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Evaluation of the Protective Effect of Cucurbita Pepo Seeds Extract on Rotenone Induced Catalepsy in Swiss Albino Mice

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ABSTRACT: This study explores the extraction, phytochemical composition, and pharmacological effects of Cucurbita pepo L. ethanolic extract (EECPL). The extract, with an 18.5% yield, contains various bioactive compounds, including amino acids, terpenoids, alkaloids, tannins, flavonoids, saponins, steroids, and cardiac glycosides. Pharmacological testing showed that EECPL improved locomotor activity and motor coordination, as evidenced by behavioral tests such as the Actophotometer, Catalepsy Bar, and Narrow Beam Walking tests. Additionally, EECPL demonstrated antioxidant properties by increasing serum Superoxide Dismutase (SOD) levels and decreasing nitrite levels. These findings suggest that EECPL holds potential for therapeutic use in neuropharmacological and antioxidative applications.

KEYWORDS: Cucurbita pepo L, Ethanol, Neuropharmacological

I. INTRODUCTION:

Neurodegenerative disorders are chronic conditions that damage and destroy parts of your nervous system over time, especially your brain. These conditions are permanent and incurable, but many are now treatable thanks to medical advances. Currently, the main goal is to treat the symptoms and slow the progress of these conditions when possible. It's marked by the gradual loss of neurons, a process called neurodegeneration, which can eventually lead to the death of these cells. This group of diseases includes amyotrophic lateral sclerosis (ALS), multiple sclerosis (MS), Parkinson's disease, Alzheimer's disease, Huntington's disease, multiple system atrophy, tauopathies, and prion diseases. Neurodegeneration can impact various regions of the brain, affecting different levels of neuronal networks, from the molecular to the systemic level.

As of now, there is no known cure for these conditions since the progressive loss of neurons cannot be reversed. However, research has highlighted oxidative stress and inflammation as key contributors to neurodegeneration. Additionally, studies in biomedical science have found commonalities among these diseases at the cellular level, such as abnormal protein formations (known as proteinopathy) and the triggering of cell death. These similarities suggest that progress in treating one neurodegenerative disease might offer benefits for others as well.

1.1 Parkinson's Disease (PD):

Parkinson's disease is a progressive disorder of the nervous system that impairs movements control. It occurs due to the loss of dopamine – producing neurons in the substantia nigra, a region of brain. This loss leads to the symptoms such as tremors, muscle stiffness, slow movements and difficulties with balance and coordination. Over time, it can also affect other aspects of motor function.

- **Symptoms:** Tremors, stiffness, slowness of movement, and postural instability.
- **Pathology:** Involves the loss of dopamine-producing neurons in the substantia nigra and the accumulation of Lewy bodies (abnormal aggregates of protein).
- **Affected area:** Primarily affects the basal ganglia, leading to motor symptoms.

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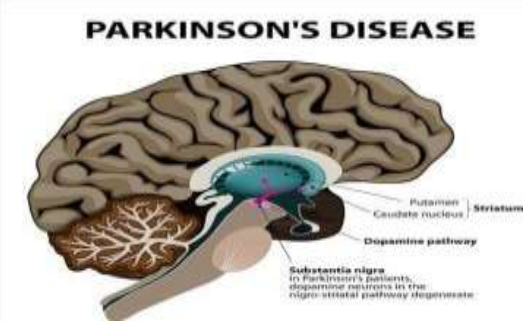

Fig: 1 Parkinson's Disease

II. PLANT PROFILE:

2.1 CUCURBITA PEPO:

Fig: 2 Cucurbita Pepo

Botanical name: Cucurbita pepo
Family: Cucurbitaceae

Kingdom	Plantae
Phylum	Tracheophyta
Class	Magnoliopsida
Order	Cucurbitales
Family	Cucurbitaceae
Genus	Cucurbita
Species	Cucurbita pepo L.

