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# Formulation and Evaluation of Alugbati-Based Nasal Spray: A Preliminary study for the Adjunct Management of Allergic Rhinitis

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**Key words:** Allergic rhinitis, Alugbati, *Basella alba*, Formulation, Nasal spray.

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## Abstract

Allergic rhinitis is a nasal inflammation produced by an allergic reaction to allergens that affects a significant number of Asian populations, particularly in tropical countries like the Philippines. The Philippine Bureau of Plant Industry promotes Basella alba, also known as Alugbati, which has been reported to have cytotoxic, anti-inflammatory, and wound-healing properties. The purpose of this study is to develop and evaluate the anti-inflammatory effect of *B. alba* Leaf extract as a nasal spray for allergic rhinitis in Sprague Dawley rats. 27 rats were divided into three groups: the negative control, positive control, and the treatment group which received the formulated B. alba nasal spray. The inflammatory impact of white blood cells (WBC) was studied in each group. Ciliary loss, reduction in goblet cells, degree of eosinophil infiltration, and vascular congestion were graded and observed during the histological investigation. Compared to the positive control group, the Formulation group showed no significant change in the histological scores but displayed clinical significance as shown in the samples microscopically. When observing WBC count, there's a significant difference between the Formulation group and the Positive control group, posthoc analysis revealed that the Formulation group was more effective in normalizing WBC count 24 hours post provocation/administration. An acute inhalation toxicity test showed that the extract is classified as “practically non-toxic”. Therefore, formulated Alugbati nasal spray is a potential alternative and is effective to use in an animal model of allergic rhinitis. Further tests such as ELISA are recommended to specifically determine the mechanism of action of the formulation.

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## Introduction

Allergic rhinitis is an inflammation of the nasal passages caused by an allergic reaction to airborne substances. This disease is the most common atopic disease globally, affecting 10-20% of the population, and the disease prevalence has shown to increase. Severe allergic rhinitis is associated with severe impairments in quality of life, sleep, and employment performance (Dykewicz *et al.*, 2010; Abong *et al.*, 2012). Allergic rhinitis is a prevalent yet underdiagnosed atopic disease that commonly presents at least one of the following clinical symptoms: persistent nasal obstruction, mucous discharge, sneezing, itching and may co-exist with asthma (Small *et al.*, 2007; Bourdin *et al.*, 2009; Chong *et al.,* 2018). The most common therapy for allergic rhinitis is intranasal corticosteroids. They work by reducing inflammation in the nasal mucosa by limiting the influx of inflammatory cells and suppressing the release of cytokines. The onset of therapeutic action for inhaled corticosteroids is 30 minutes due to its broad surface for absorption, but the peak effect might take several hours to days. (Sur *et al.*, 2010; Kimbell *et al.*, 2007). The costs of these medicines have risen dramatically, making them unaffordable for anyone who requires and needed them; thus, nasal spray becomes more and more neglected for its high price and this is why people prompts to seek out alternatives and make effective use of herbs orally which is not the fastest choice to treat allergic rhinitis (Luqman *et al.*, 2014).

The *Basella alba* plant, locally named 'Alugbati,' is one of the indigenous vegetables promoted by the Philippine Bureau of Plant Industry for consumption. The plant has enormous potential for androgenic activity, antiulcer activity, antioxidant; cytotoxic activity and antibacterial activity; anti-inflammatory activity; depressant activity of the central nervous system; nephroprotective, wound healing, and many more (Kumar *et al.*, 2013; Duke *et al.*, 1985; Grubben *et al.*, 2004; Khare 2007). Reverse Phase-High Performance Liquid Chromatography (RP-HPLC) revealed the presence of secondary metabolites such as naringin, apigenin, ascorbic acid, a-tocopherol, and Luteolin (Lut), a kind of flavonoid, possesses anti-oxidative, anti-tumor, and anti-inflammatory properties (Gunasekaran, 2015; Kachchhava, 2006; Krishna, 2012; Rodda *et al.,* 2012). With the established anti-inflammatory property and abundance of *B. alba* in Cagayan Valley, the researchers aim to develop and evaluate Alugbati (*Basella alba* L.) based nasal spray for allergic rhinitis. To the best of the researchers' knowledge, this is the first reported study using *B. alba* for allergic rhinitis using the nasal spray.

## Materials and methods

*Source of plant sample*

Leaves of *B. alba* were collected from Bitag Grande, Baggao, Cagayan. Samples were rinsed before disinfecting with Zonrox (1ml:1L), air-dried at room temperature, and pulverized using a kitchen blender.

