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Zoology

EVALUATION OF PROTEIN, LIPID PEROXIDATION AND REDUCED GLUTATHIONE LEVEL BETWEEN THE MATURE (CLITELAED) AND IMMATURE (NON CLITALATED) EARTHWORM, *EUDRILUS EUGENIAE*

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ABSTRACT Earthworms *(Eudrilus eugeniae)* were taken from the soil chemist office and acclimatized in the vermiculture bed, prepared by the laboratory for 7 days prior for different research work. The whole body of the earthworm was homogenized and processed for the biochemical analysis. Both the matured and immature earthworms were collected from the bed and the antioxidant parameter s are measured and compared by paired t-test. The lipid peroxidation (LPX), reduced glutathione (GSH) and protein content were measured and compared. Protein content was more in immature (clitellated) than in mature (non clitellated). Likewise, the GSH content (mg/g tissue) and the LPX content (mg/g tissue) were more in immature (clitelated) than in mature (non clitelated). The present work is to analyze the level of antioxidant parameter that are present in between the mature and immature earthworm.

KEYWORDS : Mature, Immature, Eudrilus Eugeniae

INTRODUCTION

Earthworms play critical roles in improving the permeability and water retention of the soil AND are one of the animals with the largest biomass in soil (Roubalova *et al.,* 2015). Earthworms play an important role in the soil ecosystem and can decompose organic matter into nutrients in the soil (Datta *et al.,* 2016). Earthworms are more sensitive to pollutants that are occurred in the soil and the water rather than the other soil organisms. Thus it is regarded as the model organism for soil (Song *et al.,* 2009). It is observed that the abundance and distribution of earthworm populations can affected by entering the pesticides into the soil. The pesticides also alter the enzyme activities and also damage the cell structure of the earthworm (Liang *et al.,* 2016; Wang *et al.,* 2016). The evaluation of traditional toxicity from the pollutants by earthworms is mainly based on ROS (Reactive Oxygen Species) level, MDA (Mass Drug Administration)content, antioxidant enzyme activity and acute toxicity (Wu *et al.,* 2012; Xu *et al.,* 2013). The traditional evaluation methods are no longer sufficient for comprehensive evaluation of pollutants. Thus the comprehensive evaluation methods such as transcriptomics, metabolomics, and proteomics are being necessary for this purpose after the development of molecular biology (Huang *et al.,* 2020; Zhang *et al.,* 2021). Thus, earthworms work as an indicator to reflect the effects of pesticides on soil biology (Vasseur and Bonnard, 2014).

MATERIAL AND METHODS

Collection And Preparation Of The Sample

The earthworm, *Eudrilus eugeniae* were purchased from soil conservation office, Markona, Balasore and were acclimatized for seven days in the laboratory condition prior to the experiment. The earthworm, were cultured in the laboratory around the year and earthworms are collected randomly from bed prior for the comparison between the content of the biochemical parameter in different stages of the life cycle. Thus 5-6 in number were picked up from each tray and their whole-body weight was measured. A 10% homogenate was prepared with phosphate buffer (pH 7.4) The supernatant was taken for biochemical assay.

Methods

Estimation of different biochemical parameter in different stages of life cycle were done by several methods such as Protein by Lowry *et al.,* (1951), reduced glutathione (GSH) by Ellman (1959), Lipid peroxidation assay by Ohkawa *et al.*, (1979).

RESULT

Protein content was highest in immature (Fig.1) and it was 38.733 ±4.858, 41.058± 4.851 in mature and immature respectively in *Eudrilus eugeniae.* The paired t-test reveals that the protein content of immature earthworm is higher than the protein content of mature ones as the mean difference of mature is less than the mean difference of immature earthworm. LPX content was highest immature (Fig.2). The LPX content (mg/g tissue) was 1.440±0.250, 1.460±0.378 in mature and immature respectively in *Eudrilus eugeniae.* The paired t-test reveals that the lipid peroxidation content of immature earthworm is higher than the LPX content of mature ones as the mean difference of mature is less than the mean difference of immature earthworm. GSH content was highest in IMmature (Fig.3) and it was 0.161±0.05, 0.273 ± 0.1 in mature and immature respectively in *Eudrilus eugeniae.* The paired t-test reveals that the reduced glutathione content of mature earthworm is higher than the GSH content of immature ones as the mean difference of mature is less than the mean difference of immature earthworm. The paired t-test reveals that the reduced glutathione content of mature earthworm is higher than the protein content of immature ones as the mean difference of mature is less than the mean difference of immature earthworm.

