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# EVALUATION OF THE ACTIVITY OF ARACHIS HYPOGAEA ON COMPLETE FREUND'S ADJUVANT-INDUCED ARTHRITIS IN RATS

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**ABSTRACT:**
Arthritis is a prevalent inflammatory condition that poses a significant burden on individuals and healthcare systems worldwide. This study aimed to evaluate the anti-arthritis activity of the ethanolic extract of Arachis hypogaea in a rat model of arthritis induced by Complete Freund's Adjuvant (CFA) injection. The extract exhibited significant reductions in paw edema volume compared to the control group, indicating its potential as an anti-inflammatory agent. Histopathological examination revealed a decrease in inflammatory cell aggregation and minimal tissue injury in the extract-treated groups, suggesting its ability to attenuate inflammation. Hematological parameters, including hemoglobin levels, RBC count, and WBC count, showed improvements in the extract-treated groups, indicating potential restoration of altered inflammatory markers. Biochemical analysis demonstrated a decrease in SGPT, SGOT, and alkaline phosphatase levels after treatment with the extract, indicating possible modulation of liver function. These findings support the potential of Arachis hypogaea as a natural anti-inflammatory agent for arthritis management. Further research is needed to elucidate its underlying mechanisms of action and isolate active compounds responsible for its therapeutic effects.

**Key Words:** Arachis hypogaea, arthritis, anti-inflammatory, histopathological examination, hematological parameters, biochemical parameters.

## INTRODUCTION

Arthritis is a highly prevalent and debilitating condition characterized by inflammation and joint pain. It poses a significant burden on individuals and healthcare systems worldwide. Although various treatment options exist, such as nonsteroidal anti-inflammatory drugs (NSAIDs) and disease-modifying antirheumatic drugs (DMARDs), they often have limitations, including adverse side effects and incomplete efficacy. Therefore, there is a need to explore alternative and complementary therapeutic strategies for arthritis management [1]. Arachis hypogaea, commonly known as peanuts, is a leguminous plant cultivated for its oil-rich seeds and renowned for its nutritional richness, including proteins, lipids, carbohydrates, vitamins, and minerals [2]. Peanut (Arachis hypogaea) finds widespread applications in traditional and pharmaceutical industries. It is utilized in ointments,

medicinal oils, and rectally for constipation relief. Peanut oil is employed in dermatology, baby care, and as a bath additive for various skin conditions. Additionally, the pharmaceutical and medical sectors use peanut oil as a vehicle for external, enteral, or parenteral medication preparations. The cosmetics industry utilizes peanut oil in skin, sun, and massage oils. Furthermore, peanut oil is consumed as a cooking oil and is believed to have cholesterol-lowering properties. The medicinal purposes of peanut include the use of its oil, seeds, and leaves. Peanuts contain various components such as flavonoids, phenolic acids, phytosterols, alkaloids, and stilbenes [3]. Notably, peanut protein constitutes a significant portion of the peanut kernel, consisting of albumin, globulins, and glutelins. However, peanuts lack essential amino acids such as lysine, methionine, and threonine. Raw peanuts and defatted flour contain varying quantities of amino acids. Peanut peptides, which offer better solubility, emulsifying capacity, and antioxidant properties compared to peanut protein isolate, have been investigated as potential food additives [4]. Peanuts exhibit a wide range of pharmacological effects, such as antioxidant, hypolipidemic, anti-inflammatory, opioid receptor affinity, sympathomimetic, immunomodulating, and anticancer activities. They have also demonstrated antimicrobial and antiparasitic effects. Certain peanut components affect endocrine function, such as aldosterone antagonism and goitrogenic activity. Peanut extracts have sedative effects and can promote sleep. Furthermore, peanut proteins and peptides have shown haemostatic and hypotensive properties. Thus, the use of peanuts and their bioactive constituents holds potential for various applications in medicine and health promotion [3]. To evaluate the activity of Arachis hypogaea in the management of arthritis, this study focuses on investigating its effects using the Complete Freund's Adjuvant (CFA)-induced arthritis model in rats. The CFA-induced arthritis model accurately mimics the pathological and immunological features of rheumatoid arthritis, making it a valuable tool for evaluating potential therapeutic agents [5]. Therefore, the aim of this study is to comprehensively assess the activity of Arachis hypogaea in the CFA-induced arthritis model in rats. Several parameters were evaluated in this study to assess the anti-arthritis activity and safety profile of the ethanolic extract of Arachis hypogoea. The parameters included in the evaluation were the morphological and physical characteristics of the plant material, the percentage yield of the extract, the presence of various phytochemical compounds, acute toxicity, paw edema volume and thickness, hematological parameters, histopathological examination, and biochemical parameters such as SGPT, SGOT, and alkaline phosphatase levels. By examining these parameters, we aim to determine the efficacy of Arachis hypogaea in alleviating arthritis symptoms and explore the potential underlying mechanisms of action. The findings from this study may contribute to the growing body of knowledge regarding natural products as potential therapeutic agents for arthritis. If proven effective, Arachis hypogaea could offer a safer and potentially more cost-effective option for arthritis management. Additionally, the identification and characterization of the active compounds within Arachis hypogaea could drive future research and drug discovery efforts. The importance of natural products in drug development cannot be overstated, as many current medications have their origins in natural compounds. By exploring the potential of Arachis hypogaea as an anti-arthritic agent, we can build upon this legacy and potentially uncover new therapeutic options for arthritis patients.

## MATERIALS AND METHODS

### *Plant Collection and Authentication*

Arachis hypogaea plants were collected from the surroundings of Kurnool on September 3, 2019. The plant was authenticated by Dr. KV Madhusudan, a botanist from Government College of Foremen, Kurnool, on September 14, 2020.

