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# Anti-inflammatory, analgesic and antipyretic activities of the aqueous leaf extract of *Kalanchoe crenata* (Andrews) Haw. traditionally used for ear disorders in Côte d'Ivoire

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**ABSTRACT**

Ear disorders, particularly otitis, remain important public health problems, and traditional medicine contributes to their management in many African countries. In Côte d'Ivoire, the leaves of *Kalanchoe crenata* (Andrews) Haw. are traditionally used for ear disorders. This study evaluated the anti-inflammatory, analgesic and antipyretic activities of its aqueous leaf extract (EAKc) using complementary experimental models. Anti-inflammatory activity was assessed using kaolin- and fresh egg albumin-induced paw edema in mice, analgesic activity using the hot-water tail immersion test, and antipyretic activity using brewer's yeast-induced pyrexia in rats. EAKc reduced paw edema in both models, with the strongest effect generally observed at 500 mg/kg. At 5 h, paw thickness was 2.27 ± 0.31 mm in the kaolin model and 1.80 ± 0.02 mm in the egg albumin model. At 500 mg/kg, tail-withdrawal latency reached 6.28 ± 0.18 s at 4 h, compared with 6.80 ± 0.06 s for aspirin. The strongest antipyretic effect was observed at 250 mg/kg, with a temperature of 35.80 ± 0.10°C at 5 h, compared with 37.30 ± 0.00°C in controls and 36.45 ± 0.05°C for paracetamol. Overall, the findings extend previous pharmacological evidence for *K. crenata* by integrating anti-inflammatory, analgesic and antipyretic activities of its aqueous leaf extract. These findings support its traditional symptomatic use in ear disorders, without establishing efficacy directly against otitis or its infectious causes. Further phytochemical, safety, mechanistic and antimicrobial studies are needed.

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## INTRODUCTION

Ear disorders, particularly otitis, remain an important public health concern worldwide. The global burden of otitis media has increased substantially over recent decades, with the estimated number of incident cases rising from 322.1 million in 1992 to 391.3 million in 2021 (Huang *et al.*, 2025). In many African countries, traditional medicine continues to contribute to primary healthcare, particularly for the management of inflammatory and painful conditions. In Côte d'Ivoire, ethnobotanical studies have documented the use of several medicinal plants for the treatment of ear, nose and throat disorders (Kouadio *et al.*, 2014; Béné *et al.*, 2016).

*Kalanchoe crenata* (Andrews) Haw. (Crassulaceae) is a medicinal plant traditionally used in Côte d'Ivoire and other African countries for the management of ear disorders, including earache and otitis. Previous phytochemical investigations have reported the presence of several classes of secondary metabolites in *K. crenata*, including flavonoids, phenolic compounds, tannins, alkaloids, saponins and terpenoids, which may contribute to its pharmacological properties (Dimo *et al.*, 2006; Assis de Andrade *et al.*, 2023). Pharmacological studies have already demonstrated analgesic and anti-inflammatory activities of *K. crenata* using different extracts, fractions and experimental models (Nguelefack *et al.*, 2004, 2006; Dimo *et al.,* 2006). These findings provide experimental support for some of the traditional uses of the species.

However, the available pharmacological evidence remains fragmented, with previous investigations focusing mainly on individual activities and using different types of extracts and experimental approaches. In particular, the pharmacological profile of the aqueous leaf extract has not been comprehensively examined through the simultaneous evaluation of inflammation, pain and fever. The present study therefore extends previous investigations by assessing the aqueous leaf extract of *K. crenata* in an integrated experimental approach combining two acute inflammation models, a thermal nociception model and a brewer's yeast-induced pyrexia model.

This complementary approach was designed to provide broader pharmacological evidence for the traditional use of the plant in the management of ear disorders, while recognizing that the present study does not directly evaluate otitis or its infectious causes.

The aim of this study was therefore to evaluate the anti-inflammatory, analgesic and antipyretic activities of the aqueous leaf extract of *K. crenata* using kaolin- and fresh egg albumin-induced paw edema, the hot-water tail immersion test, and brewer's yeast-induced pyrexia, respectively.

## MATERIALS AND METHODS

### Plant material and identification

The leaves of *Kalanchoe crenata* (Andrews) Haw. were collected in Man, western Côte d'Ivoire. The plant material was identified and authenticated by a botanist, and a voucher specimen (UCJ018569) was deposited in the herbarium for future reference. Fresh leaves were used for the preparation of the aqueous extract.

### Preparation of the aqueous extract

The leaves were washed, cut into small pieces, and dried at 18 °C for one week. The dried material was ground into powder. Fifty grams (50 g) of the powder were macerated in 500 mL of distilled water preheated to 50 °C. The mixture was mechanically agitated using a blender for 15 min and then allowed to stand for 2 min.

