

**ASSESSMENT OF ANTI-INFLAMMATORY ACTIVITY OF ETHANOLIC EXTRACT OF ASAM KANDIS (*GARCINIA XANTHOCHYMUS* HOOK. F. EX T. ANDERSON) FRUIT**HANAFIS SASTRA WINATA<sup>1\*</sup>, ROSIDAH ROSIDAH<sup>1</sup>, PANAL SITORUS<sup>2</sup><sup>1</sup>Department of Pharmacology, Faculty of Pharmacy, University of Sumatera Utara, Medan, Indonesia. <sup>2</sup>Department of Biological Pharmacy, Faculty of Pharmacy, University of Sumatera Utara, Medan, Indonesia. Email: nafis.sastrawinata@gmail.com

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**ABSTRACT**

**Objective:** The objective of this study was to evaluate the anti-inflammatory activity in acute and subacute models of inflammation from ethanolic fruit extract of Asam kandis (*Garcinia xanthochymus* Hook. f. ex T. Anderson) in animal (rats) models.

**Methods:** Preliminary phytochemical screening was carried out by using standard procedures.. Assessment of acute and subacute models of inflammation was using carrageenan-induced paw edema method and cotton pellet granuloma method using three dosage treatments; 200 mg/kg BW, 400 mg/kg BW, and 800 mg/kg BW along with a negative control group (0.5% Na CMC) and positive control (Na diclofenac 2.25 mg/kg BW). The inhibition period was observed at 30, 60, 90, 120, 150, and 180 min time intervals.

**Result:** The phytochemical screening showed that the ethanolic fruit extract from Asam kandis contain contains flavonoids, glycosides, steroids, and triterpenoids. The anti-inflammatory result showed that the strongest inhibition produced by ethanolic fruit extract of Asam kandis occurred on the dosage of 800 mg/kg BW compared to the other doses (200 and 400 mg/kg BW) throughout the observation period.

**Conclusion:** This finding indicated that ethanolic fruit extract of Asam kandis (*G. xanthochymus* Hook. f. ex T. Anderson) might become an interesting candidate for treatment of inflammation.

**Keywords:** Asam kandis, Anti-inflammatory, Carrageenan, Paw edema, Cotton pellet.

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**INTRODUCTION**

Inflammation is the defensive response of the body which is non-specific toward the tissue injury and infection. Pain and inflammation are two states associated with numerous diseased conditions [1]. There are many conditions that could trigger the inflammation such as pathogens, chemical irritation, extreme temperature, distortion or disturbances of cells, and abrasions [2-4]. There are five characteristics of inflammation such as redness, swelling, the rise of temperature, pain, and loss of function [5].

Non-steroidal anti-inflammatory drug (NSAID) is one of the most common medications for inflammation, but unexpected bronchus, gastric mucosa, cardiac, and kidney-related side effects have been reported [6]. The utilization of natural sources to search for new pharmacologically active agents has led to the discovery of many clinically beneficial drugs that contributes to a key role in the treatment of human disease [7]. Therefore, many researchers are interested in drugs from the natural sources such as plants and herbs origin due to their wide range of pharmacological activities, including anti-inflammatory potential with minimum side effects [8].

*Garcinia xanthochymus* Hook. f. ex T. Anderson, also called as Asam kandis, belongs to the Clusiaceae family. *Garcinia* species are common in Southeast Asia. This species is rich of the phytochemicals compound that includes xanthone, flavonoids, and phenolic acids. Due to its rich pharmaceutical values, mostly the *Garcinia* species have been to treat many diseases such as antifungal, antioxidant, antiprotozoal, and anti-immunosuppressive [9]. *In vitro* investigation reported that the ethanolic fruit extract of *G. xanthochymus* has a good antioxidant effect (half maximal inhibitory concentration [IC<sub>50</sub>]: 22 µg/mL) [10]. Therefore, from the previous investigation, it can be concluded that *G. xanthochymus* has a good antioxidant properties. In this regard, it makes great sense to

evaluate the anti-inflammatory activity of Asam kandis (*G. xanthochymus* Hook. f. ex T. Anderson) fruit ethanolic extract using carrageenan-induced paw edema method and cotton pellet method.

**METHODS****Identification and authentication of plant materials**

Asam kandis (*G. xanthochymus* Hook. f. ex T. Anderson) fruits were obtained from region of Lubuk Minturun, Padang, West Sumatera, Indonesia. Fresh fruits were dried and then grounded to obtain a finely divided powder.

**Plant extraction preparation and phytochemical screening**

The powder of Asam kandis (*G. xanthochymus* Hook. f. ex T. Anderson) fruits was extracted using a triple maceration process with ethanol organic solvent. 900 g of powdered Asam kandis fruits is macerated in advance with the ethanol solvent for 5 days, then filtered, do it continuously until the filtrate obtained is clear and colorless [11]. Phytochemical screening carried out on Asam kandis fruits ethanolic extract includes examining the chemical secondary metabolites of alkaloids, flavonoids, glycosides, tannins, triterpenoids, and steroids. The preliminary phytochemical screening was using widely used methods [12-14].