*Aqueous Extraction*

A 25-gram ground air-dried *B. alba* leaves (samples) were homogenized with 100 mL distilled water (solvent) in a kitchen blender (1:4 sample to solvent ratio). The prepared solution was kept in a beaker and sonicated for 45 minutes at room temperature (25±1°C). The ultrasonic treatment was performed using an ultrasonic processor. To maintain a constant room temperature (25±1 °C), an oven thermometer was used, and for any slight changes that were observed, freshwater was circulated. Moreover, all sonication was made in the dark to avoid exposure to light which can alter the process. After extraction, the sample residues were dissolved in a quantified solvent level (50 mL distilled water) and were extracted repeatedly until the extracts became clear. The gathered extracts were filtered using a Whatman No. 1 filter paper and will be stored at 4°C in an airtight container (screw type) until further analysis.

*Maintenance of Sprague Dawley rats*

This study was carried out using 25 healthy male Sprague Dawley rats with an average weight of 250–300g, obtained from the authorized seller of laboratory rats located in Santiago City. The study was started after obtaining an animal permit and

research clearance from the Department of Agriculture, Bureau of Animal Industry (Reference no. AR-2021-140 dated 12/7/2021).

The rats were maintained in cages of no more than five animals each at an ambient temperature of 26±1°C, and they were fed the usual laboratory dieting, each under a 12-hour-light–12-hour-dark cycle for seven consecutive days. During the trial, animals may drink water and consume food ad libitum.

The animals were divided into three groups of nine rats each. The negative control group (ovalbumin [OVA] sensitization and provocation + intranasal PNSS provocation). In this group of rats, sensitization and provocation were made with OVA; PNSS was given as intranasal therapy. The positive control group (ovalbumin [OVA] sensitization and provocation + intranasal Fluticasone Propionate). In this group of rats, sensitization and provocation were made with OVA, and Fluticasone Propionate was given as intranasal treatment. And the formulated group (ovalbumin [OVA] sensitization and provocation + intranasal formulated aqueous *B. alba* extract). In this group of rats, sensitization and provocation were made with OVA, and a formulated aqueous *B. alba* extract was given as intranasal treatment

*Sensitization*

The initial step of our investigations on animal immunization was conducted following acclimatization to laboratory settings. All animals have been sensitized as an antigen, with the use of OVA (0.3 mg i.p., Sigma, St. Louis, Mo., USA) and aluminum hydroxide (30 mg) in saline (1 ml i.p.) 3 every other day over 14 days (Wen, 2007; Senturk *et al.*, 2017).

*Provocation*

Daily actuations using a spray bottle with 30 ml 10% OVA solution dissolved in 1 ml PS into both nostrils were used to induce allergic rhinitis in rats sensitized with IP OVA for 14 consecutive days. Allergic rhinitis was observed regardless of the severity of typical clinical symptoms that included nasal irritation, sneezing, and nasal secretion. On day 15, which was the first day of provocation, nasal scratching, sneezing, and nasal flow were observed. The AR model was considered successful if symptoms recurred every after provocation (Wen, 2007; Senturk *et al.*, 2017).

*Drug treatment*

All groups underwent sensitization with IP OVA for two weeks. After sensitization, AR was provoked by inducing intranasal OVA 1 hour before treatment of the following: Normal Physiological saline (Negative Control group), Fluticasone Propionate (Positive Control), and the Formulated Alugbati Nasal Spray for a total of 3 treatments. The animals' respiration was monitored closely to ensure that they will not stop breathing. The animals were sacrificed after 24 hours following the last medication application. A blood sample was collected through the tail with a process of tail snip technique for complete blood count to detect the anti-inflammatory response of the extract. The nasal cavity, nasal septum, paranasal sinus, and conchae of the animals were removed in one piece. An incision will be made starting just in front of the incisors and stretching to the end of the hard palate for this reason. Tissue samples from the mucosa of the nasal septum were collected for histological analysis.

*Acute Inhalation Toxicity Test*

Six (6) healthy young adult male Sprague Dawley rats 8 to 12 weeks of age, with body weights ranging from 250-300 will be divided and nebulized individually with the extracts in increasing dose levels of 1.2.3 via nose-only nebulization. The toxicity was observed continuously for multiple durations (15, 30, 60, 120, 240 minutes). It was carried out following the OECD TG 433 (OECD, 2018) for Acute Inhalation Toxicity.

*Histological Assessment*

The nasal cavity, nasal septum, paranasal sinus, and conchae, of the test animals, were removed en bloc. For this purpose, an incision was performed

beginning at the level right in front of the incisors and stretching to the end of the hard palate, then tissue samples were taken from the mucosa of the nasal septum. The septal mucosa samples that were taken from the sacrificed animals were fixed in 10% neutral buffered formalin for 72 hours. The nasal mucosa underwent decalcification for 24 hours using nitric formol before cutting it into sections. Tissue samples were taken from the nasal mucosa of the nasal septum specifically at level 2 naso and maxillo turbinates. The tissues were dehydrated in an alcohol series of 70, 80, 90, 96, and 100 percent ethyl alcohol after fixation. The samples were softened in xylene for 3 hours before being molded in paraffin at 60 degrees Celsius. Microtome pieces of approximately 5 µm thickness were sliced after molding. Hematoxylin and eosin were used to stain the sections.