Table : 1 Taxonomical Classification

III. MATERIALS AND METHODS:

3.1 COLLECTION AND AUTHENTICATION OF PLANT:

The seed from Cucurbita pepo were collected from the natural habitat in and local areas, Tamilnadu and the plant material were authenticated by Dr.KN . Sunil kumar ., research officer /Sci- (II) HOD , Department of Pharmacognosy and by Dr. P. Elankani ., research officer (siddha), Sci – (II) / Incharge , SIDDHA CENTRAL RESEARCH INSTITUTE (central council for research in siddha ministry of AYUSH , Government of India) , Arumbakkam , Chennai – 601106.

3.2 PHYTOCHEMICAL STUDIES:

- Determination Of Ash Values
- Water Soluble Ash
- Determination Of Extractive Values
- Determination of water soluble extractive

3.3 PHYTOCHEMICAL TEST:

The ethanolic extracts of **Cucurbita Pepo.L** were subjected to the following preliminary phytochemical analysis

- Test for Carbohydrates
- Test for Alkaloids
- Test for Steroids and Sterols
- Test for Glycosides
- Test for Saponins
- Test for Flavonoids
- Test for Tri-terpenoids
- Test for Terpenoids
- Tests for Tannins and Phenolic Compounds
- Test for Gums and Mucilage
- Test for Proteins and Amino acids
- Test for Fixed Oils and Fatty acids

3.4. EXTRACTION OF SELECTED SEEDS:

Soxhlet procedure was used to extract oils from the pumpkin seeds. About 20g of crushed pumpkin seeds were fed to lab-scale Soxhlet extractor with condenser and 250ml round bottom flask. Then, 200ml of the food grade solvent (Ethanol) was added into round bottom distillation flask. The Soxhlet apparatus was then heated up using a heating mantle with temperature controller for controlling the desire value of heating. After heating for the predetermined time, the waded thimble of sample was obtained. The condensed was collected into the flask. This extraction process was carried out for predetermined time. After extracting, the flask containing the solvent and lipid was removed through distillation process. To remove the excess solvent, the solvent and extract mixture were placed on water bath. Excess solvent (Ethanol) can be evaporated at 70℃.Since the have low volatility, they retained in the flask. The extract mixture was found to be golden yellowish color. Further, this mixture was allowed to settle for 4 hrs to collect as extract and solvent as raffinate. Observed essential were separated and kept in refrigerator at 40℃ for further purification and characterization.

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3.5 EXPERIMENTAL DESIGN:

- **Animal:** Wistar albino Rat
- **Sex:** Male
- **Age:** 6-8 weeks
- **Animal number:** 25 Nos
- **Materials and methods**
- **Chemical used:**
- **Inducing agent :** Rotenone (2 mg/kg) administered subcutaneously (s.c.)
- **Test drug**:
- EECPL = ETHANOLIC EXTRACT OF CUCURBITA PEPO L
- Low dose (200 mg/kg) and High dose (400 mg/kg)

Vehicle :

- 0.5 % carboxy Methyl Cellulose (CMC) (5ml/Kg) administered orally (p.o.)
- Sunflower oil (1ml/kg) administered through subcutaneously (s.c.)

3.6 STUDY DESIGN:

Totally 25 male wistar albino rats were randomly divided into five groups each group contains five animals (n=5)

- **Group I (Control groups) (n=5)** – receives 0.5% CMC administered orally (p.o.) for about 28 days + sunflower oil (1mg/ml) was administered through subcutaneously (s.c.) for about 28 days (Both are administered simultaneously for about 28 days)
- **Group II (Negative Control groups) (n=5)**- receives rotenone (2mg/kg) emulsified in sunflower oil (2mg/ml) were given subcutaneously (s.c.) for about 28 days.
- **Group III (Standard groups) (n=5)**- received levodopa + Carbidopa (100mg/kg + 25 mg /kg) dissolved in 0.5% CMC for about 28 days.
- **Group IV (Low dose groups) (n=5)**- received rotenone (2mg/kg) emulsified in sunflower oil (2mg/ml) were given subcutaneously (s.c.) + EECPL = Ethanolic Extract Of Cucurbita Pepo L (200 mg/kg) administered orally (p.o.) for about 28 days.
- **Group V (High dose groups) (n=5)**- received rotenone (2mg/kg) emulsified in sunflower oil (2mg/ml) were given subcutaneously (s.c.) + EECPL = Ethanolic Extract Of Cucurbita Pepo L (400 mg/kg) administered orally (p.o.) for about 28 days.