### *Drugs and Chemicals*

The following materials were used: CFA (Complete Freund's Adjuvant), Indomethacin, SGOT and SGPT kits from Agappe Diagnostic Center Ltd.

***Pharmacognostical Evaluation***

***Morphological Evaluations***

The morphological characteristics, including color and taste, were observed.

***Physical Evaluation***

*Total Ash Estimation:* Three grams of the powdered plant material was incinerated in a silica crucible to determine the total ash value, which indicates the purity and quality of the crude drug.

*Moisture Content Determination:* Five grams of the plant material was heated at 105°C in a hot air oven until a stable weight was obtained to measure the moisture content [6].

***Preparation of Arachis hypogaea Extract:***

The leaves were washed under running tap water to remove any contaminants. After drying in shade for 20 days, the leaves were powdered. Seventy grams of the dried powder was extracted with ethanol using a Soxhlet apparatus for 72 hours. The obtained extract was concentrated by flash evaporation at 50°C and stored in an airtight glass container at 4°C until further use. The percentage yield of the extract was calculated [7].

***Soxhlet Extraction:***

The powdered leaves were subjected to continuous hot extraction using a Soxhlet apparatus with ethanol as the solvent in a 1:3 ratio. The extraction was conducted until the solvent became colorless [8].

***Phytochemical screening***

Phytochemical screening of the Arachis hypogaea extract was carried out for the presence of alkaloids, anthraquinone glycosides, cardiac glycosides, tannins, chlorogenic acid, and flavonoids. These compounds were identified through specific tests, producing characteristic color changes or precipitates [9].

***Acute toxicity***

An acute oral toxicity study was conducted to assess the toxicity level of Arachis hypogaea. Twenty-one Albino mice weighing 25-30 g were used for the study. The mice were fasted overnight and provided only water. The test animals were divided into four groups of three mice each and administered increasing doses of ethanolic extract of Arachis hypogaea (5, 100, 200, 400, 800, 1600, and 2000 mg/kg). The animals were closely monitored for 24 hours for any visible signs of toxicity, including salivation, lachrymation, convulsions, hair loss, abnormal behavior (such as climbing, cleaning of face, aggressive behavior including biting and scratching, licking of tail, paw, and penis, vocalization, and intense grooming behavior), as well as diarrhea. Animal mortality was recorded after a 14-day observation period [10].

***Biological Activity***

To evaluate the potential anti-arthritic activity of Arachis hypogaea, a series of experiments were conducted using a rat model [11].

Six rats in each group were assigned as follows:

Group I: Normal saline (served as the normal group)

Group II: CFA (0.1 ml) injection (served as the disease group)

Group III: Indomethacin (10 mg/kg) injection (served as the standard group)

Group IV: CFA + EEAH at a low dose (200 mg/kg) (served as the ethanolic test group at the low dose)

Group V: CFA + EEAH at a high dose (400 mg/kg) (served as the ethanolic test group at the high dose)

Arthritis was induced by injecting 0.1 ml of Complete Freund's Adjuvant (CFA) into the subplantar region of the left hind paw of the rats. The paw volume was measured using a plethysmograph on various days up to 28 days post-CFA injection. Changes in the inflammatory reaction were assessed by recording paw volume.
On evaluation days (0, 7th, 14th, 21st, and 28th day), body weights of the animals were measured using a digital balance. Blood samples were collected through retro-orbital puncture for hematological and biochemical parameter estimation.

***Hematological Parameters***
Hematological parameters such as hemoglobin content, RBC count, WBC count, and Erythrocyte Sedimentation Rate (ESR) were analyzed using an automated hematology analyzer (cell counter BC3000 Mindray Company Plus) [12].

***Biochemical Estimation***
Serum parameters were measured, including:
Estimation of SGOT (AST) using the International Federation of Clinical Chemistry (IFCC) method [13].
Estimation of SGPT (ALT) using the IFCC method [13].

***Hematological Parameters*** **[14].**
ESR (Erythrocyte Sedimentation Rate) estimation.
RBC (Red Blood Cell) count estimation.
WBC (White Blood Cell) count estimation.
Hemoglobin (HB) estimation.

***Assessment of Arthritis***
The progression of Complete Freund's Adjuvant (CFA)induced arthritis was assessed by measuring the following parameters on day 0, 7, 14, 21, and other mentioned days:
Paw Volume: The swelling in the hind paw was measured periodically using a plethysmograph.
Paw Thickness: The difference in paw thickness between day 0 and subsequent time points was measured using a screw gauge to estimate edema.
Arthritis Score: Rats were scored daily based on visual criteria, with scores ranging from 0 (no change) to 4 (gross swelling and deformity) [15].

***Histopathological Examination***
Histopathological analysis was conducted on the 10th day at the end of the experiment. All animals were anesthetized with light ether anesthesia, followed by cervical decapitation for sacrifice. The left hind limb, which had induced arthritis, was removed just distal to the knee, washed with saline, and preserved in 10% formalin. The fixed tissues were decalcified, and slides were prepared for evaluation. The histopathological assessment included analyzing soft tissue swelling, bone demineralization, pannus formation, cartilage erosion, and joint space narrowing.

***Statistical Analysis***
The results were presented as means ± standard error of mean. Statistical analysis was performed using one-way ANOVA followed by Tukey's multiple comparison tests. A significance level of $p<0.05$ was considered.

**RESULTS**
***Pharmacognostic Study***
- Morphological evaluation revealed that Arachis hypogoea had a green color and a pungent odor.
- Physical evaluation of Arachis hypogoea showed a moisture content of 7.9% and a total ash value of 9.45%.