This extraction procedure was repeated three times to improve the recovery of extractable constituents. The resulting macerate was filtered under vacuum through hydrophilic cotton and Whatman No. 1 filter paper. The filtrate was subsequently dried in an oven at 45 °C for 24 h until a dry extract was obtained. The extraction yield was calculated according to the method of Harborne (1998):

$$\text{Yield (\%)} = (\text{mass of dry extract}/\text{mass of plant powder}) \times 100 \quad (1)$$

The dried aqueous extract was stored in an airtight container until use for the pharmacological assays.

**Experimental animals**

Two experimental animal models were used. Anti-inflammatory and analgesic activities were evaluated in male and female albino mice (*Mus musculus*, Muridae), aged 8-12 weeks and weighing 15-25 g, while antipyretic activity was evaluated in male and female Wistar rats (*Rattus norvegicus*), weighing 150-200 g. The animals were acclimatized for one week under standard laboratory conditions and had free access to standard feed and water ad libitum. The experimental protocols were conducted in accordance with the relevant European Union guidelines for the care and use of animals for scientific purposes (European Union, 2010).

**Evaluation of anti-inflammatory activity**

Anti-inflammatory activity was evaluated using two acute paw edema models induced by kaolin and fresh egg albumin. The kaolin-induced paw edema model was performed according to the method described by Wagner-Jauregg *et al.* (1962), with modifications, while the fresh egg albumin-induced paw edema model was performed according to the method described by Kokon *et al.* (2016), with modifications. For each model, thirty mice were randomly divided into six groups of five animals each (n = 5 per group):

1. Group 1 received 0.9% NaCl and served as the negative control;
2. Group 2 received aspirin (100 mg/kg) as the reference drug;
3. Groups 3-6 received the aqueous leaf extract of *K. crenata* (EAKc) at doses of 62.5, 125, 250, and 500 mg/kg, respectively.

Inflammation was induced by subplantar injection of 0.1 mL of either a 20% kaolin suspension or fresh egg albumin, one hour before treatment. Paw thickness was measured before induction to establish the baseline and then at 1, 2, 3, 4, and 5 h after treatment using a digital micrometer.

The percentage reduction in paw edema was calculated using the following formula described by Muhammad *et al.* (2013):

$$\text{Percentage reduction (\%)} = [(A - B) / A] \times 100 \quad (2)$$

Where, A = mean paw thickness of the edema control group at the corresponding observation time and B = mean paw thickness of the treated group at the corresponding observation time (1, 2, 3, 4, and 5 h).

**Evaluation of analgesic activity**

Analgesic activity was evaluated using the hot-water tail immersion test according to the method described by Aydin *et al.* (1999). Thirty mice were divided into six groups of five animals each:

Group 1: negative control, receiving 0.9% NaCl control;
Group 2: aspirin, 100 mg/kg;
Groups 3-6: aqueous extract of *K. crenata* at 62.5, 125, 250 and 500 mg/kg, respectively.

After habituation to the experimental procedure, an initial tail-withdrawal latency ($T_i1$) was recorded. A second latency measurement ($T_i2$) was recorded immediately before treatment. One hour after oral administration, the distal portion of each mouse's tail was immersed in a water bath maintained at 55 ± 1 °C. Tail-withdrawal latency was then recorded at 30 min, 1, 2, 3, and 4 h after treatment. The percentage increase in latency time was calculated according to the method described by Yimer *et al.* (2020):

$$\text{Increase in latency (\%)} = [(TL_{treated} - TL_{control}) / TL_{control}] \times 100$$

where $TL_{treated}$ is the tail-withdrawal latency of the treated group and $TL_{control}$ is that of the negative control group at the corresponding observation time.

**Evaluation of antipyretic activity**

Antipyretic activity was evaluated using the brewer's yeast-induced pyrexia model in Wistar rats. Pyrexia was induced by subcutaneous injection of a 20% brewer's yeast suspension at a dose of 1 mL/100 g body weight. After induction, only animals showing an increase in rectal temperature of at least 0.5 °C were included in the experiment. The animals were randomly divided into six groups of five rats each:

1. Group 1 received saline (10 mL/kg) and served as the negative control ;
2. Groups 2–5 received the aqueous leaf extract of *K. crenata* at doses of 62.5, 125, 250, and 500 mg/kg, respectively ;
3. Group 6 received paracetamol (100 mg/kg) as the reference drug.

Rectal temperature was recorded at baseline and after pyrexia induction, and then hourly for 5 h after treatment.

### Statistical analysis

Data were expressed as mean ± SEM. Statistical comparisons among groups were performed using one-way analysis of variance (ANOVA), followed by Tukey's multiple-comparison test. Differences were considered statistically significant at $p < 0.05$. Statistical analyses were performed using GraphPad Prism version 8.0.

## RESULTS

### Extraction yield

The aqueous extraction of *Kalanchoe crenata* leaves yielded 22.12%, corresponding to 11.06 g of dry extract obtained from 50 g of powdered leaves.