3.7 PHARMACOLOGICAL STUDY:

- Actophotometer
- Catalepsy Bar Test
- Narrow Beam Walking Test
- Assessment of Serum Superoxide Dimutase
- Assessment Of Nitrite level

IV. RESULTS:

4.1 EXTRACTION APPEARANCE AND PERCENTAGE YIELD:

Drug	Ethanolic Extract Of Cucurbita Pepo L
Solvent	Ethanol
Colour	Dark Yellow
Consistency	Semi solid
Percentage yield	18.5 % w/w

Table: 2 Appearance and Percentage Yield Of EECPL

4.2 PRELIMINARY PHYTOCHEMICAL ANALYSIS:

S.No	Physio-Chemical Constant	Ethanolic Extract Of Cucurbita Pepo L
1	Total Ash	8.1±1.8
2	Acid Insoluble Ash	1.2±1.5
3	Water Soluble Extractive	31.5±1.6
4	Loss On Drying	7.6

Table : 3 Preliminary Phytochemical Analysis

4.3 PRELIMINARY PHYTOCHEMICAL SCREENING:

The Curcubita pepo extract was macerated separately. The residue were collected and the chemical test was conducted separately. Various phyto constituents were demonstrated and result were obtained. The results of this phytochemicals analysis is listed below

S.NO	CONSTITUENTS	ETHANOLIC EXTRACT
1.	Amino acids	+
2.	β-sitosterol	+
3.	Terpenoids	+
4.	Alkaloids	+
5.	Tannins	+
6.	Flavonoids	+
7.	Saponins	+
8.	Steroids	+
9.	Phenolic acid	+
10.	Cardiac glycoside	+

Table : 4 Preliminary Phytochemical Screening

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4.4 PHARMACOLOGICAL STUDIES:

4.4.1 Effect Of EECPL On Actophotometer:

S.NO	GROUPS	Locomotor Index		
		DAY 14	DAY 21	DAY 28
1	Control	81 ±1.093	82.4 ±0.567	79.3 ±1.089
2	Negative control	55 ±0.854	44 ± 1.38	31.6 ±1.154
3	Standard	75.8±1.131	78.6 ± 1.176	78 ± 1.517
4	Low dose 200 mg/kg	59.3±0.909****	63.4 ±0.406****	61.3 ±0.802****
5	High dose 400 mg/kg	72.8±1.137**	75.9 ± 1.467ns	81.4 ±1.078ns

Table: 5 Effect Of EECPL On Actophotometer

Values are represented in Mean ± SEM, n=5.
Comparison: Group III vs Group IV and Group V
Statistical significance test for comparison was done by one way ANOVA followed by Dunnet's 't' test, ns – non-significant, *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

4.4.2 Effect of EECPL On Catalepsy Bar Test:

S. NO	GROUPS	Catalepsy Behaviour		
		DAY 14	DAY 21	DAY 28
1	Control	2.3 ±0.231	3.1 ±0.332	3.1 ±0.318
2	Negative control	14.9 ±0.3	21.2 ±0.34	31.4 ±1.22
3	Standard	13.6 ±1.01****	7.7 ±0.371*****	4.9 ±.311*****
4	Low dose 200 mg/kg	18.3 ±0.3****	13.2 ±0.4****	8.1 ±0.55**
5	High dose 400 mg/kg	12.6±0.786****	9.2 ±0.311****	4.6 ±0.1****