An optical microscope was used to analyze stained materials. Ciliary loss, an increase in goblet cells, vascular congestion, and the degree of eosinophil infiltration were assessed under the microscope (Senturk *et al*, 2017). According to Ercan *et al.* (2006), the scoring was done semi-quantitatively, with each parameter receiving a score. "0" points in the absence of any modification, "1" for a slight change, "2" for noticeable change, and "3" points for severe change. The averages of the

*Complete Blood Count (CBC) Testing*

All blood samples were collected in EDTA-tube microvettes which were purchased among the pharmacies and boutiques located within the vicinity of Tuguegarao City. Blood samples were collected by cardiac perfusion prior to sacrificing the test subjects. The Complete Blood Count (CBC) testing, including white blood cell (leukocyte) differential count, was determined with the assistance of the Department of Agriculture (Carig Norte, Tuguegarao City) research specialist.

*Statistical Analysis*

The mean and standard deviation was utilized to determine the anti-inflammatory effects of Alugbati leaf extract nasal spray in terms of decrease in white blood cells, decrease ciliary loss, a decrease in eosinophil infiltration, decrease goblet cells, and decrease vascular congestion in the nasal mucosa of the test animals. Furthermore, the One Way ANOVA (Analysis of Variance) is used to determine whether there are any statistically significant differences between the formulation, positive control, and negative control. Least Significant Difference (LSD) was also used to evaluate the significant differences among various treatments. All statistical runs were performed using SPSS version 26.

**Results and discussion**

*Phytochemical analysis*

Color changes are used in phytochemical screening to determine if particular phytochemicals are present. The presence of phytochemicals such as alkaloids, diterpenes, phenols, tannins, and flavonoids in the aqueous extract was determined in previous work by Tongco *et al.* (2015) using qualitative phytochemical screening of extracts from Alugbati leaves.

**Table 1.** Qualitative screening of ethanol and aqueous extracts of *Basella alba* leaves (Tongco *et al.*, 2015).

| Phytochemical test | Aqueous extract |
|---|---|
| Alkaloids | ++ |
| Carbohydrates | - |
| Cardiac Glycosides | - |
| Anthranol Gycosides | - |
| Cyanogenic Glycosides | - |
| Saponins | - |
| Diterpenes | + |
| Triterpenes | - |
| Phenols | + |
| Tannins | + |
| Flavonoids | + |

The anticancer, antioxidant, and anti-inflammatory activities of B. alba are attributed to phytochemicals found in the aqueous extracts according to the literature. According to Gunasekaran (2015), GS-MS/MS analysis detected 25 compounds, while Reverse Phase-High Performance Liquid Chromatography (RP-HPLC) revealed the presence of or naringin, apigenin, ascorbic acid, and -tocopherol, and luteolin, a flavonoid with anti-oxidative, anti-tumor, and anti-inflammatory properties.

**Table 2.** Organoleptic properties.

| Organoleptic property | Observation |
|---|---|
| Color | Brownish-yellow |
| Odor | Odorless |
| Texture | Watery |
| Opacity | Transparent |

**Table 3.** Mortality rate during nasal toxicity test.

| Alugbati concentration (mL/L) | Mortality per Time interval | | | | |
|---|---|---|---|---|---|
| | After 15 mins | After 30 mins | After 60 mins | After 120 mins | After 240 mins |
| 5 mL/L | 0/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 3 mL/L | 0/2 | 0/2 | 0/2 | 0/2 | 0/2 |
| 1 mL/L | 0/2 | 0/2 | 0/2 | 0/2 | 0/2 |

*Formulated Alubati- based Nasal Spray*

For the formulation of Alugbati-based nasal spray, a 3 ml of Alugbati aqueous extract (obtained from Bitag Grande, Baggao, Cagayan) was added to 297 ml of plain normal saline solution (0.9% sodium chloride obtained from a local drugstore). The resulting formulation has 1% Alugbati aqueous extract and 99% of plain normal saline solution which was placed into a 30 ml bottle spray. As mentioned in the FDA Guidance of Industry for Nasal spray solution (2002) Inhalation solution and suspension drug products are typically aqueous-based formulations that contain therapeutically active ingredients and can also contain additional excipients, since the aqueous Alugbati extract has impurities and excipients it came to a decision wherein 1% of the extract be used for the formulation which also does not exceed the recommended limit concentration based on the GHS classification system when producing Aerosols. Based on the study of Papsin and McTavish (2003), it was imperative that plain normal saline solution (isotonic) must be used in the formulation because using isotonic saline had significant reductions in nasal secretions. Furthermore, in a review of Hermelingmeier *et al.* (2012) on allergic rhinitis cases treated with isotonic saline nasal irrigation from 1994 to 2010, all patients' symptoms improved, and nasal irrigation was able to reduce the dosage and side effects of glucocorticoids, accelerate nasal mucociliary clearance, and improve quality of life, so isotonic saline irrigation was recommended as a complementary therapy of AR.

