IJPSR (2014), Volume 5, Issue 10 **(Research Article)**

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 05 March 2014; received in revised form, 23 May 2014; accepted, 13 July 2014; published 01 October 2014

EVALUATION OF IMMUNOMODULATORY ACTIVITY OF THE FLAVANOID OF *KIGELIA AFRICANA*

P. K. M. Nagarathna, K. Reena *, Sriram Reddy and Johnson Wesley

Department of Pharmacology, Karnataka College of Pharmacy, Bangalore - 560054, Karnataka, India.

Keywords:

Kigelia africana, Humoral immunity, Serum immunoglobulin, Phagocytic index, Cell-mediated immunity

Correspondence to Author:
K. Reena

Department of Pharmacology, Karnataka College of Pharmacy, Bangalore - 560054, Karnataka, India.

E-mail: k.reena05@gmail.com

ABSTRACT: Objective: Modulation of the immune responses to alleviate the diseases has been of interest for many years. Thus, a real need exists to protect our immune systems and lead healthier lives. Hence the present study is aimed to evaluate the immunomodulatory activity of the Flavonoid of *Kigelia africana* (Family: Bignoniaceae). **Methods:** The assessment of immunomodulatory activity on cell-mediated and humoral mediated immunity were studied by Carbon clearance test; Cyclophosphamide induced neutropenia, Indirect Haemagglutination reaction, Neutrophil adhesion test, and effect on Serum immunoglobulin level. **Results:** Flavonoid of *Kigelia africana* was administered orally at a low dose and a high dose of 100mg/kg/day, po and 200 mg/kg/day, po respectively and *Levamisole* (2.5 mg/kg/day, po) was used as standard drug. Flavonoid of *Kigelia africana* in both doses showed a significant increase in the phagocytic index in carbon clearance assay, significant protection against Cyclophosphamide induced neutropenia. It also increased the circulating antibody titer in an indirect haemagglutination test and increased the adhesion of neutrophils in the neutrophil adhesion test. Hence, it was concluded that Flavonoid of *Kigelia africana* increases both humoral immunity and cell-mediated immunity. **Conclusion:** Flavonoid of *Kigelia africana* increases both humoral immunity and cell-mediated immunity.

INTRODUCTION: Immune system is a complex organization of white blood cells, antibodies, and blood factors that protects the body from foreign invaders, while simultaneously maintaining self-tolerance. The basic function of the immune system is to protect against foreign pathogens and infectious agents. This is achieved either through innate or natural immunological mechanisms which essentially serve as a short term first-line defense or through elaborate adaptive mechanisms which are highly specific, complex, and marked by diversity and memory.

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.5(10).4359-65

This article can be accessed online on www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.5(10).4359-65

When the immune system hits the wrong target, however, it can unleash a torrent of disorders, including allergic diseases, arthritis, and a form of diabetes. If the immune system is crippled, other kinds of diseases result in [1].

Due to the wide range of effector mechanism possessed by various groups of immune cells and its ability to exert effects with exquisite specificity, the immune system provides a good target in cancer therapy. Involvement of the host immune system in the control of cancer progression has been suspected but remained inconclusive for many years. Innate immunity, which, according to the immune surveillance theory is responsible for early detection and elimination of malignant cells, may be inefficient patients who develop a malignancy. Evidence is convincing that individuals who are older, who have been on immunosuppressive medications over prolonged periods, or have

underlying immune abnormalities, such as an autoimmune disease or a chronic infection (*e.g.*, AIDS) are particularly at risk of malignancy. Traditional and folklore medicines play an important role in health services around the globe. About three-quarters of the world's population relies on plants and plant extracts for healthcare. India has an extensive forest cover, enriched with plant diversity. Several plants have been used in folklore medicine. The rational design of novel drugs from traditional medicine offers new prospects in modern healthcare.[6] Ayurveda, the traditional medicinal system in India, describes certain plants which strengthen the host immune system [2].

