Contamination and Toxicology*, 92(2): 196-201.
- Jain, N.C. (1986) Schalm's Veterinary Haematology. 4th Edition, Lea and Febiger, Philadelphia, PA, 1221.
- Jamalzadeh, H.R., Keyvan, M. R. Ghomi, F. and Gherardi (2009). Comparison of blood indices in healthy and fungal infected Caspian Salmon (*Salmo truttacaspus*). *African Journal of Biotechnology*, 8(2):319-322.
- Johnson, P.J. (1993). Metronidazole and drug resistance. *Parasitol. Tod.*, 9: 183–186.
- Jousimies-Somer, H., Asikainen, S., Suomala, P. and Summanen, P. (1988). Activity of metronidazole and its hydroxy metabolite against clinical isolates of actinobacillus actinomycetemcomitans. *Molecular Oral Microbiology*, 3: 32–34.
- Kollner, B., Wasserrab, B., Kotterba, G. and Fischer, U. (2002). Evaluation of immune functions of rainbow trout (*Oncorhynchus mykiss*)-how can environmental influences be detected?. *Toxicology Letters*, 131: 83–95.
- Kümmerer, K. (2004). Resistance in the environment. *Journal of Antimicrobial Chemotherapy*, 54: 311–320.
- Li, Z.H., Velisek, J., Zlabek, V., Grabic, R., Machova, J. and Kolarova, J. (2011). Chronic toxicity of verapamil on juvenile rainbow trout (*Oncorhynchus mykiss*): effects on morphological indices, hematological parameters and antioxidant responses. *Journal of Hazardous Materials*, 185: 870–880.

- Marta, W., Joanna, M., Anna, B., Magda, C. and Piotr, S. (2015). Determination of metronidazole residues in water, sediment and fish tissue samples. *Chemosphere*, 119: S28–S34.
- Nwokediegwu, Z.Q.S., Daraojimba, O.H., Oliha, J.S., Obaigbena, A., Dada, M.A., Majemite, M.T. (2024). Review of emerging contaminants in water: USA and African perspectives. *International Journal of Science and Research Archive*, 11(01): 350–360.
- Radu, D., Oprea, L., Bucur, C., Costache, M., Oprea, D. (2009). Characteristics of Haematological Parameters for Carp Culture and Koi (*Cyprinus carpio* Linnaeus, 1758) Reared in an Intensive System. *Bulletin of University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca. Animal Science and Biotechnologies*, 66: 1-2.
- Rahman, M.M. (2015). Role of common carp (*Cyprinus carpio*) in aquaculture production systems. *Frontiers in Life Science*, 8:399-410.
- Rediguieri, C.F., Porta, V., Nunes, D.S.G., Nunes, T.M., Junginger, H.E., Kopp, S., Midha, K. K., Shah, V.P., Stavchansky, S., Dressman, J.B. and Barends, D.M. (2011). Biowaiver monographs for immediate release solid oral dosage forms: metronidazole. *Journal of Pharmaceutical Sciences*, 100(5): 1618–1627.
- Richardson, M.L., Bowron, J.M. (1985). The fate of pharmaceutical chemicals in the aquatic environment. *Journal of Pharmacy and Pharmacology*, 37, 1-12.
- Rizzo, A., Paolillo, R., Guida, L., Annunziata, M., Bevilacqua, N. and Tufano, M.A. (2010). Effect of metronidazole and modulation of cytokine production on human periodontal ligament cells. *International Immunopharmacology*, 10: 744–750.
- Rodriguez, L., Livengood, E.J., Miles, R.D. and Chapman, F.A. (2011). Uptake of metronidazole in *Artemia* at different developmental life stages. *Journal of Aquatic Animal Health*, 23: 100–102.
- Sánchez-Polo, M., Rivera-Utrilla, J., Prados-Joya, G. and Ocampo-Pérez, R. (2012). Metronidazole photodegradation in aqueous solution by using photosensitizers and hydrogen peroxide. *Journal of Chemical Technology & Biotechnology*, 87:1202–1208.
- Singh, N.N. and Srivastava, A.K. (2010). Haematological parameters as bioindicators of insecticide exposure in teleosts. *Ecotoxicology*, 19: 838–854.
- Svoboda, M. (2001). Stress in fishes (a review). *Bull. VURH Vod.*, 4:169- 191
- Tomaszewski, J.J. (1997). Diagnosykala boratoryjna (laboratory diagnostics). *PZWL Warszawa*, 36(4):73 -76.
- Witeska, M. (2003). The effects of metals (Pb, Cu, Cd, and Zn) on hematological parameters and blood cell morphology in common carp. *Rozprawa naukowa*, 72:113
- Wu, Y. and Fassihi, R. (2005). Stability of metronidazole, tetracycline HCl and famotidine alone and in combination. *International Journal of Pharmaceutics*, 290: 1–13.

Received: July 19, 2024

Accepted: July 30, 2024