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Anti Ulcerative Effect of Chromolaena Odorata (Siam Weed) on Indomethacin Induced Ulcer in Albino Wistar Rats

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ABSTRACT
The present study was undertaken to evaluate the anti-ulcerative effects of Chromolaena odorata extract on indomethacin-induced gastric ulcer using albino wistar rats. Crude extracts of C. odorata were obtained and reconstituted into doses 200, 400 and 600mg/kg. Albinowistar rats (84) were procured and distributed into seven groups of three control groups and four treatment groups. The control groups were groups; A (normal control), B (standard positive control treated with 0.4mg/kg Omeprazole), and C (untreated negative control group). The treatment groups; D, E, and F were treated with 200, 400, and 600mg/kg C. odorata respectively,while group G was treated with 400mg/kg C. odorata+ 0.4mg/kg omeprazole. All groups except the normal control were induced with 30mg/kg indomethacin before treatment and the study lasted for 14 days before scarifies and biochemical analysis. The results reported group A to have gained the highest weight of 21% while group D gained the least of 6.1%. The highest reduction of 52% in pepsin activity was recorded in group F while the highest decrease of 86.15% was recorded in group E for the mucin content, and a 100% decrease in the ulcer index was recorded for group B. The results show that the aqueous extract of C. odoratapossesses anti-ulcerative potentials and can be utilized in the treatment of gastric ulcer.
Keywords: Chromolaena odorata, indomethacin, ulceration, omeprazole, treatment, induction

I. INTRODUCTION

Peptic ulcer remains one of the gastrointestinal disorders that have affected many people worldwide over the centuries, with over 5-10% of the world population generally affected (Lana et al., 2017). This condition is typified by damage to the endothelial lining of the esophagus, stomach, and/or duodenum caused by the digestive action of pepsin and stomach acid; also, insufficient gastric mucosal resistance may lead to peptic ulcers (Lauret et al., 2005). Some factors that cause peptic ulcers include Helicobacter pylori infection, alcohol consumption, tobacco smoking, psychological stress, some hereditary conditions, and abuse of nonsteroidal anti-inflammatory drugs (NSAIDs) such as aspirin and ibuprofen (de Lira Mota et al., 2005).

NSAIDs can cause damage to the gastroduodenal mucosa via several mechanisms, including the topical irritant effect on the epithelium, impairment of the barrier properties of the mucosa, suppression of gastric prostaglandin synthesis, reduction of gastric mucosal blood flow and interference with the repair of superficial injury (Melcarneet al., 2016).

Ulcer is a prevalent health problem worldwide with a range of contributing factors such as prolonged use of nonsteroidal anti - inflammatory drugs and H. pylori infection. Despite the availability of orthodox treatments, the incidence of ulcer remains high and also, many anti-ulcer drugs such as Omeprazole, Sucralfate, Misoprostol, Antacids, have been reported to have adverse side effects in the body after prolonged consumption. Herbal medicine can be a mighty weapon for suppressing, modulating or eradication of the disease-associated footprints of ulcer and C. odorata has been reported by ethno-medical journals to have antimicrobial properties. This study will therefore try to explore the anti-ulcerative properties of C. odorataand justify its use in the treatment of gastric ulcers.

II. MATERIALS

The equipment's used were; Benchtop pH meter (Hanna HI 9321, Portugal), Weighing Balance (Adam AFP 800L, China), Spectrophotometer (721G, China), Micro

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Hematocrit Centrifuge (HC-120,China), Centrifuge (80-1C, China), Microscope(CECB2000C, China), Incubator (DNP-9052, China)

All the reagents and kit used were of Analytical grade and they include; Randox kit (AST, LFT, UREA, CREATININE), Deionized water, Distilled water, Indomethacin (Zhejiang Dongri Pharmaceutical Co., Ltd, China), Omeprazole (GeneithPharmaceuticalLimited Nigeria), Phenolphthalein, Sodium Chloride, Sodium Chloride, Sucrose, Sodium Acetate, Magnesium Chloride, Normal saline, Diethyl ether, Trichloroacetic Acid (TCA), Chloroform, Alcian dye, Pepsin dye, FolinCiocalteu, Hemoglobin (Sigma -Aldrich USA)

Site of Study

The location of study was at Science Village, Nnamdi Azikiwe University Awka. The biochemical analyses were conducted in Applied Biochemistry Laboratory and the animals were harbored in the Applied Biochemistry animal house at Science Village.