***Extract Percentage Yield***

The ethanolic extract of Arachis hypogoea yielded approximately 28%.

***Preliminary Phytochemical Studies***

Phytochemical screening of the extract and powder of Arachis hypogoea indicated the presence of proteins, glycosides, alkaloids, tannins, steroids, reducing sugars, saponins, and terpenoids.

**Table-1:** Preliminary phytochemical analysis of ethanolic extract of *Arachis hypogoea*

| **Phytochemicals** | **Test** | **EEAH** |
|---|---|---|
| Alkaloids | Mayer's test | + |
| | Wagner's test | + |
| | Dragendroff's test | + |
| | Hager's test | - |
| Tannins | Ferric Chloride test | + |
| | Catectrin | + |
| | Chlorogenic test | + |
| Cardiac glycosides | Baljet test | _ |
| | Legal's test | - |
| Steroids | Libermann-Buchard test | _ |
| Flavonoids | Alkaline Reagent test | _ |
| | Lead Acetate test | + |
| Glycosides | Bontrager's test | + |
| | Hydroxy anthraquinone test | _ |

+ indicates presence; - indicates absence.
EEAH: -Ethanolic Extract of *Arachis Hypogoea*

***Acute Toxicity Study***

The ethanol extract of Arachis hypogoea did not exhibit any mortality or toxic manifestations up to a dose of 2000 mg/kg body weight. Based on OECD 423 guidelines, therapeutic doses of 200 mg/kg and 400 mg/kg were selected for the ethanolic extract.

***Anti-Arthritis Activity***

Paw edema volume was significantly increased in rats injected with CFA compared to normal rats. Treatment with ethanolic extract of Arachis hypogoea at doses of 200mg/kg and 400mg/kg significantly reduced paw edema volume.

**Table 2: Observations, Signs of Toxicity, and Mortality in Control and Treated Groups**

| **Observation** | **Control group** | **Treated group** | **Signs of toxicity** | **Duration** | **Mortality** |
|---|---|---|---|---|---|
| **Alertness** | **N** | **N** | **NIL** | **24hrs** | **NIL** |
| **G rooming** | **N** | **N** | **NIL** | **24hrs** | **NIL** |
| **Touch response** | **N** | **N** | **NIL** | **24hrs** | **NIL** |
| **Pain response** | **N** | **N** | **NIL** | **24hrs** | **NIL** |
| **Limb position** | **N** | **N** | **NIL** | **24hrs** | **NIL** |
| **Grip strength** | **N** | **N** | **NIL** | **24hrs** | **NIL** |

**N= Normal**

***Effect on Paw Edema Volume***

Rats injected with CFA showed a significant increase in paw edema volume compared to the normal group. Treatment with ethanolic extract of Arachis hypogoea at both 200 mg/kg and 400 mg/kg doses led to a significant reduction in paw edema volume.

**Table 3: Changes in Paw Edema Volume Over Time**

| Group | Day 0 | Day 7 | Day 14 | Day 21 | Day 28 |
|---|---|---|---|---|---|
| Normal | 0.35 ±0022 | 0.35 ±0223 | 0.36±0.2108 | 0.36±0.0210 | 0.366±0.025 |
| Control | 0.36±0.210 | 0.716±0.040 | 0.93±0.61[d] | 1.00±0.051[d] | 1.01±0.047[d] |
| Standard 10 mg / kg | 0.36±0.210 | 0.783±0.60 | 0.716±0.60 | 0.5±0.051[a] | 0.4±0.025[a] |
| Extract 200 mg/kg | 0.35±0.022 | 0.816±0.60 | 0.8±0.057 | 0.75±0.099[b] | 0.65±0.428[b] |
| Extract 400mg/kg | 0.36±0.033 | 0.8±0.025 | 0.666±0.049 | 0.583±0.060[a] | 0.466±0.033[a] |

a=p<0.05, diseased compared with normal
b=p<0.01, diseased compared with normal
c=p<0.001, diseased compared with normal
d =p<0.05, diseased group compared with standard and test groups
e =p<0.01, diseased group compared with standard and test groups
f =p<0.001, diseased group compared with standard and test groups.

***Effect on Paw Thickness***

A gradual increase in paw thickness was observed in all groups throughout the observation period. The treatment with ethanolic extract of Arachis hypogoea at both 200 mg/kg and 400 mg/kg doses resulted in a significant decrease in paw thickness compared to the control.

**Table :4 Percentage inhibition of Paw swelling**

| Group | Day 7 | Day 14 | Day 21 | Day 28 |
|---|---|---|---|---|
| Control | --- | --- | --- | --- |
| Standard 10 mg / kg | 4.28 | 23.01 | 50.00 | 60.40 |
| Extract 200mg/kg | 0.24 | 13.98 | 25.00 | 35.64 |
| Extract 400 mg/kg | 0.20 | 28.38 | 41.70 | 53.86 |

***Effect on Body Weight***

A decrease in body weight was observed in the control group, while the treated groups showed either a slight increase or a maintenance of body weight, indicating a potential effect of the extract on weight control.

