

*Original Research Article*

**A Compare Study Effects of *Ginkgo biloba* Extract and Corticosteroid in Induce Allergic Asthma in Rabbits in Regards to Physiological and Immunological Parameters**

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**Abstract**

Thirty adult male of New Zealand Albino white rabbits were used in this article and these rabbits are haphazardly divided into five different experimental groups. Ovalbumin was used in a dose (0.1 mg /kg) to induce allergy and sensitization challenge was confirmed by the presence of the clinical symptoms of wheezing, sneezing, chest tightness, shortness of breath, changes in the level of total white blood cell, neutrophils, eosinophils, Immunoglobulin E and Interleukin-4. Eight days after allergy induction, *Ginkgo biloba* giving in the dose (25 mg/kg) and in a combination with prednisolone in the half of recommended dose (12.5 + 0.5) mg/kg (*Ginkgo biloba* + prednisolone) was giving orally for 8 days.

The results showed that ovalbumin could induce sensitivity and significantly increased the level of total white blood cell, neutrophils, eosinophils, Immunoglobulin E and Interleukin-4. Oral treatment with *Ginkgo biloba* in the dose (25 mg/kg) and in a combination with prednisolone as the half of recommended dose (12.5 + 0.5) mg /kg of *Ginkgo biloba* + prednisolone showed high significant decrease ( $p \leq 0.01$ ) in the number of total white blood cell, neutrophils and eosinophils and serum levels of Immunoglobulin E and Interleukin-4 levels. The effects of this plant extract were comparable with the observed effect of prednisolone-treated group to be present with high efficacy of herbal medical therapy than that of prednisolone treatment a combined with adverse effects when use alone.

**Key Words:** *Ginkgo biloba*; flavonoids; neutrophils; eosinophils; Immunoglobulin E; Interleukin-4; asthmarabbit's model.

**الخلاصة**

تم استخدام ثلاثين من ذكور الأرانب البيضاء النيوزيلندية في هذه الدراسة، وقسمت هذه الأرانب عشوائياً إلى خمس مجموعات تجريبية مختلفة. تم استخدام زلال البيض في جرعة (0.1 ملغ / كلغ) للحث على الحساسية وتأكد الحث من خلال وجود الأعراض السريرية للعدس، والتنفس، وضيق في التنفس، وضيق الصدر، والتغيرات في مستوى المعدلات والغلوبيولين المناعي E، انترلوكين -4. بعد ثمانية أيام من الحث التحسسي، كان الجنكبيلوبا يعطي بجرعة (25 ملغم / كلغم) وفي توليفة مع برينديزولون في نصف الجرعة الموصى بها (12.5 + 0.5) ملغم / كلغم (الجنكبيلوبا + برينديزولون) يعطي فموياً لمدة ثمانية أيام.

وأظهرت النتائج أن زلال البيض يمكن أن يسبب ربو تحسسي وزيادة كبيرة في مستوى العدلات والغلوبيولين المناعي E، انترلوكين -4. وأظهرت المعاملة بالفم لكل من الجنكبيلوبا بجرعة (25 ملغم / كلغم) وفي توليفة مع برينديزولون نصف الجرعة الموصى بها (12.5 + 0.5) ملغم / كلغم من الجنكبيلوبا + برينديزولون انخفاضاً معنوياً كبيراً ( $p \leq 0.01$ ) في عدد خلايا العدلة ومستويات المصل من الغلوبيولين المناعي E، مستويات إنترلوكين -4 وعودة درجة حرارة الجسم إلى الحالة الطبيعية. وكانت تأثيرات هذا المستخلص النباتي قابلة للمقارنة مع تأثير لوحظ في المجموعة المعالجة بعقار

البريدنيزولون (كيميائي المنشأ) ليظهر وجود فعالية عالية للعلاج الطبي العشبي مع العلاج بالبريدنيزولون جنب إلى جنب مع تقليل و تلافى التفاعلات الجانبية والآثار السلبية عند استخدام البريدنيزولون لوحدة.

**الكلمات المفتاحية:** الجذكة، الفلافينات، العدالة، الحمضة، الغلوبولين المناعي E، أنترلوكين -4، موديل الربو الارانب.

## **Introduction**

**A**sthma stays a chronic inflammatory allergic disease of the air ways that's related to raised responsiveness of the airways to environmental stimuli and reversible airflow obstruction. It's some results of the interaction between genes and atmosphere, and a number of additional genetic and environmental factors [1].