Duration of Study

The study lasted for two weeks (14days) with daily dosage administration of extract and adequate feeding with sufficient water supply for the animal consumption in a conducive environment.

Collection of Sample and Identification

The leaf of Chromolaena odorata was collected at Science Village within the locality of Nnamdi Azikiwe University, Awka, Anambra State. The plant sample collected was taken to the Department of Botany Herbarium Nnamdi Azikiwe University, Awka, where it was identified by MrIroka Chisom, a taxonomist in the Department; a voucher specimen was deposited and a voucher number NAUH-48^{D} was issued to the specimen.

Sample Preparation and Extraction of Chromolaena odorata

The leaves of C. odorata were collected, washed and air dried in Applied Biochemistry Laboratory Sample drying room for (7) days. The dried leaves were ground to fine powdery texture by using an electric blender and stored in a plastic bag. C. odorata (200g) was macerated in one liter of deionized water and allowed to stand for 24hrs. The mixture was sieved using a muslin bag and concentrated using a water bath at 50℃. The crude extract was allowed to cool, weighed and stored. The extract was graded into doses of 200, 400, 600mg/kg and administered to the experimental animals according to their body weight.

Procurement and acclimatization of Animals

Total number of eighty-four wistar albino rats were procured from Dr Chris experimental farm at Mgbakwu, Awka -North, Anambra State,weighing (70-125g).The procured Albino Wistar rats were kept in the Animal house for a period of one week (7 days) in their respective cages for acclimatization.

Ethical Approval

All experimental procedures described were approved by Animal Research Ethics Committee (AREC), Nnamdi Azikiwe University, Awka. The reference number obtained is NAU/AREC/2023/00065.

Ulcer Induction

Gastric ulceration was induced in the animals according to the procedure described by Sayanti et al. (2007). Rats were administered with a single oral dose of indomethacin (30 mg/kg body weight). The wistar albino rats were deprived of food but had free access to water 24 hours prior to ulcer induction except for the animals in normal control group. Various degrees of ulceration manifested 6 hours after indomethacin administration.

Animal Grouping

Eighty-four Wistar albino rats were randomly divided into seven groups with twelve (12) rats each in a group**.** Group A was the normal control group, group B were administered with 0.4mg/kg omeprazole only, group C were induced with 30mg/kg indomethacin but was untreated, group D were treated with 200mg/kg of C. odorata, group E were treated with 400mg/kg of C. odorata, group F were treated with 600mg/kg of C. odorata, group G were treated with 0.4 mg/kg Omeprazole + 400mg/kg of C. odorata

Treatment /Administration of Extract

Following confirmation of ulcer, oral treatment using 5ml sterile syringe commenced and lasted for a period of 14 days. At the end of seven (7) days, four rats were randomly sacrificed in each group for anti-ulcerative study

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Sacrifice and Sample Collection

Animal Sacrifice

For the first week, four (4) rats from each group were sacrificed, and then blood samples and gastric contents were collected for biochemical analysis. After 14 days which is the final week five (6) rats from each group were sacrificed and blood sample, gastric content, stomach, organ were collected for biochemical analysis.

Extraction of Blood Samples

The animals were anesthetized with chloroform and blood samples were collected using sterile 5ml syringe via closed cardiac puncture.

Biochemical Tests

Measurement of Pepsin Activity

The pepsin activity was determined by modification of the method described by Anson et al. (1938) gastric secretion was performed using pepsin standard. Various concentration of bovine pepsin ranging from 0.1-1.0mg/100ml in 0.1N HCl were transferred to a test tube and incubated for 30 minutes using 0. 2.5 % substrate (Hemoglobin) solution in a water bath for 37℃. Then, after 5 minutes 10ml of 5% TCA was added to the test tubes, allowed to stand for 10minutes at room temperature and filtered using What-man filter paper. Blanks were made for the for each concentration by adding 10ml of TCA before the addition of enzyme. Duplicate determination was performed for each sample concentration. The absorbance of the filtrate was measured in a spectrophotometer (721G) at 660nm for the determination of the pepsin activity of gastric secretion. The same procedure were followed at a concentration of 2% 0.1N HCl. A standard curve was concentrated from pepsin content of gastric secretion designed by extrapolation.