**Table 5: A. Hypogoea Effect on body weight**

| Group | MEAN BODY WEIGHT (gm) BEFORE / AFTER INDUCTION | | | | |
|---|---|---|---|---|---|
| | '0' Day | Day - 7 | Day – 14 | Day – 21 | Day - 28 |
| Normal | 266.7 ± 6.667 | 273 ± 7.211 | 291.7 ± 6.146 | 295 ± 5.774 | 306.7 ± 3.57 |
| Control | 305 ± 9.916 | 296.7 ± 9.888 | 275 ± 9.16 | 256.7 ± 9.888 | 240.8 ± 8.408 |
| Standard 10 mg / kg | 280 ± 10.33 | 274.2 ± 11.72 | 281.7 ± 15.63[b] | 288.3 ± 15.58[b] | 303.3 ± 14.06[a] |
| Extract 200 mg/kg | 285. ± 14.78 | 266.7 ± 12.82 | 256.7 ± 7.601 | 263.3 ± 7.601 | 275.8 ± 6.760[b] |
| EAC 400 mg / kg | 293.3 ± 10.22 | 292.5 ± 13.02 | 293.3 ± 11.23[b] | 298.3 ± 14.24[a] | 302.5 ± 8.827[a] |

a=p<0.001, b= p<0.01, c= p <0.05 (compared with disease control group-II)

***Hematological Parameters***

In the control group, there was a decrease in hemoglobin levels, RBC count, and WBC count, while the treated groups showed improvements in these parameters.

**Table 6: Hematological Parameters**

| Parameter | Group | 0 DAY | 7TH DAY | 14TH DAY | 21ST DAY | 28TH DAY |
|---|---|---|---|---|---|---|
| HEMOGLOBIN (gm %) | I | 10.26±1.546 | 10.28±±1.56 | 10.43±1.651 | 10.84±1.046 | 10.37±1.761 |
| | II | 10.45±1.65 | 9.10±1.237 | 9.06±0.003 | 7.85±2.651 | 7.15±2.452 |
| | III | 10.54±1.972 | 9.54±2.872 | 9.65±2.761 | 9.87±1.761[b] | 9.93±1.675 [b] |
| | IV | 10.75±0.671 | 7.892±2.867 | 7.97±2.871 | 8.12±2.856 | 8.22±2.876 |
| | V | 10.23±1.874 | 8.562±2.981 | 8.76±2.782 | 9.03±2.89 [b] | 9.14±3.961 [b] |
| RBC count (million/cu.mm) | I | 3.97±0.672 | 3.98±0.345 | 3.67±0.672 | 3.86±0.435 | 3.93±0.562 |
| | II | 3.89±0.786 | 3.48±0.672 | 3.29±0.54 | 3.01±0.673 | 2.75±0.572 |
| | III | 3.67±0.843 | 3.56±0.763 | 3.42±0.756 | 3.51±0.642 | 4.21±1.034[a] |
| | IV | 3.84±0.742 | 3.38±0.672 | 3.11±0.342 | 3.45±0.693 | 3.52±0.523 |
| | V | 3.89±0.456 | 3.45±0.531 | 3.21±0.732 | 3.67±0.627 | 3.97±0.672 [b] |
| WBC COUNT (cells/cu.mm) | I | 15.57±3.763 | 16.56±3.562 | 15.73±2.76 | 16.57±3.95 | 15.98±2.893 |
| | II | 16.73±2.865 | 20.86±3.782 | 22.67±3.782 | 27.56±4.673 | 30.45±4.923 |
| | III | 15.98±3.672 | 18.35±3.893 | 19.56±3.856 | 17.87±3.78 | 14.89±3.76[a] |
| | IV | 16.79±3.642 | 20.65±3.892 | 22.23±3.897 | 24.765±4.56 | 23.34±4.78[b] |
| | V | 16.54±3.256 | 19.67±3.892 | 20.02±3.672 | 18.74±3.82 | 16.53±3.876[a] |
| ESR (ml/hr) | I | 7.34±1.67 | 7.23±2.13 | 7.13±0.73 | 7.24±1.56 | 7.45±1.843 |
| | II | 7.23±2.89 | 7.78±1.67 | 7.98±0.56 | 8.13±1.45 | 8.87±1.782 |
| | III | 7.56±1.67 | 7.89±2.96 | 7.99±1.43 | 7.36±1.73[b] | 7.18±1.675[a] |
| | IV | 7.45±2.36 | 7.99±1.74 | 8.01±1.672 | 8.23±1.78 | 8.02±1.772 [b] |
| | V | 7.92±2.75 | 7.95±1.734 | 7.99±1.723 | 7.67±1.843[b] | 7.54±1.753[a] |

a= p<0.001, b= p<0.01, c= p<0.05 (compared with disease control group-II)

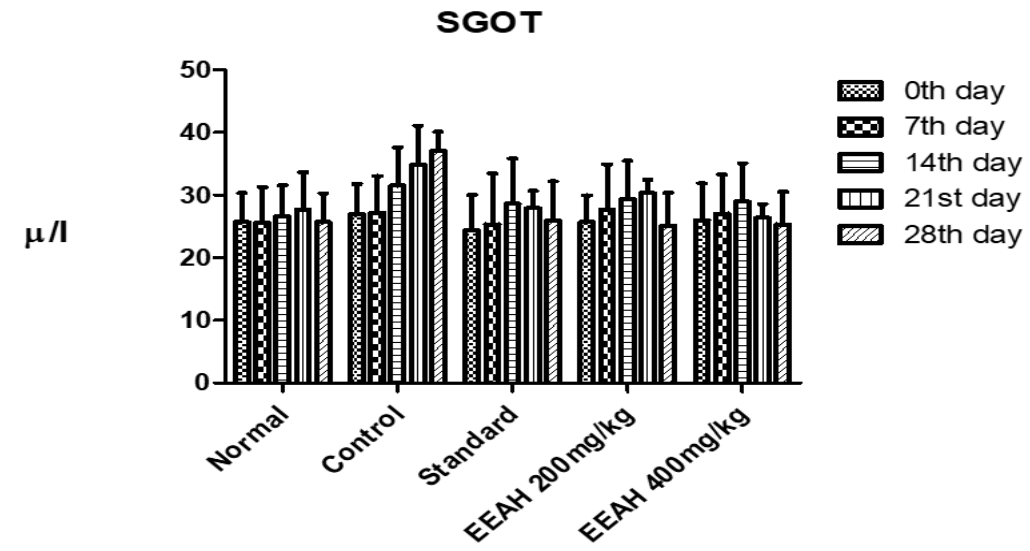
***Biochemical Parameters***

The levels of SGPT, SGOT, and alkaline phosphatase were elevated in the disease control group. However, treatment with the ethanolic extract of Arachis hypogoea resulted in a statistically significant improvement in these biochemistry parameters towards normalization by day 21.