Kigelia africana (Lam.) Benth. (Bignoniaceae) is widespread across India and Africa and is found in most wet savannah and river line areas. Growing over 20 m high, it is semi-deciduous with smooth grey-brown bark. The fruits are large grey-green "sausages" about 30-60 cm long which hang on stalks from the tree. The fresh fruit is poisonous and strongly purgative; for safety reasons, fruits are best prepared for consumption by drying, roasting, and fermentation. The scientific literature confirms the validity of many of these traditional uses due to the presence of numerous secondary metabolites. These compounds include iridoids, flavonoids, fatty acids, sterols, glycosides, and naphthoquinones [3, 4].

The present study was undertaken to evaluate the effect of flavonoid of *Kigelia africana* on the immune system using different experimental models to substantiate the traditional claim. The study helps in understanding the effect of flavonoid of *Kigelia africana* on different components on the immune system.

MATERIALS AND METHODS:

Experimental Animals: Albino Wistar rats weighing between 200-250 gm and Swiss albino mice weighing between 25-35 gm were used. Institutional Animal Ethics Committee approved the experimental protocol; animals were maintained under standard conditions in an animal house approved by Committee for Control, and Supervision on Experiments on Animals (CPCSEA). The animals were given pellet food (Lipton India Ltd., Mumbai, India) and water *ad libitum*. (Reg. no. 1564/PO/a/11/CPCSEA).

Plant Material: The leaves of *Kigelia africana* were collected from Thirupathi forest region Thirupathi District, Andhra Pradesh, India in June 2013. This plant species were authenticated by Dr. K. Madhava Chetty, Assistant professor, Department of Botany, Sri Venkateswara University, Thirupathi, Andhra Pradesh, India, where a voucher specimen has been preserved for future identification.

Preparation of Extract: 100 gm powdered leaves parts were subjected to successive extraction in a Soxhlet extractor using methyl alcohol. The extract obtained was concentrated in a rotary shaker and evaporated to dryness to get constant weight.

Preliminary Phytochemical Screening: [5] Methanolic extracts of KA were subjected to preliminary phytochemical screening using the methods described by Kokate, Trease, and Evans for the detection of various plant constituents.

Isolation of Flavonoid: [6] The method employed for the isolation of flavonoids was of Jang *et al.* (2003) with modifications. 200 gm of *K. africana* powder were defatted with hexane is a Soxhlet extractor. The marc was pressed and dried. The defatted powder was extracted with methanol in a Soxhlet extractor for 16 h. The methanol extract was filtered and concentrated to dryness in a rotary shaker evaporator. The concentrated extract was dissolved in 80% methanol with stirring and filtered. Methanol fractions were subjected to chemical analysis. Methanol extract fraction was concentrated kept in the refrigerator overnight. As there was no crystal formed, the solvent was evaporated to dryness. The residue answered the test for flavonoids.

Chemicals: Leishman's stain, Indian ink, and glut-araldehyde were purchased from Merck (Mumbai, India). WBC diluting fluid, zinc sulfate, and barium chloride were from Nice Chemicals (Cochin, India). Cyclophosphamide (Endoxan Injection) was from German Remedies (Mumbai, India).

Antigen: Fresh blood was collected from sheep sacrificed in the local slaughterhouse. Sheep red blood cells (SRBCs) were washed three times in large volumes of Alsever's solution and adjusted to a concentration of 0.5×10^9 cells/ml for immunization and challenge.

Selection of Dose and Treatment period: The animals were divided into four groups consisting of six animals each. The first group served as control (vehicle 1 ml/100 g, po), the second group received Levamisole at a dose of (2.5mg/Kg, po).The third and fourth group received the low dose (100mg/Kg, po) and a high dose of Flavanoid of Kigelia africana respectively.

Acute Toxicity Studies: [9] (http://www.epa.gov/oppts/home/guideline.htm)

The acute toxicity study was carried out according to the limit test described in the OPPTS guidelines. Briefly, a test dose of 2 g/kg and 5 g/kg were given orally to the mice. The extract was found to be safe at the dose of 2 g/kg, po. Hence, 1/10th and 1/20th of the safe dose corresponding to 100 mg/kg and 200 mg/kg orally were selected as a low and high dose, respectively.