Like any infect or irritant, the respiratory tract had been occurred and deteriorate the respiratory physiological function as a result of inflammation of the airpassageway in the lung and affects the sensitivity and also the nerve end within the airway that is stimulant throughout attack wherever the liner passage is swell and inflicting narrowing airway passage falling the air flow out of the lung that is in final come with difficult of breath [2].

A major goal of medical analysis is to outline the cause and develop the cure for chronic disease, like mucus hypersecretion, smooth muscle hyperplasia, sub-epithelial fibrosis, blood vessel proliferation, and infiltration of inflammatory cells [3]. Conventionally by targeting the adaptative immune system. Convention has additionally lead to a bipartite classification of the adaptative immune system, whereby Th1 cells mediate delayed-type hypersensitivity reactions and by selection produce IFN- $\gamma$  and IL-2, and Th2 cells promote B cell-dependent humoral immunity and produce IL-5, IL-4, and IL-13 [4]. Within the situation of asthma attack, the "Th2 hypothesis" proposes that an upregulated Th2 and a downregulated Th1 response drive the growth of disease [5]. Herbal medication (or "herbalism") is that the study and use of medicative properties of plants [6]. *G. biloba* leaves are a

conventional Chinese medication, that have survived over one hundred eighty million years, and are thought-about to be "living fossils" [7]. *G. biloba* leaves are wealthy in several biologically active compounds like flavonoids, carboxylic acids, alkylphenols, polyphenols, terpene lactones and soon [8]. *Ginkgo biloba* used for the cure of pulmonary disorders like (cough and asthma) and bladder inflammation. Also, their leaves are used to treat heart and lungs abnormalities and also for the treatment of skin disorder [9].

## **Materials and Methods**

**Animals:** Thirty adult male rabbits (breed: New Zealand white, *Oryctolagus-cuniculus*) weighing: (2100-2700) grams and 12-16 months old were used in the present search. Animals were left 4 weeks for an adaptation prior to the experiments. Each 6 animals were housed in optimized steel less steel cages and they were had been free feeding on freshly green vegetables and chaw pellets and access amount of water. Animals were reserved under the similar conditions of the temperature (20-25) °C And light automated program of 12 hrs.' light, and 12 hrs.' dark. Animals of study were separated into 5 groups each group contain of 6 male rabbits used with designed experimental protocol.

### **Experimental Protocol:**

The protocols of experimentare modified program adapted from [40,2,41]. Induction of allergic asthma was performed by intraperitoneal (I.P.) injection (immunization) by Ovalbumin (OVA) 0.1 mg and 10 mg of aluminum hydroxide in 2 ml of phosphate buffer saline (PBS) in day 1 and repeated with same preparation and concentration as a challenge dose through second sensitization in day 14 of the

immunization program the animals divided for 5 groups. As in the followings:

**Group 1 (Control group):** consist of 6 rabbits giving twice (I.P.) injection of only phosphate buffer saline at the day 1 and 14 of the experiment.

**Group 2 (OVA group):** consist of 6 rabbits giving twice (I.P.) injection of OVA 0.1 mg and 10 mg of Al(OH)<sub>3</sub>(aluminum hydroxide) in 2 ml of phosphate buffer saline by (I.P.) injection(immunization) in day one of the experiment and booster dose in day fourteen, and then all animals were euthanized in day 15 of the experiment.

**Group 3 (Prednisolone group):** consist of 6 rabbits giving twice (I.P.) injection of OVA 0.1 mg and 10 mg of Al (OH)<sub>3</sub> (aluminum hydroxide) in 2 ml of phosphate buffer saline by (I.P.) injection (immunization) in day one of the experiment and booster dose in day fourteen, and treated giving orally from the day 23 to 30 with 1mg/kg of prednisolone, then all animals were euthanized in day 31 of the experiment.

**Group 4 (Ginkgo Biloba group):** consist of 6 rabbits giving twice (I.P.) injection of OVA 0.1 mg and 10 mg of Al(OH)<sub>3</sub> (aluminum hydroxide) in 2 ml of phosphate buffer saline by (I.P.) injection (immunization) in day one of the experiment and booster dose in day fourteen, and treated giving orally from the day 23 to 30 orally with 25 mg/kg, and then all animals were euthanized in day 31 of the experiment.