**Table 7: Bio-chemical Parameters**

| Parameter | Group | 0 DAY | 7TH DAY | 14TH DAY | 21ST DAY | 28TH DAY |
|---|---|---|---|---|---|---|
| SGPT (IU/L) | I | 20.34±3.85 | 19.56±3.98 | 20.69±3.58 | 19.58±3.74 | 19.74±3.457 |
| | II | 19.75±3.87 | 24.21.5±3.58 | 28.26±5.23 | 29.91±5.21 | 32.7±5.23 |
| | III | 19.56±3.83 | 21.6±5.12 | 24.30±6.21 | 23.2±5.21 | 20.23±5.12[b] |
| | IV | 20.14±3.75 | 23.21±3.81 | 24.98±4.23 | 25.21±6.81 | 24.36±4.81 |
| | V | 19.64±3.12 | 22.5±2.18 | 24.2±5.62 | 21.23±6.21 | 19.31±4.21[a] |
| SGOT (IU/L) | I | 25.68±4.65 | 25.56±5.698 | 26.58±4.98 | 27.65±5.98 | 25.68±4.586 |
| | II | 26.89±4.865 | 27.1±5.91 | 31.5±6.12 | 34.8±6.28 | 37.0±3.123 |
| | III | 24.36±5.68 | 25.33±8.12 | 28.6±7.23 | 27.89±2.81 | 25.91±6.28[b] |
| | IV | 25.68±4.256 | 27.66±7.23 | 29.3±6.18 | 30.3±2.13 | 29.05±5.29 |
| | V | 25.98±5.863 | 27.04±6.21 | 28.93±6.12 | 26.35±2.21[b] | 25.31±5.23[b] |
| ALKALINE PHOSPHATASE (IU/L) | I | 140.58±8.25 | 145.68±9.65 | 143.76±7.65 | 140.86±8.26 | 145.98±9.65 |
| | II | 148.35±8.69 | 153.66±2.67 | 176.23±5.28 | 183.21±8.12 | 195.27±7.58 |
| | III | 149.25±9.56 | 152.81±9.21 | 166.28±23.0 | 158.21±7.21 | 152.11±9.21 |
| | IV | 143.48±7.34 | 176.378±21.8 | 182.21±11.21 | 173.0±11.23 | 162.96±9.32 |
| | V | 145.68±8.56 | 152.21±9.21 | 163.21±9.4 | 156.2±10.8 | 152.31±8.21 |