### Anti-inflammatory activity against kaolin-induced paw edema

Subplantar injection of 20% kaolin induced a marked increase in paw thickness in the control group. Treatment with the aqueous leaf extract of *K. crenata* (EAKc) reduced paw thickness at most observation times, with the strongest effect generally observed at 500 mg/kg. At 5 h, paw thickness was 2.27 ± 0.31 mm in mice treated with 500 mg/kg EAKc, corresponding to a 21.99% reduction compared with the control group. Aspirin produced a paw thickness of 2.11 ± 0.03 mm, corresponding to a 27.49% reduction compared with the control group (Table 1).

**Table 1.** Effect of the aqueous leaf extract of *Kalanchoe crenata* on kaolin-induced paw edema in mice

| Treatment and dose (mg/kg BW) | Mean paw thickness before 20% kaolin injection (mm) | Mean paw thickness immediately after 20% kaolin injection (mm) | 1 h (T1) | 2 h (T2) | 3 h (T3) | 4 h (T4) | 5 h (T5) |
|---|---|---|---|---|---|---|---|
| 0.9% NaCl control | 1.66 ± 0.06$^{a}$ | 3.04 ± 0.21$^{b}$ | 3.01 ± 0.34$^{b}$ | 3.01 ± 0.34$^{b}$ | 2.96 ± 0.20$^{c}$ | 2.94 ± 0.23$^{c}$ | 2.91 ± 0.21$^{c}$ |
| Aspirin (100) | 1.74 ± 0.02$^{a}$ | 3.12 ± 0.02$^{b}$ | 2.85 ± 0.29$^{ab}$ (5.32) | 2.40 ± 0.51$^{ab}$ (20.26) | 2.28 ± 0.14$^{c}$ (22.97) | 2.15 ± 0.23$^{c}$ (26.87) | 2.11 ± 0.03$^{c}$ (27.49) |
| EAKc (62.5) | 1.56 ± 0.10$^{a}$ | 3.36 ± 0.02$^{b}$ | 3.13 ± 0.38$^{b}$ (-4.00) | 2.96 ± 0.20$^{ab}$ (1.66) | 2.81 ± 0.40$^{c}$ (5.06) | 2.71 ± 0.15$^{c}$ (7.82) | 2.67 ± 0.27$^{c}$ (8.25) |
| EAKc (125) | 1.66 ± 0.02$^{a}$ | 3.34 ± 0.04$^{b}$ | 3.01 ± 0.39$^{c}$ (0.00) | 2.93 ± 0.04$^{c}$ (2.65) | 2.81 ± 0.17$^{bc}$ (5.06) | 2.76 ± 0.35$^{c}$ (6.12) | 2.66 ± 0.31$^{ab}$ (8.59) |
| EAKc (250) | 1.76 ± 0.40$^{a}$ | 3.18 ± 0.05$^{b}$ | 3.00 ± 0.40$^{c}$ (0.33) | 2.83 ± 0.40$^{c}$ (5.98) | 2.76 ± 0.44$^{ab}$ (6.76) | 2.70 ± 0.34$^{ab}$ (8.16) | 2.60 ± 0.15$^{c}$ (10.65) |
| EAKc (500) | 1.74 ± 0.04$^{a}$ | 3.32 ± 0.04$^{b}$ | 2.91 ± 0.21$^{d}$ (3.32) | 2.76 ± 0.08$^{c}$ (8.31) | 2.61 ± 0.33$^{c}$ (11.82) | 2.57 ± 0.12$^{c}$ (12.58) | 2.27 ± 0.31$^{c}$ (21.99) |

EAKc: aqueous leaf extract of Kalanchoe crenata; BW: body weight; n = 5. Values are expressed as mean ± SEM. Values in parentheses represent the percentage reduction in paw thickness relative to the 0.9% NaCl control group at the corresponding time point. Within each column, means followed by different superscript letters are significantly different, whereas means sharing at least one superscript letter are not significantly different according to Tukey's multiple-comparison test ($p < 0.05$).