**Group 5 (G. Biloba plus Prednisolone group):** consist of 6 rabbits giving twice (I.P.) injection of OVA 0.1 mg and 10 mg of Al(OH)<sub>3</sub> (aluminum hydroxide) in 2 ml of phosphate buffer saline by (I.P.) injection (immunization) in day one of the experiment and booster dose in day fourteen, and treated giving orally from the day 23 to 30 with G. Biloba extract 12.5 mg/kg plus prednisolone 0.5 mg/kg, and then all animals were euthanized in day 31 of the experiment.

Blood samples were collected from all control and treated groups in the day 15 of the experiment for evaluation the effects of

the OVA, also it collected in day 31 after 1 day from the last treatment dose.

#### **Physiological & immunological parameters:**

Total WBC & eosinophil count: After collection of the samples and taken blood into a test tube covered within decoagulant (EDTA) to avoidance clotting. The samples were then taken to the laboratory for counting the cells of the total WBCs, the neutrophils and eosinophils measurements through drawing the blood that performed manually, and prepared slide of blood sample for each of experimental animals involvement as a blood film and examine under the microscopical magnifications powers [10].

**Immunoglobulin E estimation:** Estimation of Rabbit Immunoglobulin E concentrations was carried out according to the kit instructions of company (Total IgE-German) by using ELISA technique.

**Cytokine Levels:** the concentrations of cytokine Interleukin -4 in the serum were calculated by sandwich ELISA using a commercial recommended available reagent in a step of the manufacturer's direction.

**Statistical Analysis:** Statistical analysis was made abuse applied mathematics Statistical Package for the Social Sciences(SPSS, version 22). data was stated by One Way ANOVA test and mean  $\pm$  SD for continuous variable. The p-value when present less than (0.05) was statistically significant, and the p-value when present less than (0.001) was statistically highly significant [11].

#### **Results**

##### **Effect of prednisolone on total WBCs for control, asthma induced & treated rabbits**

Observable data of the table in spite of showing an obvious increased in total WBCs in asthma induce rabbits when compare with the control group. But, data also presence intense significant increasing ( $P \leq 0.05$ ) in the total WBCs estimation as shown in table (1) ( $11.880 \pm 1.045$ )  $\times 10^3$  cells/ $\mu$ L when those asthmas induced rabbits receiving prednisolone (treated) daily in a dosing 1

mg/kg of the B.W. along that period of experiment.

According to the schematic protocol that lasted for 8 days start in the day 23 and end in the day 30 of the experiment.

**Effects of *Ginkgo biloba* plant extract on total white blood cells estimate in asthma induced rabbits.**

The result in figure (1) of asthma induced rabbits which treated with 25 mg/kg *Ginkgo biloba* and 12.5 mg/kg *Ginkgo biloba* + 0.5 mg/kg of prednisolone.

The results have shown a significant decrease ( $P \leq 0.05$ ) in the total WBCs estimation ( $5.66 \pm 2.504$ )  $\times 10^3$  cells/ $\mu$ L with *Ginkgo biloba* extract (25 mg/kg) of body weight. Also, that the other group of 12.5 mg/kg *Ginkgo biloba* + 0.5 mg/kg of prednisolone were showed a significant decrease ( $P \leq 0.05$ ) on total WBCs estimation ( $6.86 \pm 2.992$ )  $\times 10^3$  cells/ $\mu$ L respectively in comparison with healthy control levels of the total WBCs estimation ( $4.96 \pm 1.07$ )  $\times 10^3$  cells/ $\mu$ L in the animals, of the designated study.

Groups	Total WBC estimation $\times 10^3$ cells/ $\mu$ L (Mean $\pm$ SD)
Control Group	4960 $\pm$ 1290
Asthma Rabbits (ovalbumin induced)	5800 $\pm$ 1070
Asthma Rabbits treated with 1 mg/kg of prednisolone	11880 $\pm$ 1045

SD: Stander deviation; WBCs: White blood cells

Table (1) Total WBCs estimation (Mean  $\pm$  SD) for each of control, asthma induced and treated rabbits.