Table: 6 Effect Of EECPL On Catalepsy Bar Test

Values are represented in Mean ± SEM, n=5.
Comparison: Group II vs Group III, Group IV and Group V
Statistical significance test for comparison was done by one way ANOVA followed by
Dunnet's 't' test, ns – non-significant, *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

4.4.3 Effect Of EECPL On Narrow Beam Walking Test:

S.NO	GROUPS	Time To Cross Narrow Beam In Seconds		
		DAY 14	DAY 21	DAY 28
1	Control	3.4 ± 0.245	4.2 ±0.374	5.2 ±0.583
2	Negative control	15.8 ±0.374	226 ±0.678	29.25 ±0.587
3	Standard	12.8 ±0.583***	10 ±0.316****	6.8 ±0.735*****
4	Low dose 200 mg/kg	25 ±1.517*****	10.8 ±0.374****	8 ±0.417*****
5	High dose 400 mg/kg	14.2 ±0.374ns	7.8 ±0.38*****	4.85 ±0.2****

Table: 7 Effect Of EECPL On Narrow Beam Walking Test

Values are represented in Mean ± SEM, n=5.
Comparison: Group III vs Group IV and Group V
Statistical significance test for comparison was done by one way ANOVA followed by Dunnet's 't' test, ns – non-significant, *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

4.4.4 Effect Of EECPL On Narrow Beam Walking Test (No. Of. Slips):

S.NO	GROUPS	No. of. slips in seconds		
		DAY 14	DAY 21	DAY 28
1	Control	1.5 ± 0.231	1.2 ±0.2	1.8 ± 0.374
2	Negative control	10.5 ±0.241	12.6 ±0.113	15.7±0.1
3	Standard	11.7 ±0.715ns	7.29 ±0.2*****	6.2 ±0.2

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4	Low dose 200 mg/kg	12***	11.7 ±0.222 ns	7.4 ±0.3
5	High dose 400 mg/kg	14.7 ±0.503****	8.4 ±0.241****	5.0 ±0.1

Table: 8 Effect Of EECPL On Narrow Beam Walking Test (No. Of. Slips)

Values are represented in Mean ± SEM, n=5.
Comparison: Group III vs Group IV and Group V
Statistical significance test for comparison was done by one way ANOVA followed by Dunnet's 't' test, ns – non-significant, *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

4.4.5 Effect Of EECPL On Serum Superoxide Dimutase (SOD):

S.NO	GROUPS	SOD VALUES (U/mg)
1	Control	96.77 ± 1.498
2	Negative Control	43.88 ± 0.554
3	Standard	86.12 ± 0.514
4	Low dose 200 mg/kg	62.99 ± 0.902****
5	High dose 400 mg/kg	76.81 ± 0.921****

Table: 9 Effect Of EECPL On Serum Superoxide Dimutase (SOD)

Values are represented in Mean ± SEM, n=5.
Comparison: Group III vs Group IV and Group V
Statistical significance test for comparison was done by one way ANOVA followed by Dunnet's 't' test, ns – non-significant, *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

4.4.6 Effect Of EECPL On Nitrite Level:

S.NO	**GROUPS**	**Nitrite level (µg/ml)**
1	**Control**	131.6 ± 2.631
2	**Negative Control**	292.5 ± 6.031
3	**Standard**	161.6 ± 2.799****
4	**Low dose 200 mg/kg**	253.7 ± 2.011****
5	**High dose 400 mg/kg**	208.7 ±4.077****

Table: 10 Effect Of EECPL On Nitrite Level

Values are represented in Mean ± SEM, n=5.
Comparison: Group III vs Group IV and Group V
Statistical significance test for comparison was done by one way ANOVA followed by Turkey test, ns – non-significant, *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.

V. CONCLUSION:

The ethanolic extract of Cucurbita pepo L. (EECPL) demonstrates significant pharmacological activity, including improvement in motor coordination, increased locomotor activity, and enhanced antioxidant properties as evidenced by changes in SOD and nitrite levels. The presence of various bioactive compounds in the extract supports its potential therapeutic applications, particularly in improving motor functions and oxidative stress-related conditions. The findings suggest that EECPL may be a promising candidate for further exploration in neuropharmacological and antioxidative therapies.