Carbon Clearance Test: [11] Swiss albino mice were treated with the drug or vehicle orally for 5 days. After 48 h of the last dose of the drug, animals have injected 0.1 ml of Indian ink via the tail vein. Blood samples were withdrawn at 0 and 15 min after injection. A 50 µL blood sample was mixed with 4 ml of 0.1% sodium carbonate solution, and the absorbance of this solution was determined at 660 nm. The phagocytic index K was calculated using the following equation:

$$K= (\text{Loge OD1}-\text{Loge OD2})/15$$

Where OD1 and OD2 are the optical densities at 0 and 15 min respectively.

Cyclophosphamide Induced Neutropenia: [12] Swiss albino mice received the drug or vehicle orally for 10 days. On the 10th day, a neutropenic dose of Cyclophosphamide (200 mg/kg, sc) was injected, and this day was labeled as day zero. Blood was collected, the total leukocyte count (TLC) and DLC were performed before and on day 3 after injection of Cyclophosphamide. The TLC and neutrophil counts (%) in treated groups were compared with the values of the control group.

Indirect Haemagglutination Test (IHA Test) [13, 14]**:** Rats were pre-treated with the drugs for 14 days, and each rat was immunized with 0.5×10^9 sheep red blood cells (SRBCs) intraperitoneally, including control rats. The day of immunization was referred to as day 0. The drug treatment was continued for another 14 more days, and blood samples were collected from each rat at the end of the drug treatment, and the titer value was determined by titrating serum dilutions with SRBC (0.025×10^9 cells) in microtitre plates. The plates were incubated at room temperature for 2 h and examined visually for agglutination. The minimum volume of serum showing Haemagglutination was expressed as Haemagglutination (HA) titer.

Neutrophil Adhesion Test: [17] The rats were treated orally with vehicle or extracts for 14 days. On day 14, blood samples were collected from the retro-orbital plexus into heparinized vials and analyzed for differential leukocyte count (DLC). After the initial counts, blood samples were incubated with 80 mg/ml of nylon fibers for 10 min at 37 °C. The incubated blood samples were again analyzed for DLC. The percentage of neutrophils in the treated and untreated blood was determined, and the difference was taken as an index of neutrophil adhesion.

Effect on Serum Immunoglobulin: [18, 19] The drugs were administered to female albino rats orally for 21 days. Six hours after the last dose of the drug, blood was collected, and the serum was used for estimation of immunoglobulin levels using the method devised by Mullen (1975). Briefly, for each serum sample to be analyzed, a control tube containing 6 ml of distilled water and a test tube containing 6 ml of zinc sulfate solution was prepared. To each, 0.1 ml of serum was added from a pipette.

They were inverted to enable complete mixing of the reagents and left to stand for 1 hr at room temperature. The first tube served as blank and the second tube was taken as a sample. The turbidity developed was measured using a digital nepheloturbidity meter. The turbidity obtained (sample-blank) was compared with that obtained with standard barium sulfate ($BaSO_4$) solution. The standard $BaSO_4$ solution was prepared by adding 3 ml of barium chloride solution (1.15% w/v) to 97 ml of 0.2 N sulphuric acids. The turbidity obtained with this solution was expressed as 20 zinc sulfate turbidity (ZST) units.

Statistical Analysis: The results are expressed as mean ± S.E.M. Data analyzed by one way ANOVA followed by the "Turkey's Multiple Comparison Test." P values are $P<0.05^{*}$, $P<0.01^{**}$, $P<0.001^{***}$ as compared with control were considered.

RESULTS:

Acute Oral Toxicity: The LD_{50} was found to be 2000mg/kg. So $1/10^{th}$ and $1/20^{th}$ of the dose were taken for the study.

Preliminary Phytochemical Screening: The presence of various phytoconstituents of the extracts was detected by phytochemical screening. The KA found to contain Steroids and Triterpenes, phenolic compounds, tannins, and flavonoids.

Effect on Carbon Clearance Test: Both the doses of Flavanoid of *Kigelia africana* and LMS showed significant ($^{*}P<0.05$) increase in the phagocytic index when compared to control indicating that there was an increase in the clearance of colloidal carbon from the blood after administration of these drugs.