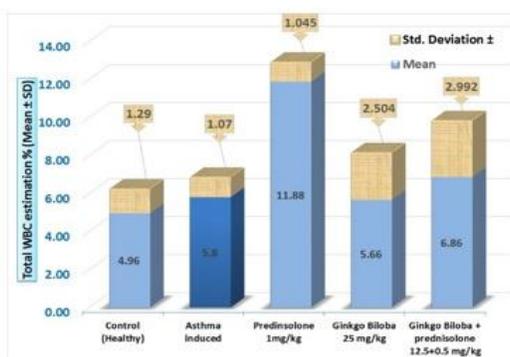


Figure (1) A comparison effect of prednisolone, *Ginkgo biloba*, and *Ginkgo biloba* + prednisolone on the Total WBCs estimation (Mean  $\pm$  SD) in asthma induced and control rabbits.

**Prednisolone effect on neutrophils approximation for each of control, asthma induced rabbits and treated rabbits.**

The immunogen was the sensitizer ovalbumin (antigen), and the immunization protocol was i/p injection program, mostly is associated with simple elevation of neutrophils in asthma induced rabbits ( $53.6 \pm 4.827$ )% in compared with neutrophils of control (healthy) rabbits ( $51 \pm 4.848$ )% as shown a table (2). Also, result pointed that regardless receiving of prednisolone for 8 days after the immunization program with ovalbumin during the experiment. Evidence show that neutrophils estimation was reduced. But, was remain significantly increase ( $P \leq 0.05$ ) ( $67.8 \pm 1.634$ )% in compared with control (healthy) rabbits ( $51 \pm 4.848$ )% as shoe in table (2).

**Effects of *Ginkgo biloba* extract plant on neutrophils estimation in asthma induced rabbits.**

Rabbits treated with 25 mg/kg have showed significant decrease ( $P \leq 0.05$ ) in neutrophils estimation in asthma induced rabbits afterward receiving a dosage of 25 mg/kg *Ginkgo biloba* extract ( $50.2 \pm 6.979$ )% which begin when asthma is induced and persevere for eight days after the immunization protocol of the experiment compared with control ( $51 \pm 4.848$ )% (healthy) rabbits as showed in figure (2).

The figure (2) show also, a significant decrease ( $P \leq 0.05$ ) in neutrophils measurements through the treatment with the 12.5 mg/kg *Ginkgo biloba* + 0.5 mg/kg of prednisolone ( $47 \pm 1.581$ )% respectively in comparison with control (healthy) rabbits.

Groups	Neutrophils Estimation (Mean $\pm$ SD) %
Control Group	51 $\pm$ 4.848
Asthma Rabbits (ovalbumin induced)	53.6 $\pm$ 4.827
Asthma Rabbits receiving 1 mg/kg B.W of prednisolone	67.8 $\pm$ 1.643

Table (2) Neutrophils estimation (Mean  $\pm$  SD) for each of control, asthma induced and treated rabbits.

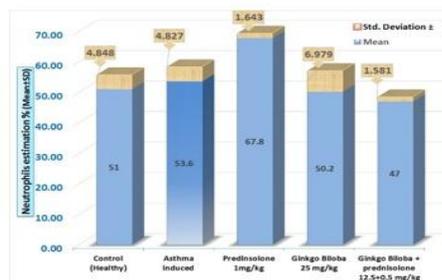


Figure (2). A comparison effect of prednisolone, *Ginkgo biloba*, and *Ginkgo biloba* + prednisolone on the neutrophils estimation (Mean  $\pm$  SD) in asthma induced and control rabbits.

### Effect of prednisolone on eosinophils measurement for each of control, asthma induced, and treated rabbits.

The eosinophils were being measured after the use of the same standard protocol for induce asthma was of i/p injection program immune gene (ovalbumin) sensitize. The result in the table (3) was shown a significant increase ( $P \leq 0.05$ ) in eosinophils measurement of asthma induced animals ( $6.2 \pm 0.837$ ) % when eosinophils measurement comprise with control (healthy) rabbits ( $0.6 \pm 0.548$ )% as it was shown in below table.

In addition, that table (3) illustrate a significant decrease in eosinophils estimation ( $P \leq 0.05$ ) ( $1.6 \pm 0.894$ )% when asthma induce rabbits gave prednisolone along for eight days started in day 23 and end in day 30 of the experiment.