a=p<0.001, b= p<0.01, c=0.05 (compared with disease control group-II)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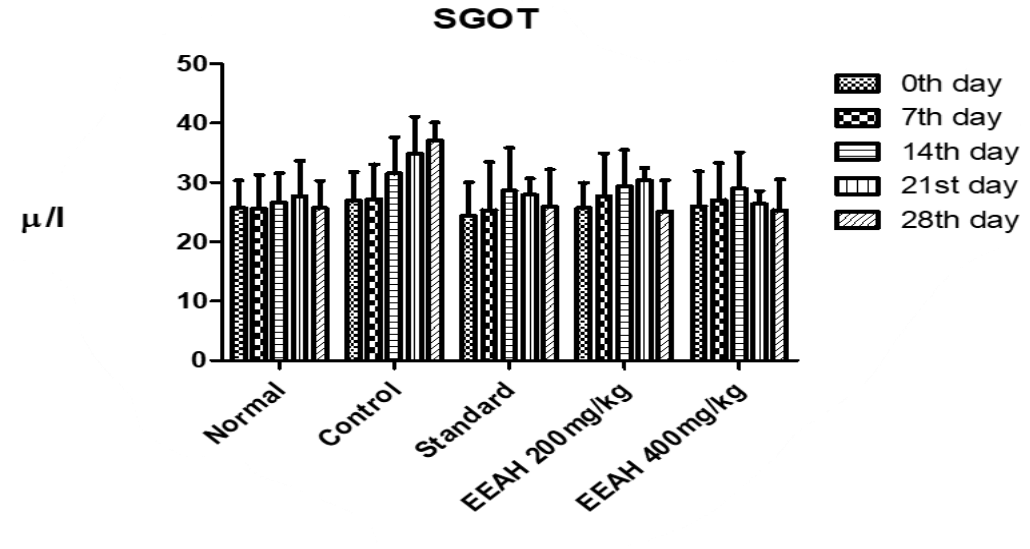
EEAH 400mg/kg
Treatment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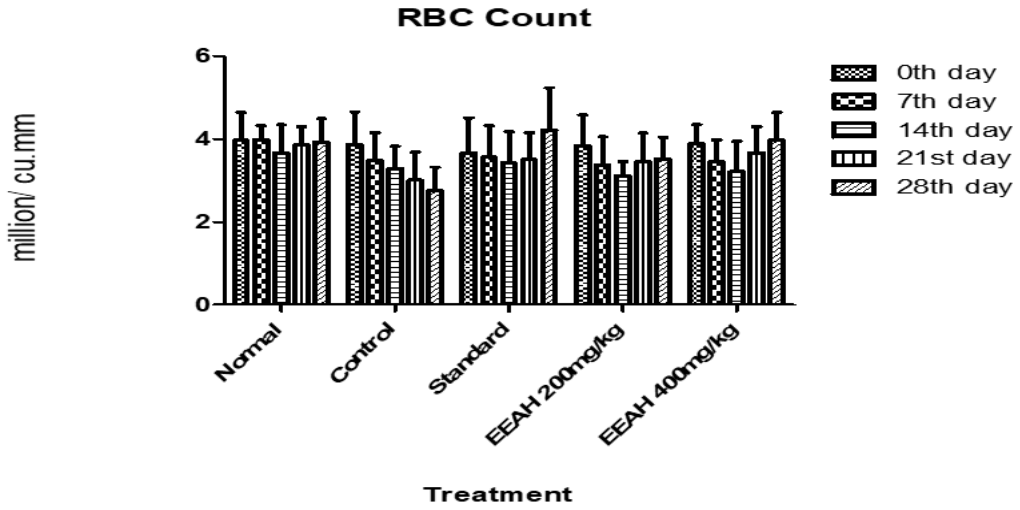
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EEAH 200mg/kg
EEAH 400mg/kg