**Table 4.** Quantity of white blood cells 24 hours post provocation/ administration of treatment.

| Variable | Treatments | Mean | Standard Deviation |
|---|---|---|---|
| Decrease in white blood cells | Formulated (Alugbati leaf aqueous extract) | 8.22 | 4.746 |
| | Fluticasone propionate (Positive Control) | 13.58 | 3.358 |
| | Normal Saline Solution (Negative Control) | 14.72 | 2.801 |

*Evaluation of the Basella alba based nasal spray to ovalbumin-provocated Sprague Dawley rats*

*Organoleptic Properties*

Table 2 shows the organoleptic test results wherein it was stated in the study of Fitriani *et al.* (2020) that the senses which are used to define the qualities of crude pharmaceuticals based on form, smell, color, and taste, are employed to specify organoleptic criteria. According to the results, the formulated Alugbati-based nasal spray exhibited no significant

smell or odorless with a brownish-yellow color. On the other hand, the formulated nasal spray has a watery texture with a transparent opacity.

*Acute Inhalation Toxicity Test*

Table 3 shows the mortality rate of Sprague Dawley rats when exposed to different concentrations of alugbati extract. After 15, 30, 60, 120, and 240 minutes of administration via nose-only nebulization.

There was no recorded mortality Therefore, according to the 2018 OECD guidelines for acute inhalation toxicity the extract is classified as "practically non-toxic".

**Table 5.** ANOVA on the difference of the quantity of WBC count when grouped according treatment.

| Variable | F-value | p-value | Decision |
|---|---|---|---|
| Decreased WBC | 6.496 | 0.006 | Reject null hypothesis |

*Quantity of White Blood Cells*

Table 4 presents the mean and standard deviation of different treatments in decreasing white blood cells. The formulated Alugbati extract has a mean of 8.22 which has a significant difference with the positive control Fluticasone propionate with a mean of 13.58. But there was no significant difference between the positive control and Plain Normal Saline Solution (negative control) with a mean of 14.72. This means that the formulated nasal spray has the potential in normalizing white blood cells 24 hours post provocation/treatment.

**Table 6.** Post-Hoc Analysis on the difference of the quantity of WBC count when grouped according treatment.

| Treatment | Mean | Mean Difference | | |
|---|---|---|---|---|
| | | Formulated | Positive Control | Negative Control |
| Formulated | 8.22 | 0.00 | -5.783* | -6.633* |
| Positive Control | 13.58 | 5.783* | 0.00 | -0.850 |
| Negative Control | 14.72 | 6.663* | 0.850 | 0.00 |

*Significant at 0.05.

Table 5 shows the one-way Analysis of Variance (ANOVA) on the white blood cell counts of the test animals when they are grouped by treatments. This demonstrates that there is a significant difference in decreasing white blood cells among the treatments in the overall comparison of the treatments in normalizing white blood cell levels 24 hours post-administration.

Table 6 displays the post-hoc analysis, LSD test, on the amounts of white blood cells in Sprague-Dawley rats when they are grouped by treatment. The table reveals that there is a significant difference in normalizing white blood cells 24 hours post administration of treatment between the formulated Alugbati Nasal Spray and the positive control Fluticasone Propionate. According to Bamidele *et al.* (2020), there was a significant decrease ($p< 0.05$) in blood glucose and white blood cell count in *Basella alba* extract treated rats when compared to the two groups of his study.

This means that the Alugbati Nasal Spray formulation has a faster response to normalizing WBC, indicating a decrease in inflammation. It was stated in the study of Kumar *et al.* (2011), that by preventing hypotonicity-induced lysis of the erythrocyte membrane, *Basella alba* leaf extracts demonstrated membrane stability. Because erythrocyte membranes are analogous to lysosomal membranes, the extract may also stabilize lysosomal membranes. The stabilization of the lysosomal membrane is critical in limiting the inflammatory response by avoiding the release of activated neutrophil lysosomal contents such as bacterial enzymes and proteases, which induce additional tissue inflammation and damage.