Estimation of Mucin Content

The method of Adzuet al. (2015) was used for the estimation of mucin content. The everted stomach were transferred in a test tube, incubated at 20℃ for 24hrs in 0.1% Alcian blue dye 8GX dissolved in 0.16M Sucrose buffered with 0.5M sodium acetate trihydrate. After uncomplexed dye was removed by two successive washes, dye complex with mucus was diluted by immersion in 5ml aliquots of 0.5M Magnesium chloride and was allowed to stand for 2hrs at room temperature. The resulting blue solution were shaken briefly with an equal volume of diethyl ether and was centrifuged at 3000rpm for 15minutes. The lower phase were decanted and the absorbance of the aqueous phase were read in a spectrophotometer(721G) at 598 nm. The stomach of the rat was weighed afterwards. The mucin content of the sample was determined from a standard curve.

Ulcer Index

In this study various scoring system were used to express gastric ulcers and to calculate ulcer indices as described by Risley et al., (1947). A scale prepared on the basis of the ratio between the area of gastric mucosa and the area of ulceration has been used in order to calculate the ulcer indices. The stomachs were opened along the greater curvature, rinsed with distilled and spread on cardboard with the mucous surface upwards avoiding corrugation. Tracing paper was placed over the stomach, the outline of the stomach and the areas of erosions of ulceration were traced on it. This was then superimposed on graph paper having a millimeter scale. The total area of the stomach mucosa and the area of ulcerations were measured. An ulcer index of 1.00 per perforation was added to the index obtained by the above method.

III. RESULT

Weight Profile

The average body weights of the rats orally treated with varying doses of C. odorata extract are expressed in Tables 1a, 1b and 1c.

The result shows a gradual increase in weight across all the groups on day 7 with the highest increase of 16.27g (21%) recorded in group A (normal control) and the lowest increase of 4.34g (6.1%) recorded in group D (200mg/kg of C. odorata). On day 14, significant increases were recorded across the groups when compared to the initial day and day 7 with the highest increase of 35.53g recorded in group A (normal control) and the lowest increase of 8.67g recorded in group D (200mg/kg C. odorata) when compared to the day initial (Day 1).

However, among the treatment groups, group E (400mg/kg C. odorata showed the highest gain in weight of 10.11g (14%) on day 7 when compared to day 1, while the group G (0.4mg/kg Omeprazole + 400 mg/kg C. odorata) displayed the lowest weight gain of 8.12g (12%) on day 7 when compared to the initial day. Groups administered 600 mg/kg C.odorata extract (Group F) gained the highest weight of 18.24g (25.3%) on day 14 when compared to day 1 (initial) while those administered 200mg/kg gained the lowest weight of 9.16g (11.4%) on day 14 when compared to the Day 1 as shown in table 1a and 1c.

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A gross significant increase ($p<0.05$) in the weights of the animals was recorded on day 14 when compared to the initial day as shown in table 1b.

Table 1a: Average weights of all experimental albino wistar rats

Groups	Initial ± SD (g)	Day 7 ± SD (g)	Day 14 ± SD (g)
Group A (Normal control)	77.16 ± 20.42	93.49 ± 20.22	109.76 ± 22.11
Group B (Standard positive control)	71.83 ± 18.72	81.68 ± 19.79	88.2 ± 19.21
Group C (Negative control)	78.91 ± 18.88	86.83 ± 17.87	92.63 ± 16.83
Group D (200mg/kg C. odorata)	71.33 ± 11.01	75.67 ± 8.13	80.49 ± 8.91
Group E (400mg/kg C. odorata)	57.07 ± 6.89	67.17 ± 9.03	72.47 ± 11.74
Group F (600mg/kg C. odorata)	71.93 ± 15.37	81.81 ± 14.82	90.17 ± 13.59
Group G (Omeprazole + C. odorata)	66.70 ± 14.70	74.82 ± 8.84	82.94 ± 10.6