REFERENCE:

[1]. Alhawiti, A.O., Toulah, F.H. & Wakid, M.H. Anthelmintic Potential of Cucurbita pepo Seeds on Hymenolepis nana. Acta Parasit. **64**, 276–281 (2019). https://doi.org/10.2478/s11686-019-00033-z

[2]. Amin, M. Z., Rity, T. I., Uddin, M. R., Rahman, M. M., & Uddin, M. J. (2020). A comparative assessment of antiinflammatory, anti-oxidant and anti-bacterial activities of hybrid and indigenous varieties of pumpkin (Cucurbita maxima Linn.) seed oil. Biocatalysis and agricultural biotechnology, 28, 101767.

[3]. Chitra V, Manasa K*, Mythili A, TamilanbanvT, gayathiri K : Effect of hydroalcoholic extract of Achyranthes aspera on haloperidol induced Parkinson's disease in wistar rats.

[4]. "Cucurbita pepo: A Review of Its Pharmacological and Therapeutic Properties" (2020) by M. A. Khan et al., editors. Springer Nature.

[5]. Ekpono, E. U., Eze, E. D., Adam, A. M., Ibiam, U. A., Obasi, O. U., Ifie, J. E., ... & Aja, P. M. (2024). Ameliorative Potential of Pumpkin Seed Oil (Cucurbita pepo L.) Against Tramadol-Induced Oxidative Stress. Dose-Response, 22(1), 15593258241226913.

[6]. Ekpono EU, Eze ED, Adam AM, et al. Ameliorative Potential of Pumpkin Seed Oil (Cucurbita pepo L.) Against Tramadol-Induced Oxidative Stress. Dose-

Response. 2024;22(1). doi:10.1177/15593258241226913

[7]. Hanaa H. Ahmed, Mona Abdel-Rahman, Rania S. Ali, Ahmed E. Abdel Moniem : Protective Effect of Ginkgo Biloba Extract and Pumpkin Seed Oil Against Neurotoxicity of Rotenone in Adult Male Rats.

[8]. Hassan et al. (2020). Cucurbita pepo seed extract improves motor function and reduces oxidative stress in a rat model of Parkinson's disease. Journal of Pharmaceutical Sciences, 109(2), 833-842. DOI: 10.1016/j.xphs.2019.12.00

[9]. Jitendra O. Bhangale and Sanjeev R. Acharya : Anti-Parkinson Activity of Petroleum Ether Extract of Ficus religiosa (L.) Leave.

[10]. Jong-Bo Kim, Spandana Rajendra Kopalli and Sushruta Koppula : Cuminum cyminum Linn (Apiaceae) extract attenuates MPTP-induced oxidative stress and behavioral impairments in mouse model of Parkinson's disease.

[11]. Kim et al. (2018). Neuroprotective effects of Cucurbita pepo seed extract on SH-SY5Y cells. Journal of Ethnopharmacology, 211, 345-353. DOI: 10.1016/j.jep.2017.10.0.

[12]. K. Ray Chaudhuri,Eduardo Tolosa,Anthony H. V. Schapira,Werner Poewe : Non motor Symptoms of Parkinson's Disease.

[13]. Lee et al. (2019). Anti-inflammatory effects of Cucurbita pepo seed extract on microglial cells. Journal of Medicinal Food, 22(10), 1039-1046. DOI: 10.1089/jmf.2019.0103.

[14]. Manal K Abdel-Rahman : Effect of pumpkin seed (Cucurbita eo L.) diets on benign prostatic hyperplasia (BPH) : Chemical and morhometric evalution in rats, 2006.

[15]. Mani Rupesh Kumar, Madhura.M.Bose, Swati Mittal, A.Muthukumar, Md.Salahuddin : A systemic review on neuroprotective potential of cucurbita pepo on neurological disease, 2023.