**Table 7.** White blood cell count 24 hours post-provocation-administration.

| Lab No. | Replicates | WBC |
|---|---|---|
| | | 4.0-19.0 x10^9/L |
| RADDLD-215182 | F1S1 | 4 |
| RADDLD-215182 | F1S2 | 7.6 |
| RADDLD-215182 | F1S3 | 2.5 |
| RADDLD-215182 | F2S1 | 52.6 |
| RADDLD-215182 | F3S1 | 14.8 |
| RADDLD-215182 | F3S2 | 7.9 |
| RADDLD-215182 | F3S3 | 12.5 |
| RADDLD-215182 | P1S1 | 17.1 |
| RADDLD-215182 | P1S2 | 19 |
| RADDLD-215182 | P1S3 | 10.2 |
| RADDLD-215182 | P2S1 | 14.7 |
| RADDLD-215182 | P2S2 | 10.3 |
| RADDLD-215182 | P2S3 | 13.2 |
| RADDLD-215182 | P3S1 | 15.3 |
| RADDLD-215182 | P3S2 | 13.5 |
| RADDLD-215182 | P3S3 | 8.9 |
| RADDLD-215182 | N1S1 | 15.9 |
| RADDLD-215182 | N1S2 | 13.9 |
| RADDLD-215182 | N1S3 | 16.2 |
| RADDLD-215182 | N2S1 | 19.4 |
| RADDLD-215182 | N2S2 | 11.9 |
| RADDLD-215182 | N2S3 | 12.1 |
| RADDLD-215182 | N3S1 | 13 |
| RADDLD-215182 | N3S2 | 18.1 |
| RADDLD-215182 | N3S3 | 12 |

**Table 8.** T-test on the difference on the decrease ciliary loss in the nasal mucosa when grouped according treatment.

| Variable | t-value | *p*-value | Decision |
|---|---|---|---|
| Quantity of ciliary loss in the nasal mucosa | 0.707 | 0.496 | Accept null hypothesis |

The data also demonstrates that there is no statistically significant difference between Fluticasone Propionate (positive control) and Plain Normal Saline Solution (negative control).

**Table 9.** T-test on the difference on the quantity of eosinophils infiltration in the nasal mucosa when grouped according treatment.

| Variable | t-value | p-value | Decision |
|---|---|---|---|
| Quantity of eosinophils infiltration in the nasal mucosa | -0.349 | 0.734 | Accept null hypothesis |

This means that both have a comparable response when it comes to normalizing WBC 24 hours post-administration. Furthermore, there is a significant difference in formulation between alugbati nasal spray and the negative control. Table 7 showed the levels of white blood cell count after 24 hours of the last treatment administration. From the different replicates it presents the majority of WBC count within normal values however, the results 52.6, 13.5, and 19.4 were found in the formulated, positive and

negative control, respectively, which indicates an above normal result. Moreover, it can be seen that the one replicate from the formulated group yielded 2.5 which indicates a below-normal count of WBC. According to Sur and Scandale (2010), the onset of therapeutic action for inhaled corticosteroids is 30 minutes, but the peak effect might take several hours to days.

**Table 10.** T-test on the difference on the quantity of Goblet Cells when grouped according treatment.

| Variable | t-value | p-value | Decision |
|---|---|---|---|
| Quantity of Goblet Cells | -0.415 | 0.687 | Accept null hypothesis |

**Table 11.** T-test on the difference on the quantity of vascular congestion when grouped according treatment.

| Variable | t-value | p-value | Decision |
|---|---|---|---|
| Quantity of vascular congestion | 1.265 | 0.235 | Accept null hypothesis |

The decrease in WBC counts in the present work is in contrast to the work done by Bamidele *et al.* (2010) in animals exposed to aqueous extract of *B. alba* in which WBC was increased.

**Fig. 1.** Nasal Spray with Alugbati Extract.

*Quantity of Ciliary loss*

Table 8 represents the test of difference in terms of decreased ciliary loss in nasal mucosa once grouped by treatment. Because *p*-value=0.496 > 0.05 alpha level, there is no significant difference between the formulated Alugbati nasal spray and the positive control Fluticasone propionate. This means that the formulated nasal spray is comparable to that of the positive control in terms of the quantity of ciliary loss. Figure 2 showed no visible ciliary loss on both the Formulated nasal spray and the Positive control compared to the negative control where evident ciliary loss is present. This means that there is a clinical significance present in terms of histopathological observations.

*Quantity of Eosinophil infiltration*

Table 9 shows the test of difference in terms of the decrease of eosinophils infiltration in the nasal mucosa when grouped according to treatment. Since p-value= 0.734 is greater than the alpha level of 0.05 it shows that there is no significant difference between the positive control and the formulated nasal spray.

This implies that the formulated nasal spray is comparable to the positive control in terms of the quantity of eosinophil infiltration in the nasal mucosa.