Table 1b: Gross Weight of the Wistar Rats

	Initial	Day 7	Day 14
All Groups	70.7 ± 7.24	80.21 ± 8.61[a]	88.05 ± 11.78[A,b]

[A] Significant when compared to Initial Day, [B] Significant when compared to Day 7, [a] Not significant when compared to Initial Day, [b] Not significant when compared to Day 7

Table 1c: Percentage Increase in Weight when compared to the Initial Day

Groups	Day 7	Day 14
Normal Control	21.12%	42.25%
Positive Control	13.89%	28.96%
Negative Control	10.04%	25.85%
200mg/kg C.odorata	6.08%	25.57%
400mg/kg C. odorata	17.69%	28.68%
600mg/kg C. odorata	13.74%	22.57%
Omeprazole + C. odorata	12.17%	13.88%

%Percentage Decrease
%Percentage Increase

Pepsin Activity Levels of the Experimental Rats

Table 2 shows result of the pepsin concentrations of the experimental animals determined on days 7 and 14 of treatment.

After the first week of treatment, the trend of the result showed a dose-dependent decrease in the pepsin concentration; from the highest value of 0.7±0.10µg/m obtained in group D (200mg/kg C. odorata), to a lower value of 0.48±0.08µg/m obtained in group F (600mg/kg C. odorata)..Groups E (400mg/kg C. odorata), F (600mg/kg C. odorata) and G (omeprazole + 400mg/kg C. odorata) showed a nonsignificant decrease ($p>0.05$) in the pepsin concentration when compared to normal control. The lowest pepsin mean value of 0.41 ± 0.17µg/m was obtained in groups administered with standard drug while the highest value of 0.70 ± 0.10µg/m was obtained for group D (administered 200mg/kg C. odorata).

Furthermore, on day 14, the trend repeated by displaying a dose dependent decrease among the treatment groups from a value as high as 0.74 ± 0.14µg/m obtained for group D (200mg/kg of C. odorata); to the lowest value of 0.21 ± 0.09µg/m obtained in group F (600mg/kg C. odorata). Except for group D, the pepsin levels of the treatment groups were lower when compared with the negative control group. The highest value of 0.84 ± 0.17µg/m was obtained for the normal control group. Among the treatment groups, the highest value of 0.74 ± 0.14µg/m was obtained for the group administered 200mg/kg C. odorata while the lowest value of 0.21 ± 0.09µg/m was obtained for the group administered 600mg/kg C. odorata shown in table 2.

The pepsin concentration of the negative control group showed a 55.32% increase when both weeks of treatment are compared. However, except for group D (200mg/kg C. odorata) and group G; which showed a 5.77% and 30.77% increase respectively, the treatment groups showed reductions in the pepsin levels when both weeks of treatment are compared. The highest increase of 75.6% was recorded for the positive control group while the highest decrease was recorded for group F (600mg/kg C. odorata).

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Table 2: Average Pepsin activityafter 7 and 14days of treatment

Groups	Mean±SD (µg/ml) (WEEK 1)	Mean±SD (µg/ml) (WEEK 2)	Percentage Difference
Group A (Normal control)	0.62 ± 0.18	0.84 ± 0.17	35.48%
Group B (Standard positive control)	0.41 ± 0.17	0.72 ± 0.16	75.60%
Group C (Negative control)	0.47 ± 0.18	0.73 ± 0.19	55.32%
Group D (200mg/kg C. odorata)	0.70 ± 0.10[a,b,d]	0.74 ± 0.14[a,b,d]	5.71%
Group E (400mg/kg C. odorata)	0.55 ± 0.17[a,b,d]	0.37 ± 0.03[a,b,d]	32.73%
Group F (600mg/kg C. odorata)	0.48 ± 0.08[a,b,d]	0.21 ± 0.09[a,b,d]	52.25%
Group G (Omeprazole + C. odorata)	0.52 ± 0.15[a,b,d]	0.68 ± 0.18[a,b,d]	30.77%

Values are mean ± SD of triplicate determination

[A] Significant when compared to Normal control, [B] Significant when compared to Positive control, [D] Significant when compared to Negative control, [a] Not significant when compared to normal control, [b] Not significant when compared to positive control, [d] Not significant when compared to Negative control.
%Percentage Decrease
%Percentage Increase

Mucin Concentration of the Experimental Animals

Table 3 shows the mucin contents of the experimental animals after the first and second week of treatment.