TABLE 1: EFFECT OF FLAVONOID OF *KIGELIA AFRICANA* ON PHAGOCYTIC ACTIVITY BY CARBON CLEARANCE TEST IN SWISS ALBINO MICE

Treatment	Phagocytic index
Vehicle	0.0052 ± 0.0035
LMS (2.5 mg/Kg,po/day/5days)	0.0211 ± 0.0033*
FKA (100 mg /Kg,po/day/5days)	0.0129 ± 0.0033*
FKA (200 mg /Kg,po/day/5days)	0.0165 ± 0.0035*

Values are expressed as mean ± SEM. n = 6 $^{*}P<0.05$ when compared to control. Statistically analyzed by one way ANOVA followed by Tukey-Kramer multiple comparison test.

Effect on Cyclophosphamide Induced Neutropenia: The neutropenic dose of Cyclophosphamide reduced the TLC in control animals by 57.87%. Administration of Flavonoid of *Kigelia africana* for 10 days before Cyclophosphamide administration at a low dose and high dose produce 51% and 38.64% reduction in TLC respectively, while LMS pre-treatment produced 30.99% decrease in TLC when compared to control. The neutrophils count (%) was reduced by 75.71% in Cyclophosphamide treated control, 59.14% and 13.7% in low dose and a high dose of FKA, respectively. LMS pre-treatment produced 13.7% decrease in neutrophil count T**able 2**.

Effect on Indirect Haemagglutination Test: Antibody molecules which are secreted by plasma cells mediate the humoral immune response.

The FKA showed a significant (*P*<0.001) increase in the hemagglutination titer at doses of 100 mg/kg, po, and 200 mg/kg, po in animal studies compared to the control group. This augmentation of the humoral response to SRBC indicated enhanced responsiveness of the macrophages and T and B lymphocyte subsets involved in antibody synthesis T**able 3**.

Effect on Neutrophil Adhesion Test: Incubation of neutrophils with nylon fibers produced a decrease in the neutrophil counts due to adhesion of neutrophils to the fibers. Both doses of Flavonoid of *Kigelia africana* and LMS showed significant (***$P<0.001$) increase in the neutrophil adhesion when compared to control **Table 4**.

Effect on Serum Immunoglobulin: Both the doses of Flavanoid of *Kigelia africana* and LMS showed a significant ($^{**}P<0.01$, $^{***}P<0.001$) increase in the serum immunoglobulin levels when compared to control.

TABLE 2: EFFECT OF FLAVONOID OF *KIGELIA AFRICANA* ON CYCLOPHOSPHAMIDE INDUCED NEUTROPENIA IN SWISS ALBINO MICE

Treatment	Number of leucocytes after cypm treatment	% Reduction of leucocytes	Number of neutrophils after cypm treatment	% reduction of Neutrophils
Vehicle (1 ml /Kg,po/day/10days)	3030 ± 111.36	57.87	16.2 ± 0.74	75.7
LMS (2.5mg/Kg,po/day/10days)	2110 ± 92.73***	30.99	02.4 ± 0.25***	9.03
FKA (100mg/Kg,po/day/10days)	2550 ± 72.45	51	13.6 ± 0.68	59.14
FKA (200mg/Kg,po/day/10days)	2260 ± 88.60***	38.64	03.4 ± 0.25***	13.7

Values are expressed as mean ± SEM. n = 6 $^{***}P<0.001$ when compared to control. Statistically analyzed by one way ANOVA followed by Tukey-Kramer multiple comparison test.

TABLE 3: EFFECT OF FLAVONOID OF *KIGELIA AFRICANA* ON INDIRECT HAEMAGGLUTINATION TEST IN WISTAR RATS

Treatment	HA titer value (µL/100 µL)
Vehicle (1 ml /Kg,po/day/28days)	2083 ± 0.03
LMS (2.5 mg/Kg,po/day/28days)	0.942 ± 0.05***
FKA (100 mg /Kg,po/day/28days)	0.641 ± 0.05***
FKA (200 mg /Kg,po/day/28days)	0.822 ± 0.04***

Values are expressed as mean ± SEM. n = 6. $^{***}P<0.001$ when compared to control. Statistically analyzed by one way ANOVA followed by Tukey-Kramer multiple comparison test.