### Effects of *Ginkgo biloba* plant extract on Eosinophils estimation in asthma induced rabbits.

*Ginkgo biloba* extraction as in figure (3) showed that significantly decrease ( $P \leq 0.05$ ) in eosinophils estimation of asthmatic rabbits afterward receiving a dosage of 25 mg/kg B.W. of *Ginkgo biloba* extract ( $1.4 \pm 0.548$ )% which begin after showed of asthma and continue for eight days later the asthma induce protocol of the experiment in compared with control ( $0.6 \pm 0.548$ )% (healthy) rabbits.

Meantime, figure (3) show also, significantly decrease ( $P \leq 0.05$ ) in eosinophils measurements through the treatment with the 12.5 mg/kg *Ginkgo biloba* + 0.5 mg/kg of prednisolone ( $1.4 \pm 0.548$ )% respectively in compared with control ( $0.6 \pm 0.548$ ) % healthy rabbits.

Groups	Eosinophils % estimation (Mean ± SD)
Control Group	0.6 ± 0.548
Asthma Rabbits (ovalbumin induced)	6.2 ± 0.837
Asthma Rabbits treated 1 mg/kg B.W of prednisolone	1.6 ± 0.894

Table (3) Eosinophils estimation (Mean ± SD) for each of control, asthma induced and treated rabbits.

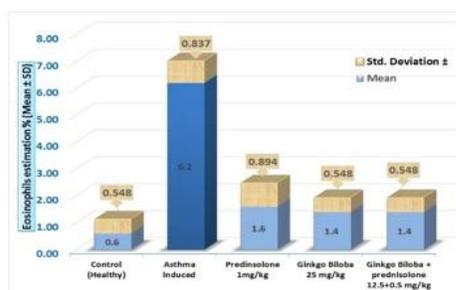


Figure (3) A comparison effect prednisolone, *Ginkgo biloba*, and *Ginkgo biloba* + prednisolone on the Eosinophils estimation (Mean ± SD) in asthma induced, and control rabbits.

### Prednisolone receiving effect on Immunoglobulin E level in asthma induced rabbits.

Result present in table and figure (4) show a significant increase ( $P \leq 0.05$ ) in the immunoglobulin E level as a reflect of ovalbumin induced asthma in rabbits ( $381.52 \pm 4.177$ )  $\mu\text{g/ml}$  compared with the healthy (control) rabbits ( $48.26 \pm 4.47$ )  $\mu\text{g/ml}$ . Meanwhile, the table show significantly decrease ( $P \leq 0.05$ ) in the levels of immunoglobulin E ( $53.49 \pm 3.91$ )  $\mu\text{g/ml}$  on asthma rabbits when received (treated) with prednisolone at a dose of 1 mg/kg of body weight which last for eight days started from the day 23 and end in day 30 of the experiment.

### Effects of *Ginkgo biloba* plant extract on Immunoglobulin E level in asthma induced rabbits.

A valuable obtained data in the table (4) showed asthma induced rabbits when treated with *Ginkgo biloba* plant extract of 25 mg/kg body of weight. Data showed a decrease in the level of Immunoglobulin E ( $67.21 \pm 4.14$ )  $\mu\text{g/ml}$ . While, the treatment with 12.5 mg/kg *Ginkgo biloba* + 0.5 mg/kg of prednisolone were reveal significant decrease ( $P \leq 0.05$ ) on the level of Immunoglobulin E ( $53.09 \pm 5.57$ )  $\mu\text{g/ml}$  respectively in comparison with healthy control levels of the immunoglobulin E ( $48.26 \pm 4.47$ )  $\mu\text{g/ml}$  in the animals, of designated study.

Groups	IgE levels $\mu\text{g/ml}$ (Mean $\pm$ SD)
Control Group	48.26 $\pm$ 4.478
Asthma induced (Ovalbumin received)	381.52 $\pm$ 4.177
Asthma Rabbits treated 1 mg/kg B.W of prednisolone	53.49 $\pm$ 3.914

SD: Stander deviation; **IgE**: Immunoglobulin E

**Table (4)** Immunoglobulin E levels (Mean  $\pm$  SD) in each of control, asthma induced and treated rabbits.

**Prednisolone receiving effect on the levels of IL-4 in each of control and asthma induced rabbits.**

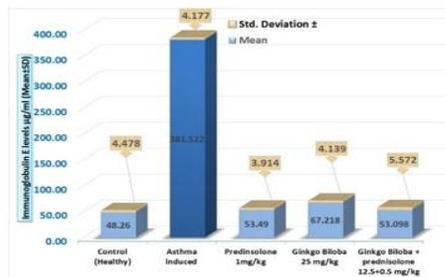
The present result in the table (5) show that Interleukin-4 levels were increased significantly ( $P \leq 0.05$ ) in asthma induced rabbits with ovalbumin injection ( $358.87 \pm 60.08$ ) pg/ml compared with healthy rabbits ( $30.44 \pm 4.577$ ) pg/ml.