**Table 2.** Effect of the aqueous leaf extract of *Kalanchoe crenata* on fresh egg albumin-induced paw edema in mice

| Treatment and dose (mg/kg BW) | Mean paw thickness before egg albumin injection (mm) | Mean paw thickness after egg albumin injection (mm) | 1 h | 2 h | 3 h | 4 h | 5 h |
|---|---|---|---|---|---|---|---|
| 0.9% NaCl control | 1.44 ± 0.04$^{a}$ | 2.47 ± 0.03$^{b}$ | 2.40 ± 0.03$^{b}$ | 2.39 ± 0.02$^{b}$ | 2.30 ± 0.09$^{c}$ | 2.28 ± 0.02$^{c}$ | 2.10 ± 0.03$^{d}$ |
| Aspirin (100) | 1.38 ± 0.03$^{a}$ | 2.52 ± 0.05$^{b}$ | 2.28 ± 0.03$^{b}$ (5.00) | 2.06 ± 0.05$^{c}$ (13.81) | 2.06 ± 0.05$^{c}$ (10.43) | 1.64 ± 0.41$^{d}$ (28.07) | 1.26 ± 0.31$^{e}$ (40.00) |
| EAKc (62.5) | 1.30 ± 0.04$^{a}$ | 2.76 ± 0.02$^{b}$ | 2.60 ± 0.02$^{b}$ (-8.33) | 2.53 ± 0.06$^{b}$ (-5.86) | 2.30 ± 0.05$^{ab}$ (0.00) | 2.15 ± 0.04$^{b}$ (5.70) | 2.00 ± 0.05$^{c}$ (4.76) |
| EAKc (125) | 1.42 ± 0.03$^{a}$ | 2.60 ± 0.02$^{b}$ | 2.50 ± 0.06$^{b}$ (-4.17) | 2.30 ± 0.09$^{c}$ (3.76) | 2.20 ± 0.03$^{c}$ (4.34) | 2.10 ± 0.04$^{c}$ (7.89) | 1.97 ± 0.03$^{d}$ (6.19) |
| EAKc (250) | 1.42 ± 0.03$^{a}$ | 2.71 ± 0.02$^{b}$ | 2.36 ± 0.02$^{b}$ (1.66) | 2.25 ± 0.02$^{c}$ (5.86) | 2.18 ± 0.04$^{c}$ (5.21) | 2.00 ± 0.03$^{cb}$ (12.28) | 1.90 ± 0.02$^{d}$ (9.52) |
| EAKc (500) | 1.38 ± 0.03$^{a}$ | 2.42 ± 0.21$^{b}$ | 2.24 ± 0.04$^{ab}$ (6.67) | 2.19 ± 0.05$^{ab}$ (8.37) | 2.16 ± 0.04$^{ab}$ (6.08) | 1.90 ± 0.02$^{c}$ (16.66) | 1.80 ± 0.02$^{c}$ (14.29) |

EAKc: aqueous leaf extract of *Kalanchoe crenata*; BW: body weight; n = 5. Values are expressed as mean ± SEM. Values in parentheses represent the percentage reduction in paw thickness relative to the 0.9% NaCl control group at the corresponding time point. Negative values indicate an increase in paw thickness relative to the control group. Within each column, means followed by different superscript letters are significantly different, whereas means sharing at least one superscript letter are not significantly different according to Tukey's multiple-comparison test ($p < 0.05$).

**Table 3.** Analgesic effect of the aqueous leaf extract of *Kalanchoe crenata* on hot-water tail immersion-induced nociception in mice

| Treatment and dose (mg/kg BW) | Habituation latency ($T_1$) (s) | Pre-treatment latency ($T_2$) (s) | 30 min | 1 h | 2 h | 3 h | 4 h |
|---|---|---|---|---|---|---|---|
| 0.9% NaCl control | 2.02 ± 0.002$^a$ | 2.05 ± 0.009$^a$ | 1.041 ± 0.012$^b$ | 1.064 ± 0.01$^b$ | 2.06 ± 0.01$^a$ | 2.15 ± 0.15$^a$ | 2.90 ± 0.03$^a$ |
| Aspirin (100) | 2.00 ± 0.006$^a$ | 2.022 ± 0.009$^a$ | 1.089 ± 0.002$^b$ (4.61) | 1.12 ± 0.02$^b$ (5.26) | 2.95 ± 0.019$^a$ (43.20) | 4.48 ± 0.11$^d$ (108.37) | 6.80 ± 0.06$^e$ (134.48) |
| EAKc (62.5) | 1.81 ± 0.07$^a$ | 1.75 ± 0.14$^a$ | 1.24 ± 0.03$^b$ (19.12) | 1.68 ± 0.12$^b$ (57.89) | 1.96 ± 0.02$^a$ (-4.85) | 2.14 ± 0.05$^a$ (-0.47) | 2.76 ± 0.10$^a$ (-4.83) |
| EAKc (125) | 2.03 ± 0.015$^a$ | 2.033 ± 0.008$^a$ | 1.131 ± 0.01$^b$ (8.65) | 1.80 ± 0.10$^b$ (69.17) | 1.90 ± 0.02$^a$ (-7.77) | 2.10 ± 0.05$^a$ (-2.33) | 2.70 ± 0.10$^a$ (-6.90) |
| EAKc (250) | 2.00 ± 0.01$^a$ | 2.072 ± 0.05$^a$ | 2.14 ± 0.01$^b$ (105.57) | 2.25 ± 0.02$^b$ (111.47) | 2.38 ± 0.01$^b$ (15.53) | 2.50 ± 0.01$^{ab}$ (16.28) | 2.81 ± 0.04$^a$ (-3.10) |
| EAKc (500) | 2.01 ± 0.005$^a$ | 2.038 ± 0.008$^a$ | 2.50 ± 0.002$^{ab}$ (140.15) | 3.34 ± 0.02$^b$ (213.91) | 4.60 ± 0.10$^c$ (123.30) | 6.19 ± 0.19$^d$ (187.91) | 6.28 ± 0.18$^e$ (116.55) |

EAKc: aqueous leaf extract of *Kalanchoe crenata*; BW: body weight; n = 5. Values are expressed as mean ± SEM. Values in parentheses represent the percentage increase or decrease in tail-withdrawal latency relative to the 0.9% NaCl control group at the corresponding time point. Positive values indicate an increase in latency, whereas negative values indicate a decrease in latency relative to the control group. Within each column, means sharing the same superscript letter are not significantly different, whereas means with different superscript letters are significantly different according to Tukey's multiple-comparison test ($p < 0.05$).