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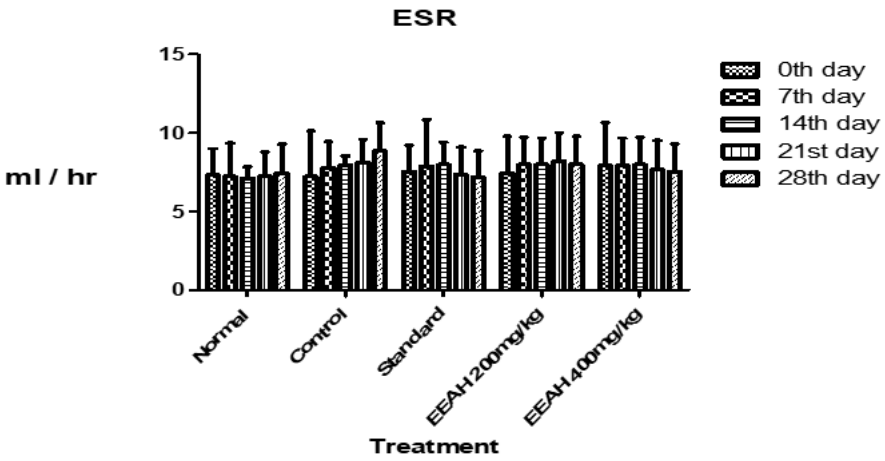
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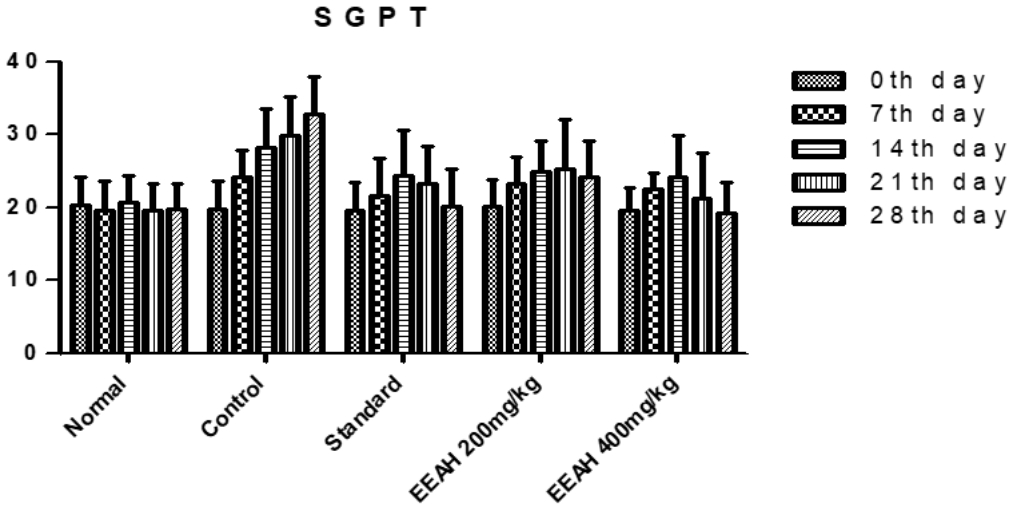
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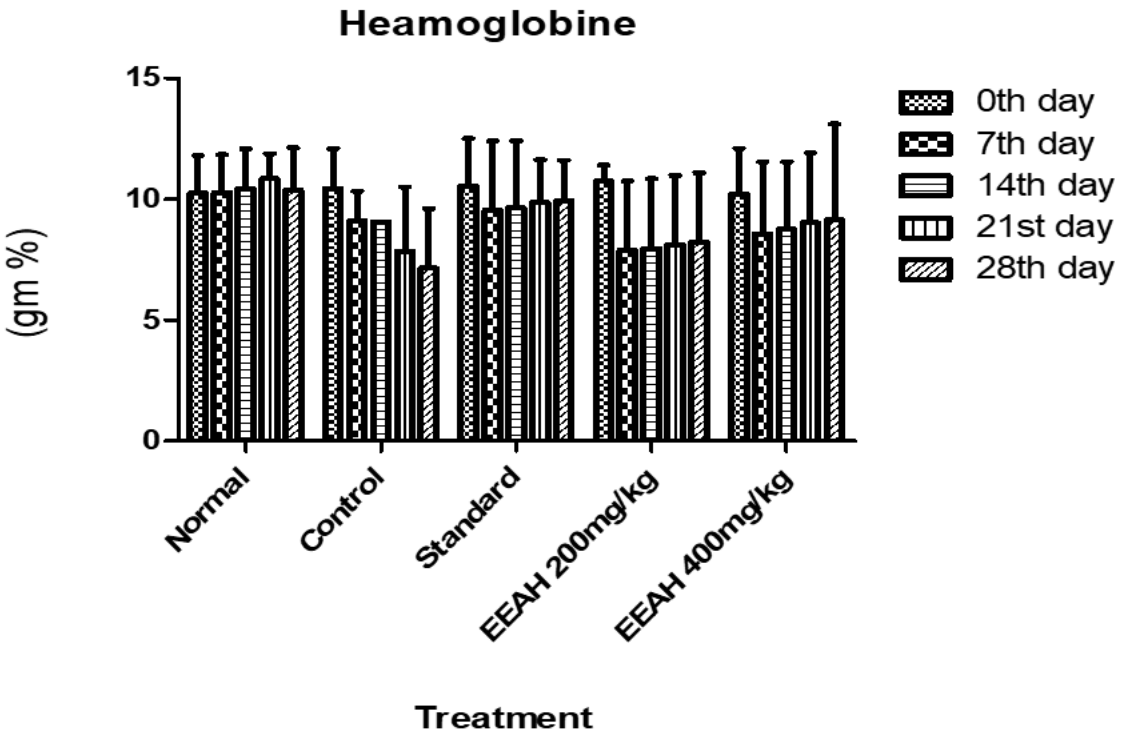
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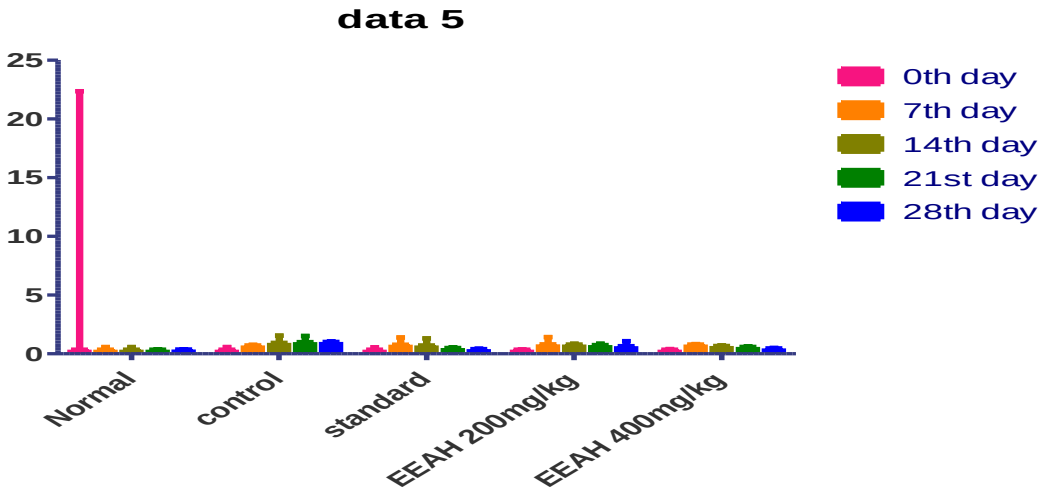
Treatment