While, the table show a significant decrease ( $P \leq 0.05$ ) in the IL-4levels ( $27.64 \pm 2.78$ ) pg/ml when those asthma induced rabbits treated with prednisolone at a daily dose of 1 mg/kg of B.W.at that time of same period. According to the schematic protocol that

Groups	IL4 levels $\text{pg/mL}$ (Mean $\pm$ SD)
Control Group	30.44 $\pm$ 4.577
Asthma induced (Ovalbumin received)	358.87 $\pm$ 60.085
Asthma Rabbits treated 1 mg/kg B.W of prednisolone	27.64 $\pm$ 2.789

SD: Stander deviation; **IL 4**: Interleukin 4

**Table (5)** Interleukin-4 (Mean  $\pm$  SD) levels in each of control, asthma induced and treated rabbits.

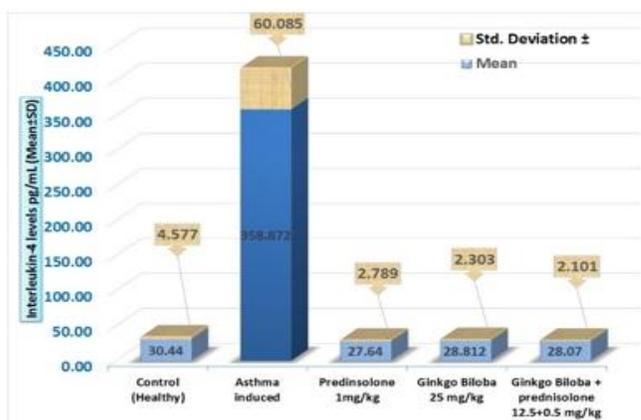


**Figure (4)** A comparison effect prednisolone, *Ginkgo biloba*, and *Ginkgo biloba* + prednisolone on the levels of Immunoglobulin E levels (Mean  $\pm$  SD) in asthma induced and control rabbits.

lasted for eight days started in day 23 and end in day 30 of the experiment.

**Effects of *Ginkgo biloba* plant extract on the Interleukin-4 level in asthma induced rabbits.**

After receiving treatment with a *Ginkgo biloba* plant extract of 25 mg/kg body weight for eight days a significant decrease ( $P \leq 0.05$ ) was showed in the levels of IL-4 ( $28.81 \pm 2.30$ ) pg/ml, also after treatment with the(12.5 mg/kg *Ginkgo biloba* + 0.5 mg/kg prednisolone)for eight days was shown a significant decrease ( $P \leq 0.05$ ) in the levels of IL-4 ( $28.07 \pm 2.10$ ) pg/ml as show in Figure (5).



**Figure (5)** A comparison effect of prednisolone *Ginkgo biloba*, and *Ginkgo biloba* + prednisolone on the levels of Interleukin-4 (Mean  $\pm$  SD) in asthma induced and control rabbits.

## **Discussion**

The outcomes of data of figures (1), (2) and (3) were showing significant increase in total WBCs, eosinophils and neutrophils measurements were in a good agreement with other scientists [2,12] whom, pointed significant elevation with these two divergent types of hematological parameters and extend their role in expression of asthma symptoms by the release of inflammatory mediators of chemokines and cytokines that reproduce the use of ovalbumin to finally show a major role in distinctive allergic asthma signs. However, it also showed a significant reduction in eosinophils that measured donate to the flavonoids of *Ginkgo biloba* inhibitory effect, in the current study.

The enlisting of eosinophils into bronchial cartilaginous tube mucous membrane during which allergic inflammation happens could be an essential donor to the late asthma reaction of mucus hypersecretion and congestion [13,14]. once the eosinophils cells reach degranulation occur and carry on inflammation of underlying airway. These cells are a rich supply of lipid mediators, cytotoxic proteins, free radicals, cytokines, and  $O_2$  [13].