TABLE 4: EFFECT OF FLAVANOID OF KIGELIA AFRICANA ON NEUTROPHIL ADHESION TEST IN WISTAR RATS

Treatment	Neutrophils adhesion index(UB-NFTB)
Control- Distilled Water (1 ml /Kg, po /day / 14days)	12.01 ± 0.67
LMS (2.5 mg /Kg,po /day / 14days)	34.48 ± 2.23 ***
FKA (100 mg /Kg,po /day /14days)	30.12 ± 1.22 ***
FKA (200 mg /Kg,po /day/ 14days)	33.04 ± 2.14 ***

Values are expressed as mean ± SEM. n=6, $^{***}P<0.001$ when compared to control. Statistically analyzed by one way ANOVA followed by Tukey-Kramer multiple comparison test

TABLE 5: EFFECT OF FLAVONOID OF *KIGELIA AFRICANA* ON SERUM IMMUNOGLOBULIN LEVELS IN WISTER RATS

Treatment	Serum immunoglobulin level (ZST-UNITS)
Control-Distilled Water (1 ml /Kg,po/day/21days)	21.12 ± 0.55
LMS (2.5 mg/Kg,po/day/21days)	38.14 ± 1.40***
FKA (100 mg /Kg,po/day/21days)	28.4 ± 0.85**
FKA (200 mg /Kg,po/day/21days)	33.04 ± 1.33***

Values are expressed as mean ± SEM. n = 6, $^{**}P<0.01$, $^{***}P<0.001$ when compared to control. Statistically analyzed by one way ANOVA followed by Tukey-Kramer multiple comparison test

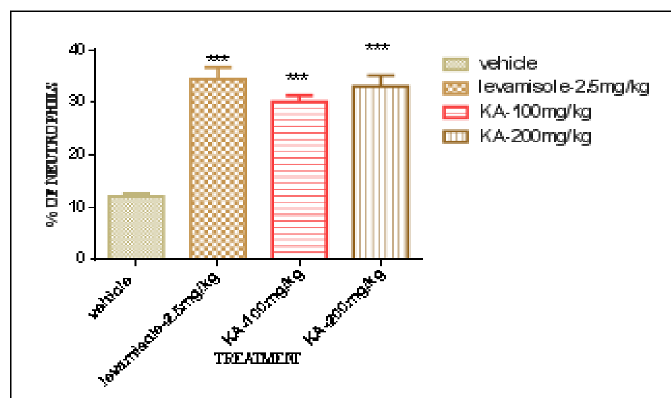

FIG. 1: EFFECT OF FLAVONOID OF *KIGELIA AFRICANA* ON NEUTROPHIL ADHESION TEST IN WISTAR RATS. Values are expressed as mean ± SEM. n=6, $^{***}P<0.001$ when compared to control.

DISCUSSION: The results of the present study suggest that Flavonoid of *Kigelia africana* affects humoral immunity as shown by its effect in the indirect Haemagglutination test and serum immunoglobulin levels and it also has an effect on the cell-mediated immunity as it showed a significant increase in the neutrophil adhesion, carbon clearance and a reduction in cyclophosphamide-induced neutropenia.

The carbon clearance test was done to evaluate the effect of drugs on the reticuloendothelial system. The reticuloendothelial system (RES) is a diffuse system consisting of phagocytic cells. Cells of the RES play an important role in the clearance of particles from the bloodstream. When colloidal carbon particles in the form of ink are injected directly into the systemic circulation, the rate of clearance of carbon from the blood by macrophage is governed by an exponential equation. Flavonoid of *Kigelia africana* at both doses and Levamisol showed an increase in the phagocytic index. Hence, these agents may increase the activity of the reticuloendothelial system [20].

The Cyclophosphamide induced Neutropenia model concentrates on the effect of drugs on the hemopoietic system. Flavonoid of *Kigelia africana* at 100mg/Kg, po/day/10 days and 200mg/Kg, po/ day/10 days caused 59.14% and 13.7% decrease in the Cyclophosphamide induced Neutropenia suggesting that it attenuates the effect of Cyclophosphamide on the hemopoietic system.