DISCUSSION

protein is one of the major building blocks and an important source of energy for animal tissue. Reports on the effects of organic matter in soil on earthworm tissue protein are rare. However, it is well known that protein contributes significantly to the total biomass in animals (Fonte et al., 2009) and reported that earthworm population and average biomass were significantly higher in soil rich in organic matter. Lipid peroxidation is a chain reaction initiated by hydrogen abstraction or addition of oxygen radical, resulting in oxidative damage of polyunsaturated fatty acids (Halliwell and Gutteridge, 1984). The production of free radicals in the organism, and the imbalance between the concentrations of these and the antioxidant defences, may be related to processes such as ageing and several diseases. The ageing process has been described by various theories. In particular, the free radical theory of ageing has received widespread attention, which proposes that deleterious actions of free radicals are responsible for the functional deterioration associated with ageing. It attributes the ageing process to non-specific damage of macromolecules such as DNA, lipids, and proteins by free radicals. The theory predicts that the ability of cell to resist or prevent oxidative stress is a key determinant of longevity (Harman, 1956).

But here in the present work it is observed that season had a great impact on antioxidant system and enzymatic activity of the earthworm also affected by the season.

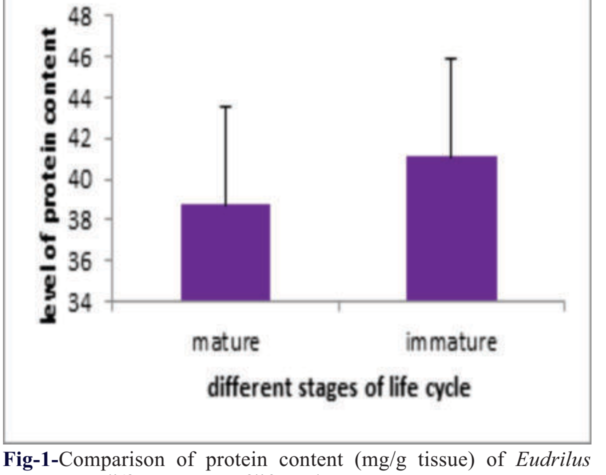

Fig-1-Comparison of protein content (mg/g tissue) of *Eudrilus eugeniae* at different stages of life cycle

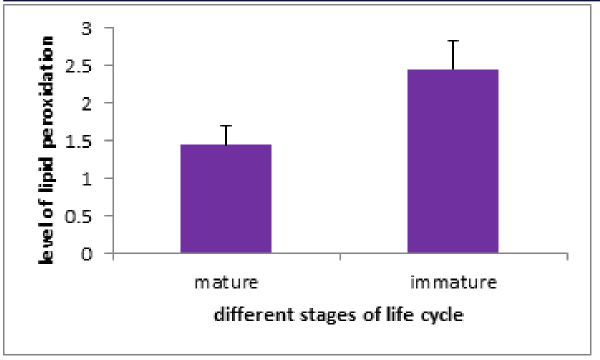

Fig-2-Comparison of lipid peroxidation content (mg/g tissue) of *Eudrilus eugeniae* at different stages of life cycle

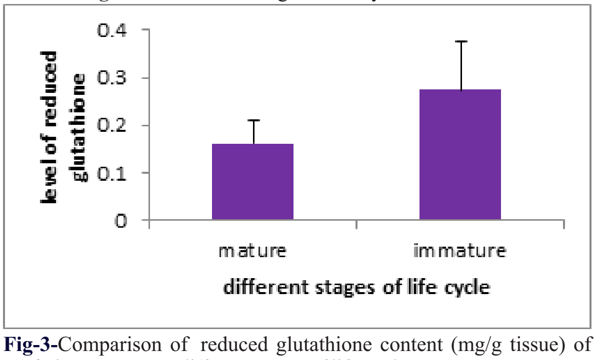

Fig-3-Comparison of reduced glutathione content (mg/g tissue) of *Eudrilus eugeniae* at different stages of life cycle

CONCLUSION

It was concluded that the antioxidant system of *E. eugeniae* is well developed to defend against the stress or reactive oxygen species produced due to exposure to various stress, it may be the variation of temperature in different stages of life cycle. When the different biochemical parameters were taken in consideration, it was concluded that the mature has more protein, LPX and GSH in comparison to the immature. Moreover, it might be assumed that several pesticides may be detoxified by earthworm to a small extent, but they have strong stress factors influencing the detoxification system of the animal. Long exposure might lead to deleterious effect on earthworm and limit their survival rate. The conclusions of this investigations are that *Eudrilus eugeniae* is a fast-growing and productive earthworm in animal waste i.e., ideally suited as a source of animal feed protein as well as for rapid organic waste conversion. It is more productive in terms of rates of growth than other species and would seem to be a suitable candidate for vermicomposting systems, in regions where the optimal temperature of 25°C is both feasible and economic. Although the large size of *E. eugeniae* makes it much easier to handle and harvest, than commonly-used species such *E. fetida* and *P. excavatus,* it is much more sensitive to disturbance and handling and may occasionally migrate from breeding beds. From the above study it is found that the antioxidant system of the mature is well developed than the immature.

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