In patients with asthma, once trans epithelial tissue migration, eosinophils transmigrate and cling to bronchial cartilaginous tube epithelial tissue wherever eosinophils degranulate and unharness substances that are cytotoxic for epithelial cells (major basic protein, eosinophil ion macromolecule, superoxide, and WBC oxidase) [15]. Organic phenomenon and Injury of cells desquamation, tissue epithelial secretion and cilliostasis [16] manifest the airway epithelial tissue toxicity. Major basic protein could be a selective, allosteric adversary for muscarinic receptors ( $M_2$ ) [17].

The damage of muscarinic receptor ( $M_2$ ) perform leads to enhanced airway tone attributable to enhanced unharness of neurotransmitter and synergism of vagally mediate reflex bronchoconstriction and

bronchial cartilaginous tube hyperresponsiveness [18]. MBP additionally stimulates production of histamine from mast cells and basophils. lipid bodies are evoked to be advanced within eosinophils that activated, and are the positions for increased formation of each cyclooxygenase-derived eicosanoids and lipoxygenase [19].

Eosinophils are capable of manufacturing large amounts of leukotrienes (specially IL-4). leukotrienes bond the smooth muscle of airway (100-1000) fold firmer bronco constrictors than histamine, rise vasculature permeability, enhance secretion of mucus, reduction mucousciliary clearance, enhance neutrophils and eosinophils into the airways enlisting, cause neural dysfunction and enhance proliferation of airway smooth muscle [20].

Eosinophils have a powerful to release and synthesize variety of chemokines and cytokines. Cytokines created by eosinophils included the autocrine-eosinophil active growth factors (Interleukin-3, Interleukin-5, GM-CSF), cytokines that immunoregulatory include (Interleukin-1, Interleukin-2, Interleukin-4,  $IFN-\gamma$ ,  $TGF-\beta$ ), pro-inflammatory cytokines include (Interleukin-6, Interleukin-1, Interleukin-16, Tumor necrosis factor- $\alpha$ ) and chemokines where include of Interleukin -8 and  $MIP-1\alpha$  [21].

After reaching of eosinophils at the airways inflammatory site, eosinophils become stimulated with phenotype (hypodense) [22]. Stimulated eosinophil expression a various of receptors for immunoglobulins, chemokines, cytokines and complement. throughout the response to inflammation, domestically created complement-derived anaphylatoxins  $C5a$  and  $C3a$  bounded to specific receptors on the cells and invigorate respirational spurt in eosinophils.  $C5a$  performances as a chemoattractant for eosinophils and neutrophils and signifies a large eosinophils metabolic activator causing to free a tinny granule of protein and

free O<sub>2</sub> radicals that reason destruct and injury for tissue [23].

(1), (2) and (3) figures showed the effect of the (chemical treatment) prednisolone one of corticosteroids collection and had overcome the parallel action on hematological parameters.

The WBCs count could be a routine lab. check which mirrors the score of distribution of leukocytes within blood [24]. Neutrophils reside in some compartments; the 2 compartments associated with this subject are the bordering section (hooking up of neutrophils to the blood vessel endothelium) and therefore the circulating cells (cells circulate within the blood vessels together with different cells) [24].

This difference is very significant as neutrophils inside the vessel lumen roving on the endothelial surface (i.e., within the bordering compartment) aren't mirrored during a white cell count. solely the polymorphonuclear circulating freely in the circulatory section are found within the blood vessel sample that used for examination. as like anything that lead the marginal neutrophils to separate from the vessel wall epithelium surface can lead to a larger absorption of neutrophils within the circulatory compartment and therefore rise the white cell count. This is notable to do by glucocorticoids [25]. as the WBCs harvested in our work.

Corticosteroid side effect on a hematological parameter such as elevated RBCs and Hb content of blood, probably by delaying erythroid- phagocytosis. This impact was incontestable via the incidence of blood disorder (polycythemia) in Cushing syndrome and in Addison disease gentle normochromic anemia. Corticosteroids additionally have an effect on circulating white cells. adrenal cortical steroid treatment leads to enhanced polymorphonuclear WBCs in blood as a consequence of increment rate of the doorway from marrow and a reduced rate of elimination from the vasculature compartment. In distinction, the

eosinophils, basophils, monocytes, and lymphocytes reduction in number after glucocorticoids administration [26].

