

Protective Effect of *Portulaca quadrifida* on Rotenone Induced Locomotor Impairment in *Drosophila* Model

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Abstract:

Portulaca quadrifida Linn., family Portulacaceae, commonly known as Chicken weed is used as an edible food. It has been explored for medicinal property. The rotenone-induced locomotor impairment in *Drosophila* model is used in studying the potential neuroprotective activity. The aim of the study was to evaluate and compare neuroprotective activity *Portulaca quadrifida*. The whole plant material was subjected to successive extraction using elutropic solvent system to obtain hexane extract, chloroform extract, ethyl acetate extract and ethanol extract. The adult *Drosophila* flies were divided in to 6 different groups i.e., control, disease, HE, CE, EAE and EE group. The locomotor behavior of flies was evaluated by negative geotaxis assay. The neuroprotective activity was found prominent in hexane extract followed by chloroform extract of *Portulaca quadrifida*.

Keywords —*Drosophila melanogaster*, Neuroprotection, Negative geotaxis assay, *Portulaca quadrifida*, Rotenone..

I. INTRODUCTION

The neurodegenerative disorder is one of the debilitating disorders leading to morbidity worldwide. The major drawback of neurodegeneration is the inability of the degenerated neuron to regenerate[1], [2]. The neurodegeneration leads abnormal locomotor activity like imbalance, tremors, altered voluntary movements. The current therapy available is mainly targeting for the symptomatic relief and is associated with many adverse effects, which drastically hampers the patient compliance towards the treatment[3]. Thus, need of hour is for the agents which can exert neuroprotection with minimal side effect. The Indian traditional medicine has gained way better acceptance mainly due to the lesser and less severe adverse effect. Many herbs which have been consumed as vegetables have found to have medicinal value and folklore used as remedy in varied ailments[4].

The *Portulaca quadrifida* belonging to the family of Portulacaceae is one such herb which has been used for treating different ailments. Also, the various pharmacological activities are reported like neuropharmacological, anti-inflammatory, anti-diabetic, anticonvulsant. The plant is native to Africa and Asia. It is commonly known as chicken weed or baralunia[5], [6]. The present study was targeted to evaluate and compare the neuroprotective activity of different extracts of *Portulaca quadrifida* in *Drosophila* fly model.

II. MATERIALS AND METHODS:

A. Materials

The plant material was collected from Satpuda region and authenticated by Dr. Rajendra Shinde, St. Xavier's College, Mumbai (Herbarium number: 16926 dated 6/8/2018). The wild type flies were received as gift sample from IISER, Pune and was further cultivated in Bharati Vidyapeeth's College

of Pharmacy Animal house. Rotenone was purchased from Sigma Aldrich. All other reagents were used of analytical grade.

B. Successive extraction of *Portulaca quadrifida*

The whole dried plant was used for extraction with varying solvent polarity. Initially the plant was treated with hexane followed by chloroform, ethyl acetate and finally ethanol to obtain hexane extract (HE), chloroform extract (CE), ethyl acetate extract (EAE) and ethanol extract (EE). The content was stored in airtight container prior to study.

C. Negative geotaxis assay [7], [8]

The flies approximately 1 week old were divided randomly in different groups to get 16 flies in each group. The equal number of male and female flies were maintained in all groups. The flies were grown on culture media consisting mainly of corn starch, glucose, and yeast. The groups consisted of control group, disease group, treatments groups i.e., HE, CE, EAE and EE. The concentration of respective treatment group was 2.5 mg/ml. The flies of all group except control were exposed to rotenone (500 micromolar). The rotenone and the treatment drugs were added to the culture media and flies were exposed to it for 1 week. Followed by the exposure flies were analysed for the negative geotaxis assay and hence change in locomotor behaviour. Briefly, the flies were transferred to observation tube marked at 100mm height. The tube was tapped to settle all the flies at the bottom of the observation tube. The flies were observed for 1 min. The count of flies able to cross the 100 mm height were recorded. The locomotor behaviour was evaluated six times for all the groups.

III. RESULT:

The flies exposed to the rotenone had decreased locomotor activity in comparison to the control group flies; hence only 29.17 % of flies were able to cross the 100mm distance. While in case of HE, CE, EAE and EE treated flies the improvement in locomotor activity was noticed with 89.58,

85.42, 71.88 and 78.13 % respectively. The detailed data is reflected in Table 1.

TABLE 1. LOCOMOTOR BEHAVIOUR IN DROSOPHILA FLIES TREATED WITH DIFFERENT EXTRACTS OF *PORTULACA QUADRIFIDA*.

Treatment Groups	Total Number of Flies Escaped 10 cm Distance (Percent Locomotor Behaviour)
I (Vehicle)	15.00 ± 0.37* (94.79)
II (Disease)	4.67 ± 0.33 (29.17)
III (HE)	13.83 ± 0.31* (89.58)
IV (CE)	13.67 ± 0.42* (85.42)
V (EAE)	11.50 ± 0.43*# (71.88)
VI (EE)	12.67 ± 0.49*# (78.13)

Values are indicated as average ± S.E.M. *indicates significance (One way ANOVA with P > 0.01) as compared to disease group

IV. DISCUSSION

The drosophila flies also known as fruit flies share common physiology with humans and hence the study on flies can be extrapolated to predict the drug action in human beings. The fruit fly has a relatively shorter life span and maintenance is easier [9], [10]. The fruit fly model has been well explored to study the neuroprotection activity of the study drug. The locomotor activity is monitored as the ability of the fruit fly to fly across the marked distance [11]. Thus, the test drug treated flies if show better flying ability it is indicating improvement in the locomotor activity and hence the neuroprotective effect. The rotenone which is used as the inducing agent in this study is highly lipophilic which fastens the crossing of blood brain barrier. The rotenone inhibits the mitochondrial chain reaction which ultimately leads to damage to the mitochondria and hence decreased functioning of the neurons which are responsible for the locomotion [12]. In present study average number of flies i.e., 4.67 out of 16 were able to cross the fixed distance of 100 mm, indicating the locomotor disability as a virtue of mitochondrial dysfunction. The number of average flies pretreated simultaneously with rotenone and the different extracts of *Portulaca quadrifida* i.e., HE, CE, EAE and EE were 13.83, 13.67, 11.50 and 12.67 respectively. Thus, the increase in the number of flies of extract treated group specify the neuroprotective effect of *Portulaca quadrifida*. The

present study conclude that the hexane, chloroform and ethanol extract exert equally the neuroprotection and ethyl acetate was least effective with a statistical significance of $P > 0.01$.

V. CONCLUSIONS

It can be concluded that the *Portulaca quadrifida* extracts exert good neuroprotective activity by cause of improved locomotion in fruit fly. Further studies are to be designed to explore and understand the bio-actives present in respective extracts. Also, activity guided fractionation can be performed to identify the potential bioactive from the plant.

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