**Table 4.** Antipyretic effect of the aqueous leaf extract of *Kalanchoe crenata* on brewer's yeast-induced pyrexia in Wistar rats

| Treatment and dose (mg/kg BW) | Mean normal rectal temperature (°C) | Mean rectal temperature after brewer's yeast injection (°C) | 1 h | 2 h | 3 h | 4 h | 5 h |
|---|---|---|---|---|---|---|---|
| 0.9% NaCl control | 37.17 ± 0.09$^a$ | 37.77 ± 0.03$^a$ | 37.63 ± 0.03$^a$ | 37.50 ± 0.10$^a$ | 37.47 ± 0.07$^a$ | 37.37 ± 0.03$^a$ | 37.30 ± 0.00$^a$ |
| EAKc (62.5) | 37.70 ± 0.61$^a$ | 37.43 ± 0.42$^a$ | 37.53 ± 0.61$^a$ | 37.33 ± 0.30$^a$ | 37.10 ± 0.15$^{ab}$ | 36.83 ± 0.23$^a$ | 36.55 ± 0.25$^{ab}$ |
| EAKc (125) | 37.23 ± 0.26$^a$ | 37.47 ± 0.13$^a$ | 36.50 ± 0.40$^a$ | 36.77 ± 0.35$^a$ | 36.67 ± 0.32$^{ab}$ | 36.50 ± 0.17$^{ab}$ | 36.30 ± 0.12$^b$ |
| EAKc (250) | 36.43 ± 0.79$^a$ | 36.83 ± 0.54$^a$ | 36.63 ± 0.47$^a$ | 35.97 ± 0.64$^a$ | 36.13 ± 0.42$^b$ | 35.77 ± 0.28$^b$ | 35.80 ± 0.10$^b$ |
| EAKc (500) | 37.47 ± 0.24$^a$ | 37.67 ± 0.28$^a$ | 37.83 ± 0.15$^a$ | 37.30 ± 0.21$^a$ | 37.13 ± 0.22$^{ab}$ | 37.27 ± 0.35$^a$ | 36.80 ± 0.30$^a$ |
| Paracetamol (100) | 37.27 ± 0.03$^a$ | 37.70 ± 0.06$^a$ | 37.43 ± 0.03$^a$ | 37.03 ± 0.03$^a$ | 36.77 ± 0.09$^{ab}$ | 36.57 ± 0.09$^{ab}$ | 36.45 ± 0.05$^{ab}$ |

EAKc: aqueous leaf extract of *Kalanchoe crenata*; BW: body weight; n = 5. Values are expressed as mean ± SEM. Within each column, means sharing the same superscript letter are not significantly different, whereas means with different superscript letters are significantly different ($p < 0.05$). The overall ANOVA p-values for normal temperature, post-induction temperature, and at 1, 2, 3, 4, and 5 h were 0.471, 0.349, 0.101, 0.065, 0.032, 0.0029, and 0.0063, respectively.

**Anti-inflammatory activity against egg albumin-induced paw edema**

The aqueous leaf extract of *K. crenata* reduced paw thickness at most observation times, with the greatest reduction observed at 500 mg/kg. At 5 h, paw thickness was 2.00 ± 0.05, 1.97 ± 0.03, 1.90 ± 0.02, and 1.80 ± 0.02 mm at doses of 62.5, 125, 250, and 500 mg/kg, respectively, corresponding to reductions of 4.76%, 6.19%, 9.52%, and 14.29% compared with the control group. Aspirin produced a greater reduction of 40.00%, with paw thickness reaching 1.26 ± 0.31 mm (Table 2).

**Analgesic activity in the hot-water tail immersion test**

The aqueous extract produced a variable antinociceptive response, with the most pronounced effect observed at 500 mg/kg. At this dose, tail-withdrawal latency increased from 2.50 ± 0.002 s at 30 min to 6.28 ± 0.18 s at 4 h. A moderate increase was also observed at 250 mg/kg, with a latency of 2.81 ± 0.04 s at 4 h. In contrast, the responses at 62.5 and 125 mg/kg remained close to, or slightly below, those of the control group at several observation times. At 4 h, the latency produced by the 500 mg/kg dose (6.28 ± 0.18 s) approached that of aspirin (100 mg/kg; 6.80 ± 0.06 s). Overall, the results indicate that the antinociceptive effect was most evident at the higher extract doses, particularly at 500 mg/kg (Table 3).