Figure 3 showed no visible Eosinophil Infiltration on both the Formulated nasal spray and the Positive control compared to the negative control where evident eosinophil migration from the thickened basement membrane is present. This means that there is a clinical significance present in terms of histopathological observations.

*Quantity of Goblet Cells*

Table 10 demonstrates the test of difference in terms of decrease in Goblet cells in the nasal mucosa when grouped by treatment. Since the p-value of 0.687 is more than the alpha level of 0.05, it indicates that there is no significant difference between the positive control and the formulated treatment.

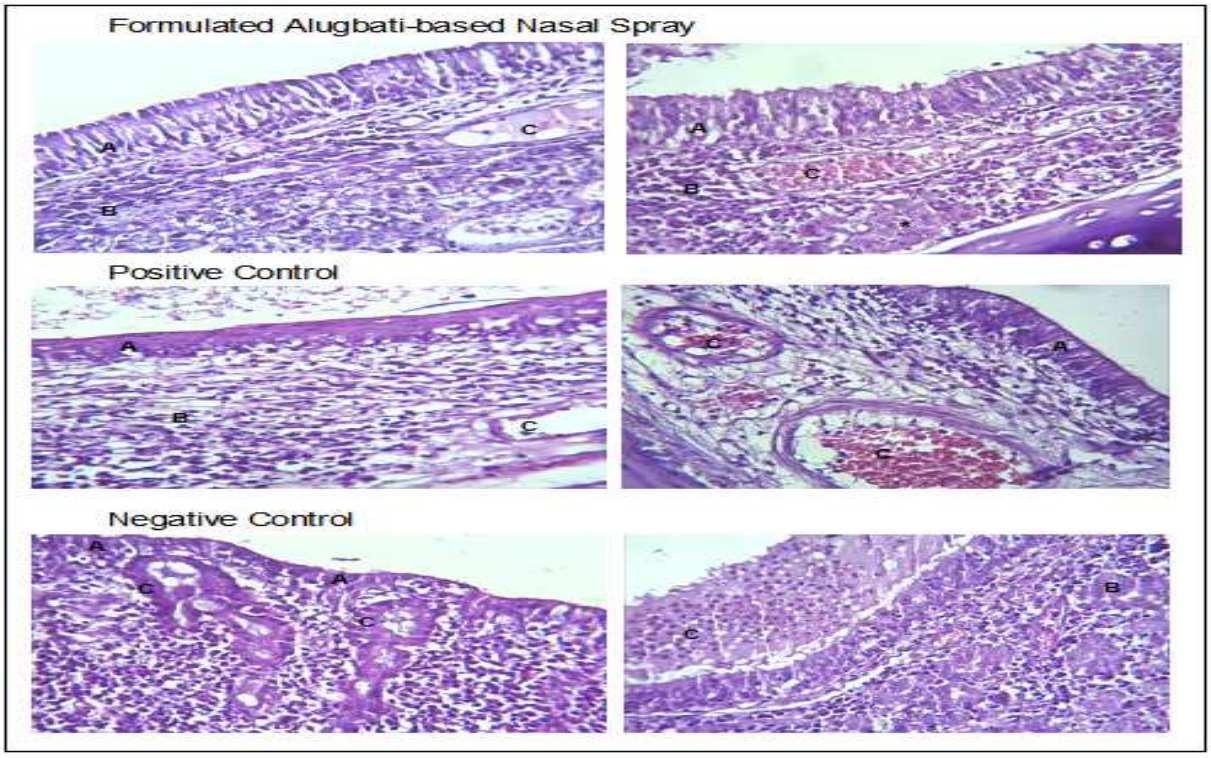


**Fig. 2.** Histolopathogical examination of different samples among treatment when ciliary loss is observed.

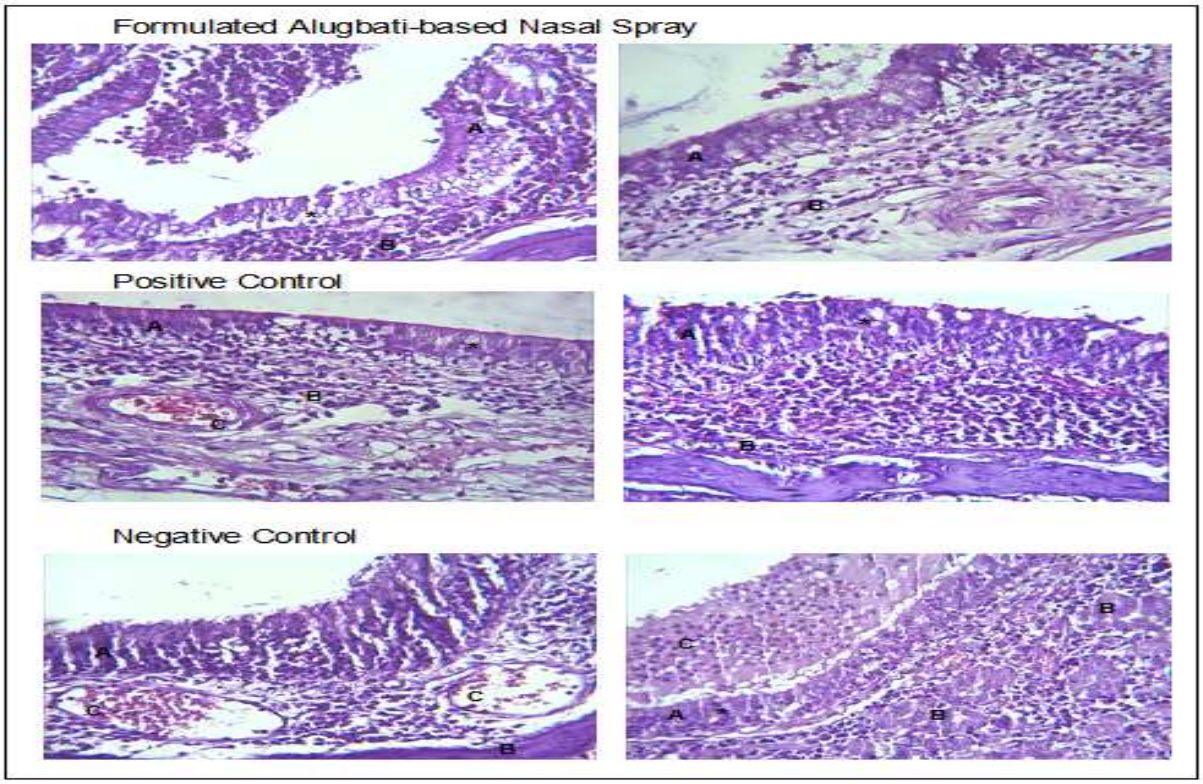


**Fig. 3.** Histopathological examination of different samples among treatment when eosinophil infiltration is observed.

This indicates that the prepared nasal spray is comparable to the positive control in terms of the quantity of Goblet cells in the nasal mucosa.Figure 4 showed some visible presence and alteration of goblet cells on both the Formulated nasal spray and the Positive control compared to the negative control

were in a massive presentation and alteration of goblet cells. This means that there is a clinical significance present in terms of histopathological observations.

*Quantity of Vascular Congestion*

Table 11 presents the test of difference in terms of the decrease in vascular congestion in nasal mucosa when grouped according to treatment. Since p-value=0.235 > α=0.05 it shows that there is no significant difference between the formulated Alugbati nasal spray and positive control Fluticasone propionate.

This implies that the formulated nasal spray is comparable with the positive control in terms of the quantity of vascular congestion in the nasal mucosa.