The result obtained on day 7 shows varying decreases in the mucin content among the treatment groups when compared to the negative control group The treatment group also showed non-significant ($p>0.05$) decrease when compared with normal control group. The highest value of 6.17 ± 2.60 was obtained for the negative control while the lowest value of 2.46 ± 0.94 was obtained for groups administered 600mg/kg C. odorata when compared with normal control group. This shows that C. odorata was effective at lowering the mucin content of the animals when compared to the untreated group. Also, the positive control shows that the standard drug lowered the mucin content of the experiment animals when compared to the other control groups.

After the second week of treatment, results show that the mucin contents of the treatment groups were lower when compared to the negative control groups. The highest value of 2.35 ± 0.39 was obtained for group B (Positive control) while the lowest value of 0.73 was obtained for group E (administered 400mg/kg C. odorata). The mucin content of the standard drug was high when compared to the negative and normal control groups. This suggests that continued treatment with the standard drug affected the normal mucin lowering process in the experimental animals.

However, the mucin content in all groups showed a gross reduction on day 14 when compared to day 7 with the highest decrease of 4.54µg/ml recorded in group E (400mg/kg C. odorata). The negative control group recorded a 68.72% reduction in the mucin content when both weeks of treatment are compared. Except for group E, that value is higher than the reductions recorded among the treatment groups after comparing both weeks of treatment. The mucin content of group G showed the lowest reduction of 4.34% when comparing results from the first week to the second week of treatment as shown in table 3.

Table 3: Average values of Mucin Contents after 7 and 14days of treatment

Groups	Mean±SD (µg/ml) (Week 1)	Mean±SD (µg/ml) (Week 2)	Percentage Difference
Group A (Normal control)	5.73 ± 1.92	1.47 ± 0.47	74.35%
Group B (Standard positive control)	4.77 ± 1.91	2.35 ± 0.39	50.73%
Group C (Negative control)	6.17 ± 2.60	1.93 ±0.40	68.72%
Group D (200mg/kg C. odorata)	4.05 ±1.14[a,b,d]	1.42 ±0.56[a,b,d]	64.94%
Group E (400mg/kg C. odorata)	5.27 ± 1.12[a,b,d]	0.73 ±0.20[a,b,d]	86.15%
Group F (600mg/kg C.	2.46 ± 0.94[a,b,d]	1.40 ± 0.78[a,b,d]	43.09%

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odorata)			
Group G (Omeprazole + C. odorata)	4.40 ± 0.79[a,b,d]	0.89 ± 0.17[a,b,d]	4.34%

Values are mean ± SD of triplicate determination

[A] Significant when compared to Normal control, [B] Significant when compared to Positive control, [D] Significant when compared to Negative control, [a] Not significant when compared to normal control, [b] Not significant when compared to positive control, [d] Not significant when compared to Negative control
%Percentage Decrease
%Percentage Increase

Ulcer Index Measurement

The mean values of the ulcer index of the experimental animals obtained on days 7 and 14 are expressed in tables 7a.

On day 7, as expected the normal control group exhibited an ulcer index of 0.0 making it the lowest value among the experimental animals. As a result, all the test groups showed a significant elevation ($p<0.05$) in their ulcer index value when compared to the normal control. However, the highest value of 58.53 ± 10.11was obtained for the negative control group, then subsequent reductions were recorded in the treatment groups when compared to negative control. Also the results show that all the test groups have mean values that are significantly higher than the mean values of the positive control after the first week of treatment.

However, on day 14, it becomes evident among the test groups that there is a significant ($p<0.05$) reduction in the mean values of the treatment groups when compared to the negative control. The highest value of 43.65 ± 2.25 was recorded for the negative control group while the lowest value of 4.10 ± 0.25 was recorded among the test group for group D (200mg/kg C. odorata) as shown in table 7a and 7b.

The result showed the highest reduction of 100% reported for the positive control group when comparing the results from the first week and the second week. The rate of decrease in the ulcer index was markedly higher in the treatment groups than it is in the negative control group between the first and second weeks of treatment. However, a 25% decrease in the ulcer index of the negative control was recorded on day 14 when compared to day 7.