**Antipyretic activity in brewer's yeast-induced pyrexia**

Brewer's yeast injection produced an increase in rectal temperature in the control group. Following treatment, EAKc produced a gradual reduction in rectal temperature, with the most pronounced decrease observed at 250 mg/kg. At 5 h, rectal temperature reached 35.80 ± 0.10 °C in the EAKc

250 mg/kg group, compared with 37.30 ± 0.00 °C in the 0.9% NaCl control group.

EAKc at 125 mg/kg also reduced rectal temperature, reaching 36.30 ± 0.12 °C at 5 h, whereas the effect at 500 mg/kg was less pronounced, with a temperature of 36.80 ± 0.30 °C. Paracetamol (100 mg/kg) produced a temperature of 36.45 ± 0.05 °C at 5 h. Differences among treatments were not statistically significant at 1 or 2 h ($p = 0.101$ and $p = 0.065$, respectively), but became significant at 3 h ($p = 0.032$) and remained significant at 4 and 5 h ($p = 0.0029$ and $p = 0.0063$, respectively) (Table 4).

## DISCUSSION

The present study provides experimental evidence consistent with the traditional use of *Kalanchoe crenata* (Andrews) Haw. for the management of ear disorders in Côte d'Ivoire. The aqueous leaf extract exhibited anti-inflammatory, analgesic, and antipyretic activities in complementary experimental models. The most pronounced effects were generally observed at 500 mg/kg in the anti-inflammatory and analgesic assays, whereas the greatest reduction in rectal temperature in the antipyretic assay was observed at 250 mg/kg. The simultaneous occurrence of these pharmacological effects is relevant to the symptomatic management of inflammatory ear disorders, in which pain, inflammation, and fever may occur concomitantly.

The anti-inflammatory activity of the aqueous extract was evaluated using two models of acute paw edema induced by kaolin and fresh egg albumin. Paw edema is a widely used experimental model for evaluating acute anti-inflammatory activity. In the present study, acute paw edema was induced using kaolin and fresh egg albumin. The aqueous extract produced a progressive reduction in paw thickness with increasing dose, although its effect remained lower than that of aspirin. In the kaolin model, the 500 mg/kg dose produced a 21.99% reduction in paw thickness at 5 h, compared with 27.49% for aspirin. A similar trend was observed in the fresh egg albumin model, where the 500 mg/kg dose produced a 14.29% reduction, compared with 40.00% for aspirin. Although the extract was less active than the reference drug, its ability to reduce paw edema in two complementary inflammatory models supports the presence of anti-inflammatory constituents in the aqueous extract.

These findings are consistent with previous pharmacological investigations of *K. crenata*. Dimo *et al.* (2006) reported anti-inflammatory activity of leaf extracts and fractions of *K. crenata* in experimental models of acute and chronic inflammation. In particular, the n-butanol fraction showed marked activity in carrageenan-, histamine-, serotonin-, and formalin-induced inflammatory models, suggesting that polar constituents may contribute to the anti-inflammatory properties of the plant. More broadly, reviews of the genus *Kalanchoe* have documented anti-inflammatory and analgesic activities among its reported pharmacological properties and have highlighted flavonoids and other phenolic constituents as important classes of secondary metabolites in the genus (Milad *et al.*, 2014; Assis de Andrade *et al.*, 2023).

The anti-inflammatory activity observed in the present study may therefore be related, at least in part, to the phytochemical constituents previously reported in *K. crenata*. Flavonoids and other polyphenolic compounds can modulate several inflammatory pathways, including cyclooxygenase- and lipoxygenase-related processes and NF-κB-dependent signaling, thereby influencing the production of inflammatory mediators such as TNF-α, IL-1β, IL-6, and COX-2 (Panche *et al.*, 2016; Al-Khayri *et al.*, 2022). However, because individual constituents and molecular targets were not investigated in the present study, these mechanisms should be regarded as plausible explanations rather than experimentally demonstrated mechanisms. The analgesic assay further demonstrated the pharmacological potential of the aqueous extract.

In the hot-water tail immersion test, the extract increased tail-withdrawal latency in a generally dose- and time-related manner, with the strongest response observed at 500 mg/kg. At 4 h, the latency reached

6.28 ± 0.18 s, compared with 6.80 ± 0.06 s for aspirin. This result is consistent with previous studies reporting analgesic effects of *K. crenata* leaf extracts. Nguelefack *et al.* (2004) demonstrated analgesic activity of aqueous and ethanol extracts in several experimental models of pain, including thermal nociception. In a subsequent study, Nguelefack *et al.* (2006) also demonstrated analgesic effects of *K. crenata* leaf extracts in chemical and mechanical pain models. These studies provide pharmacological support for the analgesic activity observed in the present investigation.

The progressive increase in tail-withdrawal latency observed after administration of the extract suggests that its analgesic effect may persist over several hours. However, the tail immersion test primarily assesses the response to a thermal nociceptive stimulus and cannot by itself establish the precise mechanism of analgesia. The observed effect may involve modulation of central nociceptive pathways, while inhibition of prostaglandin-mediated processes may also contribute. Further experiments using selective antagonists and biochemical markers would be required to identify the mechanisms involved.