The indirect Haemagglutination test was performed to confirm the effect of Flavonoid of *Kigelia africana* on the humoral arm of the immune system. The humoral immunity involves the interaction of B cells with the antigen and their subsequent proliferation and differentiation into antibody-secreting cells. Antibody functions as the effectors of the humoral response by binding to antigen and neutralizing it or facilitating its elimination by cross-linking to form latex that is more readily ingested by phagocytic cells. Flavonoid of *Kigelia africana* at both the doses (100 & 200 mg/kg) and Levamisole showed that levels of circulating antibodies are significantly increased [21]. The adhesion of neutrophils to nylon fibers describes the margination of cells in the blood vessels and the number of neutrophils

reaching the site of inflammation. Flavonoid of *Kigelia africana* at both doses showed a significant increase in the neutrophils adhesion to nylon fibers. This might be due to the up-regulation of the β2 integrins, present on the surface of the neutrophils through which they adhere firmly to the nylon fibers. Hence, it was inferred that Flavonoid of *Kigelia africana* causes stimulation of neutrophils towards the site of inflammation [23].

The estimation of serum immunoglobulin levels was used to evaluate the increase in serum immunoglobulin production after the administration of the drugs. Immunoglobulins are antibodies that react specifically with the antigen. The zinc sulfate turbidity test is used to gain a rough estimation of the amount of Immuno-globulins present in the serum. Zinc sulfate causes precipitation of the Immunoglobulins making the solution cloudy. A lack of cloudiness signifies lack of immunoglobulins. The turbidity is expressed as ZST units, which in turn indicate the amount of immunoglobulin present in the sample. Flavonoid of *Kigelia africana* at both the doses (100&200mg/kg) showed a significant increase in the serum immunoglobulin levels. [24, 25]

Findings of the present study showed an overall stimulatory effect of Flavonoid of *Kigelia africana* on both humoral and cellular immunity.

CONCLUSION: In the present study, Flavonoid of *Kigelia africana* showed immunostimulant activity. Thus, it can be concluded that Flavonoid of *Kigelia africana* has therapeutic potential and could be served as an effective immunomodulatory candidate without any side effects and support the traditional claim of *Kigelia africana* for medicinal purposes.

ACKNOWLEDGEMENT: Authors are thankful to the Department of Pharmacology, Karnataka College of Pharmacy, Bangalore, Karnataka, for providing constant support and lab facilities.

CONFLICT OF INTEREST: Nil

REFERENCES:

1. Premanathan M, Rajendran S, Ramanathan T, Kathiresan K, Nakashima H and Yamamoto N: A survey of some Indian medicinal plants for anti-human immunodeficiency virus (HIV) activity. Indian J Med Res 2010; 112: 73-7.
2. Fong HH: Integration of herbal medicine into modern medical practices: issues and prospects. Integr Cancer Ther 2002; 1: 287.
3. Huffman MA: Animal self-medication and ethnomedicine: exploration and exploitation of the medicinal properties of plants. Proc Nutr Soc, 2003; 62: 371.
4. Miller KL, Liebowitz RS and Newby LK: Complementary and alternative medicine in cardiovascular disease: a review of biologically based approaches. Am Heart J 2004; 147: 401.
5. Limei Y, Mouming Z, Jin SW, Chun C, Bao Y, Yueming J and Qiangzhong Z: Antioxidant, immunomodulatory and anti-breast cancer activities of phenolic extract from pine (Pinus massoniana Lamb) bark. Innovative Food Sci Emerging Techno 2008; 9: 122.
6. Thatte UM, Kulkarni MR and Dahanukar SA, Immunotherapeutic modification of Escherichia coli peritonitis and bacteremia by Tinospora cordifolia, J Postgrad Med 1992; 38: 13.
7. Rege NN, Thatte UM and Dahanukar SA: Adaptogenic properties of six rasayana herbs used in Ayurvedic medicine, Phytother Res 2008; 3: 275.
8. Parab S, Kulkarni R and Thatte UM: Heavy metals in ‘herbal’ medicines, Indian J Gastroenterol 2003; 22: 111.
9. Yin L, Jie F, Yunpeng Z and Shaoyuan C: Immunomodulatory activity of aqueous extract of *Actinidia macrosperma*. Asia Pac J Clin Nutr 2007; 16: 261-65.
10. Ismail S and Asad M: Immunomodulatory activity of *Acacia catechu*. Indian J Physiol Pharmacol 2010; 53(1): 25-33.
11. Gaur K, Kori ML and Nema RK: Comparative screening of immunomodulatory activity of hydro-alcoholic extract of *Hibiscus rosa sinensis* Linn. and ethanolic extract of *Cleome gynandra* Linn. Global Journal of Pharmacology 2012; 3(2): 85-89.
12. Bafna A and Mishra S: Antioxidant and immunomodulatory activity of the alkaloidal fraction of *Cissampelos pareira* Linn. Sci Pharm 2010; 78: 21-31.
13. Bharani S, Asad M, Sunil SD and Chandrakala GK: Immunomodulatory activity of methanolic extract of *Morus alba* linn. (Mulberry) leaves. Pak J Pharm Sci 2010; 23(1): 63-68.
14. Gang X, Schepetkin IA and Quinn MT. Immunomodulatory activity of acidic polysaccharides isolated from *Tanacetum vulgare* L. Int Immunopharmacology 2010; 1639-50.
15. Khanittha P, Christopher P, Watchara K and Usanee V: Immunomodulatory activities of *Centella asiatica* and *Rhinacanthus nasutus* extracts. Asian Pac J Cancer Prev 2005; 6: 396-00.
16. Syamsudin, Rita and Kusmardi: Immunomodulatory and *in-vivo* antiplasmodial activities of propolis extracts. Am J Pharmacol Toxicol 2009; 4(3): 75-79.
17. Satpute KL, Jadhav MM, Karodi RS, Katare YS, Patil MJ and Rub R: Immunomodulatory activity of fruits of *Randia dumetorum* Lamk. J. Pharmacognosy Phytother 2009; 1.
18. Fariba S, Shirin P, Moslem A, Ramin R, Omid R and Amin P: Immunomodulatory activity of aqueous extract of *Heracleum persicum* Desf. in Mice. Iran J Pharm Res 2012; 8(4): 287-92.
19. Mayank T, Shilpi B and Dixit VK: Immunomodulatory activity of *Chlorophytum borivilianum* Sant. ECAM 2007; 4(4): 419-23.
20. Geo-Vigila A and Baskaran X: Immunomodulatory effect of coconut protein on cyclophosphamide induced immune

suppressed swiss albino mice. Ethnobotanical Leaflets 2008; 12: 1206-12.

21. Ramanatha KR, Lakshminaryana R and Gopal T: Potency test of Duck Pasteurella vaccine in mice. Mysore J Agri Sci 1995; 29: 155-57.
22. Thatte UM, Chhabria SN, Karandikar SM and Dahanukar SA: Protective effects of Indian medicinal plants against cyclophosphamide neutropenia. J Postgrad Med 1987; 33(4): 185-88.
23. Jayathirtha MG, Mishra SH: Preliminary immunomodulatory activities of methanol extracts of *Eclipta alba* and *Centella asiatica*. Phytomedicine 2004; 11: 361-65.
24. Gokhale AB, Damre AS and Saraf MN: Investigations into the immunomodulatory activity of *Argyreia speciosa*. J Ethnopharmacol 2003; 84: 109-14.
25. Mullen PA: Zinc sulfate turbidity test as an aid to diagnosis. Veter Annu 1975; 15: 455-57.

How to cite this article:

Nagarathna PKM, Reena K, Reddy S and Wesley J: Evaluation of immunomodulatory activity of the flavanoid of *Kigelia africana*. Int J Pharm Sci & Res 2014; 5(10): 4359-65. doi: 10.13040/IJPSR.0975-8232.5(10).4359-65.

All © 2013 are reserved by International Journal of Pharmaceutical Sciences and Research. This Journal licensed under a Creative Commons Attribution-NonCommercial-ShareAlike 3.0 Unported License.

This article can be downloaded to **ANDROID OS** based mobile. Scan QR Code using Code/Bar Scanner from your mobile. (Scanners are available on Google Playstore)