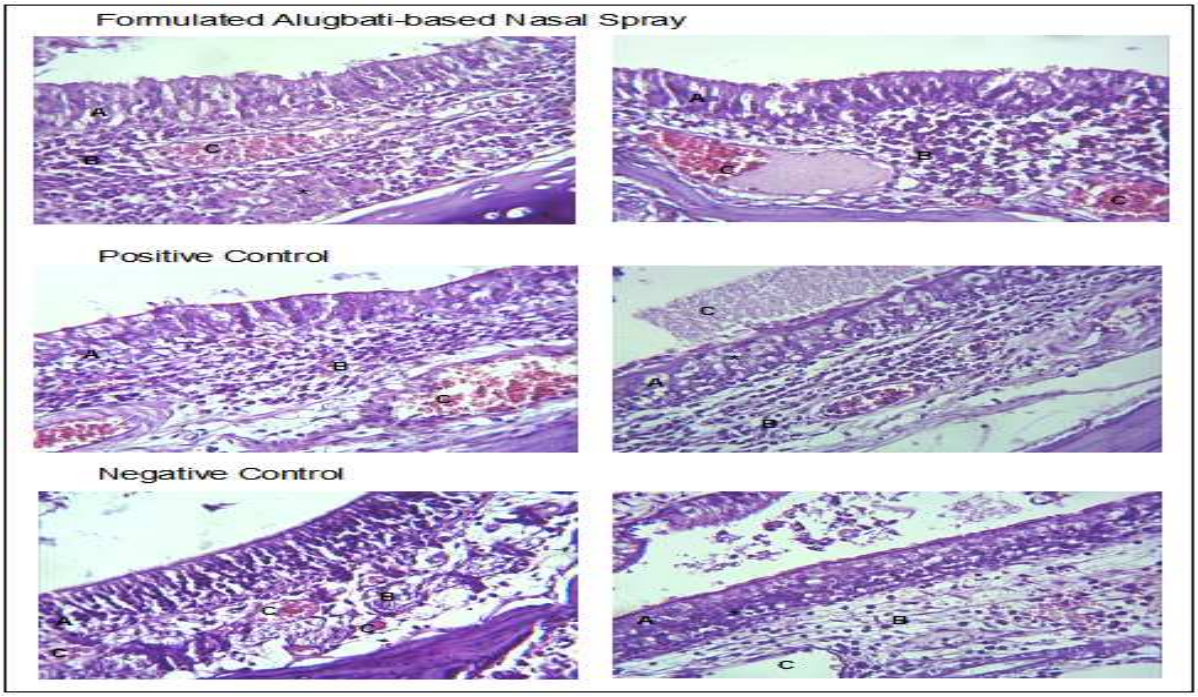


**Fig. 4.** Histopathological examination of different samples among treatment when Goblet cells are observed.

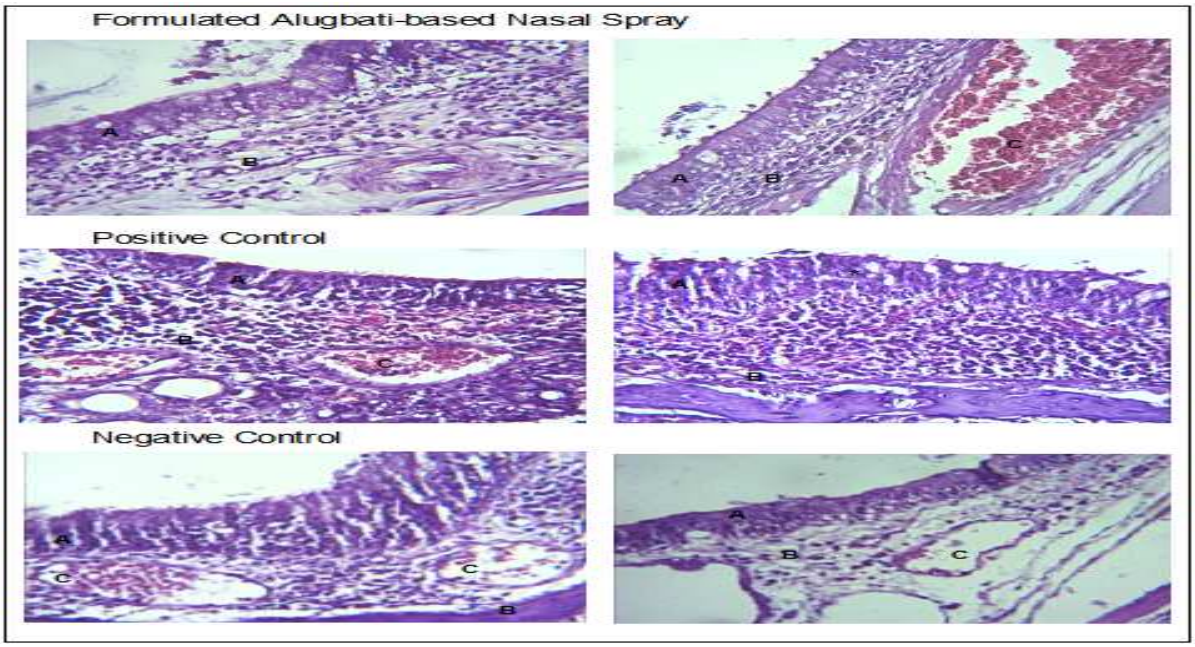


**Fig. 5.** Histopathological examination of different samples among treatment when vascular congestion is observed.

Figure 5 showed visible vascular congestion during all the treatments. Additionally, the negative control had dilation of blood vessels aside from vascular congestion. This means that there is a clinical significance present in terms of histopathological observations.

**Conclusion**

The formulated nasal spray with Alugbati (*Basella alba* L.) aqueous extract is a potential treatment when used in an animal model of experimental allergic rhinitis. It can normalize the number of white blood cells within 24 hours after treatment which indicates an increased response to decreasing inflammation in the nasal passages caused by Allergic rhinitis in Sprague Dawley rats. In the histological parameters, the formulated Alugbati nasal spray has an increased effect in the decrease of eosinophils infiltration, decrease ciliary loss in the nasal mucosa, a decrease of goblet cells, and a decrease in vascular congestion when compared to the positive control statistically. From a clinical perspective, it is evident when observing histopathological samples microscopically there is clinical significance when comparing between treatments. Therefore, formulated Alugbati nasal spray is a potential alternative for treating Allergic Rhinitis.

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