The antipyretic assay also showed a pharmacological response. Brewer's yeast induced an increase in rectal temperature, whereas treatment with the aqueous extract reduced body temperature, with the most pronounced reduction observed at 250 mg/kg. At 5 h, the temperature reached 35.80 ± 0.10 °C at this dose, compared with 37.30 ± 0.00 °C in the 0.9% NaCl control group. EAKc at 125 mg/kg also produced a reduction in temperature, reaching 36.30 ± 0.12 °C at 5 h, while the temperature in the 500 mg/kg group was 36.80 ± 0.30 °C. Paracetamol (100 mg/kg) produced a temperature of 36.45 ± 0.05 °C at 5 h. These findings indicate that the antipyretic response was not strictly dose-dependent, as the greatest reduction was observed at 250 mg/kg rather than at the highest tested dose.

Yeast-induced pyrexia is associated with the release of endogenous inflammatory mediators and activation of prostaglandin-dependent thermoregulatory mechanisms. Because cyclooxygenase-derived prostaglandins, particularly PGE2, contribute to the elevation of the hypothalamic thermoregulatory set point, compounds capable of modulating inflammatory mediator production may attenuate experimentally induced fever.

The antipyretic effect observed here may consequently be related to the anti-inflammatory properties of the extract. Flavonoids and other polyphenolic compounds reported in *K. crenata* may contribute to this effect through modulation of inflammatory signaling and oxidative processes. Recent experimental work has also provided additional evidence of anti-inflammatory and antioxidant effects associated with an aqueous extract of *K. crenata*, although the experimental model and endpoints differed from those used in the present study (Kandeda *et al.*, 2025).

The traditional relevance of these findings is supported by previous reports describing the use of *K. crenata* in African traditional medicine, including for otitis and inflammatory disorders. Nguelefack *et al.* (2006) specifically reported that *K. crenata* is traditionally used for otitis and inflammation, while subsequent reviews have documented the ethnomedicinal and pharmacological relevance of the genus (Milad *et al.*, 2014; Assis de Andrade *et al.*, 2023). The present results therefore provide pharmacological evidence consistent with the traditional use of the plant, particularly with respect to symptoms such as pain, inflammation, and fever.

Nevertheless, the findings should be interpreted with caution. The present study evaluated the pharmacological effects of a crude aqueous leaf extract in experimental animals and did not directly investigate otic inflammation or the infectious causes of ear disorders. Therefore, the results should not be interpreted as demonstrating that the extract can treat otitis itself. Rather, they indicate that the extract possesses pharmacological activities potentially relevant to the symptomatic management of inflammatory conditions associated with ear disorders.

Further studies should therefore focus on the phytochemical characterization of the aqueous extract, identification of active or marker compounds, evaluation of toxicity and safety, and investigation of the molecular pathways underlying the observed effects. In addition, antimicrobial studies against clinically relevant pathogens associated with ear infections would be important to determine whether the traditional use of *K. crenata* may involve both symptomatic and anti-infective effects.

Overall, the present findings, together with previous pharmacological evidence, support the biological plausibility of the traditional use of *K. crenata*. However, additional phytochemical, pharmacological, toxicological, and microbiological studies are required before standardized preparations or therapeutic applications can be proposed.

## CONCLUSION

The present study demonstrated that the aqueous leaf extract of *Kalanchoe crenata* (Andrews) Haw. exhibits anti-inflammatory, analgesic and antipyretic activities in complementary experimental models. While previous studies have documented analgesic and anti-inflammatory properties of *K. crenata*, the present investigation extends the existing pharmacological evidence by providing an integrated assessment of these activities together with antipyretic activity using an aqueous leaf extract. The observed activities provide additional experimental support for the traditional use of *K. crenata* in the symptomatic management of ear disorders in Côte d'Ivoire.

However, these findings should not be interpreted as evidence of efficacy against otitis itself or against its infectious causes, since no direct otic infection model was investigated. Further studies are warranted to characterize the active constituents, establish the safety profile, investigate the underlying mechanisms and assess the antimicrobial activity of the extract against relevant ear pathogens.

## RECOMMENDATION(S)

Further studies should focus on phytochemical characterization, safety assessment and clarification of the mechanisms underlying the observed activities. Antimicrobial studies against relevant ear pathogens and appropriate models of ear inflammation and infection are also needed to further assess the therapeutic relevance of *Kalanchoe crenata* for ear disorders.