A solitary dose of corticosteroid ends up in a seventieth percentage reduction in lymphocytes and a ninetieth percentage reduction in monocytes, occurring four to six hrs. after treatment and continuous for approximately twenty-four hrs. Cell numbers then rise twenty-four to seventy-two hrs. after treatment.

The reduction in eosinophils, lymphocytes and monocytes is believed to be a significance of the distribution of those cells, while sure lymphocytes additionally tolerate glucocorticoid-induced cell death. T lymphocytes have less sensitivity to glucocorticoid-induced cell death than are B lymphocytes, and T-cell subpopulations dissent in their glucocorticoid sensitivity. A reduction in basophils happens by an unidentified mechanism.

But, the extended intake corticoid treatment results in rise in neutrophils and reduce in eosinophils, basophils and lymph cell, that such results were in matched with existing obtained subject, additionally to concomitant immunological disorder, additionally leaves patients at risk of invasive diseases and fungous infections [27].

In the present study, had been shown that the *Ginkgo biloba* extract and its main constitute from flavonoid can modulate the appearance of pro-inflammatory cytokines and their role in the severe phase of the inflammatory response.

The obtained data from figure (4) and (5) showed the inhibitory impact of *G. biloba* extract on IgE and IL-4 levels in treated groups with the plant extract, and this effect contributes to the concentration of flavonoid.

Degranulation unharness apart from histamine; mast cells show variance roles within the allergic inflammation by starting and arranging immune responses via the discharge of chemokines and

cytokines via difference intracellular communication paths [28].

PMACI and immunoglobulin-E are best-known to facilitate the expression of cytokines and also the histamine discharging from mast cells [29]. The HMC-1 cells activation by PMACI raised levels of proinflammatory protein [30].

Though, immature human mastocyte line (HMC-1), wasn't impressed in unharness or content of histamine by immunoglobulin E or PMACI. The intracellular Ca is the important efficient compartments within the mast cells degranulation. Ca crossing the mast cells membranes represent a significant mark for antiallergic medication effectiveness [31].

Together

cells varieties are necessary players in the allergic inflammatory responses regulation by feature of their capabilities to proinflammatory mediators providing as well as histamine [28]. Proinflammatory cytokines like interleukin-1 $\beta$ , interleukin -6, interleukin -8 and TNF- $\alpha$  and aid inflammation, infiltration of white blood cell, formation of granuloma and tissue fibrosis, and are thought to be leader of inflammation states connected to cytokine by motivating production of cytokine [32].

Flavonoids is stated to impede immunoglobulin E-mediated histamine unharness and production of TNF- $\alpha$  from mast cells derived cultivated from bone marrow and mast cells serosa [33]. Flavonoids inhibit immune globulin mediate unharness of proinflammatory mediators from umbilical cord blood-derived cultivated mast cells by inhibition of PKC signaling and Cainflux [34]. Moreover, it had been investigated that the impact of flavonoids on the RBL-2H3 cells and granule-stored mediator accumulation in HMC-1 proliferation [35].

Subsequent antigenic challenge in immunoglobulin E-sensitized mast cells, flavonoids decrease level of ROS and deeply suppressed release of histamine [36]. However, flavonoids markedly decrease

pro-inflammatory cytokines through defeat of MAPKs and NF- $\kappa$ B [30].

Corticosteroids were supposed of from the foremost active treatment accessible for the allergic diseases management, as well as dermatitis, allergic rhinitis and asthma attack. Their effects are helpful primarily mediated by multiple mechanisms of anti-inflammation [37].

Especially, the repression of the many inflammatory cytokines and chemokines that amplify and preserve allergic inflammation. Suppression of inflammatory proteins depends on the interaction of glucocorticoid receptors, stimulated by binding to corticosteroids, with varied transcript factors that are stimulated by induce inflammation [38].

Also, corticosteroids decline the endurance of eosinophils and T cells by growing programmed cell death, contributive to their defeat of chronic allergic inflammation. Additional apparently damaging impact of corticosteroids involves the interleukin-4 stimulated creation of immunoglobulin E that's seen in hydrocortisone treated B lymphocytes [41].

### **Conclusion**

Present experimental study emerges with slight or no adverse effects for *Ginkgo biloba* plant extract treatment compared with standard asthma attack synthetic preventive treatments, and regarding the results of this study, *Ginkgo biloba* deserves within observance long-term plans for such patients with asthma attack or allergic reaction, But, with taking in to account a number of individual reservation.

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