**WBC**




***Histopathological examination***

Revealed that the control group exhibited aggregation of inflammatory cells and tissue injury. Treatment with ethanolic extract of Arachis hypogoea resulted in a decrease in inflammatory cell aggregation and minimal tissue injury compared to the control group.

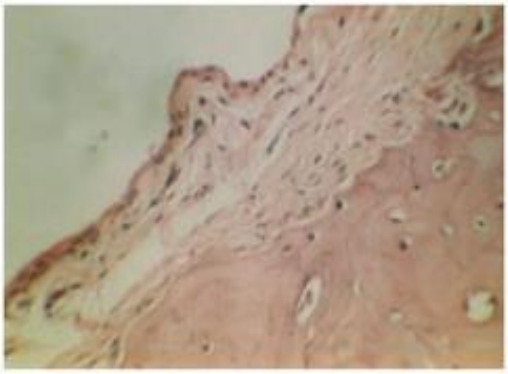


**Fig 2A : Normal**

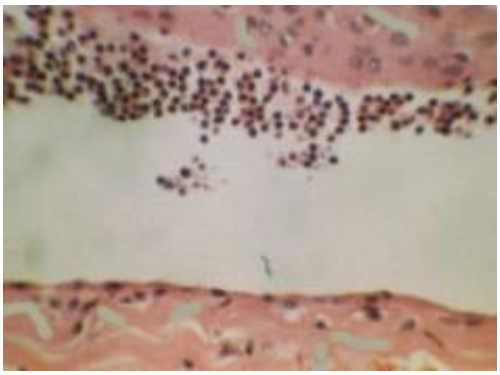


**Fig 2B : Disease control**

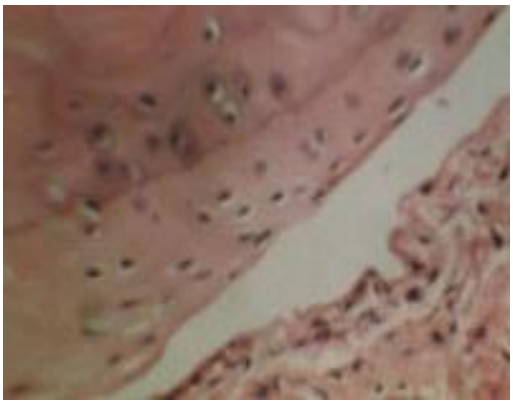
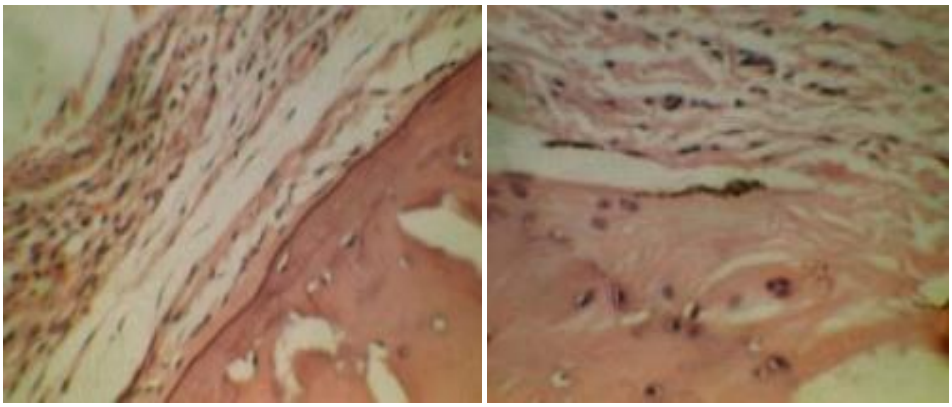


**Fig 2C : Standard (10 mg / kg)**



**EEAH;200mg/kg** **EEAH;400mg/kg**

## DISCUSSION

Arthritis is a chronic inflammatory disorder affecting a significant portion of the population worldwide. The management of arthritis involves the use of various treatment modalities, including nonsteroidal anti-inflammatory drugs (NSAIDs), disease-modifying antirheumatic drugs (DMARDs), and immunosuppressive agents. However, these treatments are often associated with adverse effects and have limited efficacy in some patients. Therefore, search for safe and effective natural alternatives is of great interest. In this study, we aimed to evaluate the potential anti-arthritis activity of the ethanolic extract of Arachis hypogoea in a rat model of arthritis induced by CFA injection. Our results provide compelling evidence regarding the beneficial effects of Arachis hypogoea extract in attenuating inflammation and edema. The successful induction of arthritis in the control group rats injected with CFA validated the model used in our experiment. The marked increase in paw edema volume in the control group compared to the normal group is consistent with previous studies using CFA injection to induce arthritis. The significant reduction in paw edema volume in the extract-treated groups indicates the potential anti-inflammatory properties of the Arachis hypogoea extract. The observed reduction in paw edema volume following treatment with the ethanolic extract at both 200 mg/kg and 400 mg/kg doses suggests the potential of Arachis hypogoea as an anti-inflammatory agent. The decrement in paw edema volume demonstrates the ability of the extract to mitigate inflammatory processes, resulting in reduced edema formation. The presence of phytochemicals such as alkaloids, glycosides, flavonoids, and tannins in the extract could potentially contribute to its anti-inflammatory activity. These phytochemicals have been reported to possess anti-inflammatory properties in various studies. The arterial hypogoea could act through the modulation of inflammatory mediators, inhibition of pro-inflammatory enzymes, and attenuation of oxidative stress. Our findings support the traditional use of Arachis hypogoea in folk medicine for treating inflammatory conditions. The dose-dependent effect observed in the Arachis hypogoea-treated groups, particularly in the 400 mg/kg dose, suggests the potential therapeutic benefit of higher dosages. The presence of phytochemicals in the extract, such as alkaloids, glycosides, and flavonoids, could be responsible for the observed anti-inflammatory effects. Alkaloids have been reported to possess anti-inflammatory and analgesic properties, while glycosides and flavonoids have shown antioxidant and anti-inflammatory activities. Tannins, on the other hand, have been associated with anti-inflammatory and immunomodulatory effects. Histopathological examination revealed a significant reduction in the aggregation of inflammatory cells and tissue injury in the extract-treated groups compared to the control group. This histological improvement further supports the anti-inflammatory activity of the Arachis hypogoea extract. The reduction in inflammatory cell aggregation indicates the potential of the extract to suppress immune-mediated inflammation associated with arthritis. The improvement in hematological parameters, including hemoglobin levels, RBC count, and WBC count, following treatment with the extract further supports its beneficial effects in restoring the altered inflammatory markers associated with arthritic conditions. The observed improvements in these parameters may be attributed to the antioxidant and immunomodulatory activities of the extract. Antioxidants present in the extract can scavenge free radicals generated during the inflammatory process, reducing oxidative

stress and preserving hemoglobin and RBC count. Additionally, the immunomodulatory effects of the extract may contribute to restoring the altered WBC count. The absence of signs of toxicity and mortality in the ethanolic extract-treated groups up to a dose of 2000 mg/kg indicates the safe profile and increased tolerability of the extract. This is an important finding as the safety of potential therapeutic agents is of utmost importance. However, it is crucial to note some limitations of our study. First, we examined the acute toxicity of the extract; hence, further investigations are required to evaluate its chronic toxicity and long-term safety profile. Moreover, our study focused on evaluating the anti-arthritis activity of the extract, thus future studies should investigate the extract's mechanism of action at a cellular and molecular level to understand its interactions with specific inflammatory pathways.

## CONCLUSION

In conclusion, the present study provides compelling evidence regarding the potential anti-arthritis activity of Arachis hypogoea extract. The observed reduction in paw edema volume, histopathological improvements, and favorable changes in hematological parameters support the traditional use of Arachis hypogoea in folk medicine for treating inflammatory conditions. These findings highlight the potential of Arachis hypogoea as a natural anti-inflammatory agent. Further investigations are warranted to elucidate the exact mechanisms of action and isolate the active compounds responsible for its therapeutic effects. This research contributes to our understanding of safe and effective natural alternatives to traditional anti-inflammatory drugs for managing arthritis.

## CONFLICT OF INTEREST

The authors declare no conflict of interest.

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