Table 7a: Average values of the ulcer index after 7 and 14days of treatment.

Groups	**Mean** ± SD **Week 1**	**Mean** ± SD **Week 2**	**Percentage Increase**
Group A (Normal control)	0.00 ± 0.00	0.00 ± 0.00	-
Group B (Standard positive control)	18.72 ± 1.16	0.00 ± 0.00	100%
Group C (Negative control)	58.53 ± 10.11	43.65 ± 2.25	25.42%
Group D (200mg/kg C. odorata)	45.07 ± 10.11[A,B,d]	4.10 ± 0.25[a,b,D]	90.90%
Group E (400mg/kg C. odorata)	48.23 ± 6.75[A,B,d]	8.61 ± 1.16[a,b,D]	82.15%
Group F (600mg/kg C. odorata)	46.69 ± 11.19[A,B,d]	7.30 ± 4.18[a,b,D]	84.36%
Group G (Omeprazole + C. odorata)	50.38 ± 3.39[A,B,d]	10.51 ± 1.16[a,b,D]	79.14%

Values are mean ± SD of triplicate determination

[A] Significant when compared to Normal control, [B] Significant when compared to Positive control, [D] Significant when compared to Negative control, [a] not significant when compared to normal control, [b] not significant when compared to positive control, [d] Not significant when compared to Negative control
%Percentage Decrease
%Percentage Increase

IV. DISCUSSION

Ulcer is a recurrent disease affecting large populations in all geographical regions in Nigeria. Peptic ulcers result from; an imbalance between factors that damage the mucosa and the normal mucosal defense and repair mechanisms; factors associated with Helicobacter pylori infection; and increased use of non-steroidal anti-inflammatory drugs like aspirin and indomethacin (Corne et al.,

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2006). These cause damage by inhibiting the biosynthesis of cytoprotective prostaglandins (Rainsford, 2010).

The results of the study showed that indomethacin (30mg/kg) was effective in the induction of ulcer in the experimental rats. Indomethacin is a Non-SteroidalAnti Inflammatory drug (NSAID) which has been implicated as one of the causes of ulcer and exacerbation of ulceration (Munial et al., 2023).

The weight profile of the animals showed significant increases ($p<0.05$) across all groups, after 7 and 14days of treatment. However, it was noticed that the normal control group showed the highest weight gain of 32.6g (21.2%) while the treatment groups showed a significantly lower gain in weight when compared with the normal control group. Since the gain in weight of the untreated group was found to be significantly reduced when compared to the normal control after 14 days of administration, it becomes evident in the result that ulcer; via induction with indomethacin, reduced the gain in weight among the experimental animals. This means that ulceration has a negative effect on the weight of the experimental rats. This agrees with Woolf et al., (2023) who reported weight loss as a major symptom of ulcer among other symptoms like nausea, heartburn, and stomach pain. Moreover, the reduction in weight gain among most of the treatment groups after the second week of treatment when compared to the negative control was also manifest in the result. This result agrees with Mori et al., (2018) who reported weight loss as a consequence of improvement in ulcer treatment. The result also summarily shows that the treatment with extract significantly reduced the rate of weight gain among the experimental animals, which were fed and given water until the day of sacrifice. This result agrees with the suggestions of Herreeaet al. (2014) who reports that C. odorata extract reduced the weight of experimental animals significantly when compared to the control group; and also the result agrees with the observations of Mori et al. (2018) who reports; as stated above, weight loss as a consequence of improvement in ulcer treatment.