## REFERENCES

**Al-Khayri JM, Sahana GR, Nagella P, Joseph BV, Alessa FM, Al-Mssallem MQ.** 2022. Flavonoids as potential anti-inflammatory molecules: A review. Molecules **27(9)**, 2901. https://doi.org/10.3390/molecules27092901

**Assis de Andrade E, Machinski I, Terso Ventura AC, Barr SA, Pereira AV, Beltrame FL, Strangman WK, Williamson RT.** 2023. A review of the popular uses, anatomical, chemical, and biological aspects of *Kalanchoe* (Crassulaceae): A genus of plants known as "Miracle Leaf". Molecules **28(14)**, 5574. https://doi.org/10.3390/molecules28145574

**Aydin S, Demir T, Oztürk Y, Başer KHC.** 1999. Analgesic activity of *Nepeta italica* L. Phytotherapy Research **13(1)**, 20–23. https://doi.org/10.1002/(SICI)1099-1573(199902)13:1<20::AID-PTR380>3.0.CO;2-J

**Béné K, Camara D, Fofie NBY, Kanga Y, Yapi AB, Yapo YC, Ambe SA, Zirihi GN.** 2016. Étude ethnobotanique des plantes médicinales utilisées dans le Département de Transua, District du Zanzan (Côte d'Ivoire). Journal of Animal and Plant Sciences **27(2)**, 4230–4250.

**Dimo T, Fotio A, Nguelefack TB, Asongalem EA, Kamtchouing P.** 2006. Antiinflammatory activity of leaf extracts of *Kalanchoe crenata* Andr. Indian Journal of Pharmacology **38(2)**, 115–119. https://doi.org/10.4103/0253-7613.24617

**European Union.** 2010. Directive 2010/63/EU of the European Parliament and of the Council of 22 September 2010 on the protection of animals used for scientific purposes. Official Journal of the European Union **L276**, 33–79.

**Harborne JB.** 1998. Phytochemical methods: A guide to modern techniques of plant analysis. 3rd ed. Chapman and Hall, London/New York. 302 pp.

**Huang GJ, Lin BR, Li PS, Tang N, Fan ZJ, Lu BQ.** 2025. The global burden of otitis media in 204 countries and territories from 1992 to 2021: A systematic analysis for the Global Burden of Disease study 2021. Frontiers in Public Health **12**, 1519623. https://doi.org/10.3389/fpubh.2024.1519623

**Kandeda AK, Mezui C, Kengni S, Baldagai N, Mabou ST.** 2025. *Kalanchoe crenata* Haw. (Crassulaceae) decreases hippocampal neuron loss and improves memory and executive function in aged rats: Implications for anti-inflammatory and antioxidant mechanisms. Brain and Behavior **15**, e70261. https://doi.org/10.1002/brb3.70261

**Kokon JE, Davis K, Nwidu LL.** 2016. Anti-inflammatory and antinociceptive activities of *Solenostemon monostachyus* aerial part extract in mice. Avicenna Journal of Phytomedicine **6(3)**, 284–294.

**Kouadio KK, N'Guessan K, Tra Bi FH, Koné MW.** 2014. Enquête ethnobotanique sur les plantes utilisées dans le traitement traditionnel des affections ORL dans trois régions de Côte d'Ivoire. Phytothérapie **12(5)**, 295–302.

**Milad R, El-Ahmady S, Singab AN.** 2014. Genus *Kalanchoe* (Crassulaceae): A review of its ethnomedicinal, botanical, chemical and pharmacological properties. European Journal of Medicinal Plants **4**, 86–104.

**Muhammad N, Saeed M, Khan H, Raziq N, Halimi SMA, Abass M.** 2013. Antipyretic and anticonvulsant activity of n-hexane fraction of *Viola betonicifolia*. Asian Pacific Journal of Tropical Biomedicine **3(4)**, 280–283. https://doi.org/10.1016/S2221-1691(13)60063-5

**Nguelefack TB, Fotio AL, Watcho P, Wansi SL, Dimo T, Kamanyi A.** 2004. Analgesic properties of the aqueous and ethanol extracts of the leaves of *Kalanchoe crenata* (Crassulaceae). Phytotherapy Research **18(5)**, 385–388. https://doi.org/10.1002/ptr.1444

**Nguelefack TB, Fotio AL, Watcho P, Wansi SL, Dimo T, Kamanyi A.** 2006. Analgesic and anticonvulsant effects of extracts from the leaves of *Kalanchoe crenata* (Andrews) Haworth (Crassulaceae). Journal of Ethnopharmacology **106(1)**, 70–75. https://doi.org/10.1016/j.jep.2005.12.003

**Panche AN, Diwan AD, Chandra SR.** 2016. Flavonoids: An overview. Journal of Nutritional Science **5**, e47. https://doi.org/10.1017/jns.2016.41

**Wagner-Jauregg T, Jahn U, Buech O.** 1962. The antiphlogistic testing of known antirheumatics on kaolin-induced rat paw edema. Arzneimittel-Forschung **12**, 1160–1162.

**Yimer T, Birru EM, Adugna M, Geta M, Emiru YK.** 2020. Evaluation of analgesic and anti-inflammatory activities of 80% methanol root extract of *Echinops kebericho* M. (Asteraceae). Journal of Inflammation Research **13**, 647–658. https://doi.org/10.2147/JIR.S267154