The results of the pepsin activity showed a dose dependent decrease was evident in the result for pepsin activity across the test groups from a value as high as 0.74 ± 0.14µg/ml obtained for the least dose of 200mg/kg; to a lower value of 0.21 ± 0.09µg/ml obtained for the highest dose of 600mg/kg on day14. This is so because for every increase in dose, there was a corresponding decrease in the pepsin activity. According to Malik et al. (2023), pepsin or gastric acid secretion characterizes peptic ulcer disease; therefore, it can be suggested that the dose-dependent decrease noticed among the test groups from 0.74 ± 0.74µg/ml (200mg/kg) to 0.21±0.8µg/ml (600mg/kg) may prove the potency of anti-ulcerative effects of C. odorata. This finding is in line with the findings of Ogundajor et al. (2014) who reported that C. odorata extract inhibits pepsin activity in dose dependent manner. Increase in pepsin activity can exacerbate damage to the gastric mucosal since pepsin can penetrate the epithelium and cause tissue damage, resulting to ulcer formation (Rao et al., 2016). Also, the pepsin concentration of the negative control group showed a 55.32% increase when both weeks of treatment are compared. However, except for group D (200mg/kg C. odorata) and group G; which showed a 5.77% and 30.77% increase respectively, the treatment groups showed reductions in the pepsin levels when both weeks of treatment are compared. Since increase in the pepsin concentration causes and exacerbates the damage to the gastric mucosal (Rao et al., 2016), it can be said that the C. odorata has a positive effect in the treatment of ulcer since it reduced the activity of pepsin.

The result of mucin content after 7 days of treatment showed; the negative control to have the highest value of 6.17 ± 2.60µg/ml, then a dose dependent decrease in the test groups with the lowest value of 2.46 ± 0.94ug/ml recorded in groups administered 600mg/kg. Decreases were also recorded in week 2 (day 14) when compared to the negative control. This result is in agreement with the observations of Yamakoet al. (2004), who reported that an increase in mucin content is related to the inflammatory response associated with ulcer formation. Also, according to Kesimeret al. (2013), inflammation (by inflammatory cytokines) and infection (by bacterial or viral pathogens) are amongst the several factors that can cause increase in the mucin content. After one week of treatment, it was observed that mucin content of the negative control was the highest, this might be due to the inflammatory response as a result of the untreated ulcers on the gastric mucosal of the negative control groups (Johansson and Hansson, 2016). Therefore, the rise noticed in the negative control, and the subsequent dose-dependent reductions reported in the treatment groups show that after induction of ulceration using indomethacin, the reductions in the mucin content seen in the treatment groups affirms that treatment with C. odorata is effective. Even, it can be suggested that at higher doses, C. odorata is more effective at

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reducing the mucin content than the standard drug, omeprazole. However, is prudent to note that gastric mucus (mucin) is an important protective factor for the gastric mucosa and consist of a viscous elastic, adherent and transparent gel formed by 95% water and 5% glycoproteins that cover the entire gastrointestinal mucosa (Johansson et al., 2013). Also, the mucin content in the treatment groups showed a gross reduction on day 14 when compared to day 7 with the highest decrease of 4.54µg/ml (86.15%) recorded in group E (400mg/kg C. odorata). This suggests that C. odorata is effective in the treatment of gastric ulcer having been implicated in the reduction of the mucin content of the experimental animals (Kesimeret al., 2013).

The result of the ulcer index shows that the negative control reported the highest value after the first and second weeks of treatment. The ulcer index of the negative control was significantly ($p<0.05$) higher when compared to the treatment groups after the second week of treatment. The ulcer index among the test groups reduced by; approximately over 87%, on day 14 when compared to day 7. This shows that the extract of C. odorata is effective even in low doses of 200mg/kg. The result reports a 100% reduction reported for the positive control group after the two weeks of treatment. The decrease in the ulcer index was markedly higher in the treatment groups than in the negative control group when the results between the first and second weeks of treatment are compared. However, a 25.42% decrease in the ulcer index of the negative control was recorded on day 14 when compared to day 7. The report agrees with Olaleye et al. (2012) where he noted that C. odorata showed significant anti-ulcer activity by reducing the ulcer index and increasing healing rate. Afolabi et al. (2007) attributes this ability of reducing ulceration to the phytometabolite constituents inherent in the C. odorata. Significant increase in ulcer index following oral administration of indomethacin in the negative control group may be attributed to either free radical's formation or inhibition of prostaglandins synthesis (Sabiu et al., 2015).

V. CONCLUSION

The result obtained from this study shows that the aqueous extract of Chromolaena odoratapossesses anti-ulcerative potentials.C.odorataleaf is safe for consumption at a low doses ≤400mg/kg and might elicit toxic effect when taken at dosesup to 600mg/kg or more.The use of C. odorata as a herbal medicine would provide good health to human life considering its effective wound healing property; using multiple mechanism to inhibit inflammatory mediators.

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