**Preparation of animals**

Healthy adult male Wistar rats (150–200 g body weight) from animal house of Faculty of Pharmacy, University of Sumatera Utara, were used for the study. Rats were housed in a polycarbonate cages in a room with 12 h day-night circle. They were fed on a standard pellet diet and water *ad libitum*. The study was approved by Animal Research Ethics Committees (AREC) of University of Sumatera Utara (AREC No: 120/KEPH-FMIPA/2017) and the experiments were conducted according to the ethical norms and AREC guidelines.

### Anti-inflammatory activity

#### Carrageenan-induced paw edema

Healthy adult male Wistar rats were divided into five groups of six rats each. Edema was induced by injecting 0.05 mL of 1 % carrageenan suspension into the subplantar region of the right hind paw of the rats. Control group rats received 0.5% (w/v) Na CMC and the reference group of rats received 2.25 mg/kg BW sodium diclofenac, orally. The test groups of rats were treated orally with 200, 400, and 800 mg/kg BW ethanolic extract of Asam kandis fruits 40 min before carrageenan injection. The paw volume was measured by plethysmometer before carrageenan injection ( $V_0$ ) and 30, 60, 90, 120, 150, and 180 min after ( $V_t$ ). The inflammation was calculated as the increase in volume (mL) of the paw after treatment subtracted of basal volume. Results were expressed as percentage of inhibition of edema, calculated according to the formula [15,16]:

$$\text{Percentage inhibition (\%)} = \frac{\text{Mean paw inflammation of control} - \text{Mean paw inflammation of test}}{\text{Mean paw inflammation of control}} \times 100\%$$

#### Cotton pellet granuloma method

Wistar rats were divided into five groups of six rats each. Adsorbent cotton was made into pellets weighing  $20 \pm 1$  mg. The pellets were sterilized in a hot air oven at  $120^\circ\text{C}$  for 2 h. The abdomen was shaved cleanly, swabbed with 70% ethanol, and the sterilized cotton pellet was implanted subcutaneously under light ether anesthesia. Test drugs were administered once daily throughout the experimental period of 7 days. On the 8<sup>th</sup> day after implantation, rats were anesthetized and the pellets were dissected and dried at  $60^\circ\text{C}$  for 6 h, weighed after cooling. The mean weight of the cotton pellets of the control group as well as of the test groups was calculated. The transudative weight and percent granuloma inhibition of the test compound were calculated [17].

#### Statistical analysis

The significant difference of data between different groups was compared by ANOVA followed by Duncan's test.

### RESULTS AND DISCUSSION

This study investigated the anti-inflammatory activity of Asam kandis (*G. xanthochymus* Hook. f. ex T. Anderson) fruits ethanolic extract. The phytochemical screening of ethanolic extract of Asam kandis fruits revealed the presence of flavonoids, glycosides, steroids, and triterpenoids (Table 1). These results are related to previous research [18] that shown the presence of flavonoids, glycosides, steroids, and triterpenoids.

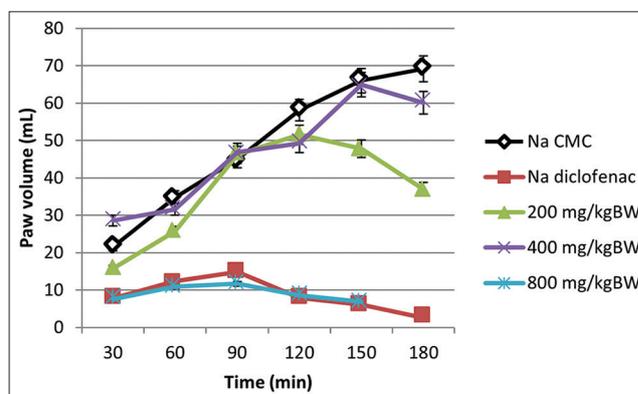
To evaluate the extract effect on anti-inflammatory activity, the ethanolic extract of Asam kandis fruits was tested using carrageenan-induced paw edema and cotton pellet granuloma method. In carrageenan-induced paw edema method, the paw volumes and percentage of inhibition of the control, standard, and test groups are shown in Figs. 1 and 2. The ethanolic fruit extract of Asam kandis showed anti-inflammatory activity compared to Na diclofenac. A dose of 800 mg/kg BW elicited a greater percent of inhibition of inflammatory after 60–180 min. These results showed that the ethanolic extract of Asam kandis fruit (dose 800 mg/kg BW) has the most potent anti-inflammatory activity.

The cotton pellet granuloma method has been widely used to access the subacute inflammation. The results are shown in Fig. 3. As shown in Fig. 3, all the test groups have the inhibitory activity, but the best doses are 400 and 800 mg/kg BW. It can be shown by the percentage of inhibition for 400 mg/kg BW is 58.10% and 35.89% for 800 mg/kg BW. These results suggest that the ethanolic extracts exhibit antitransudative effect.

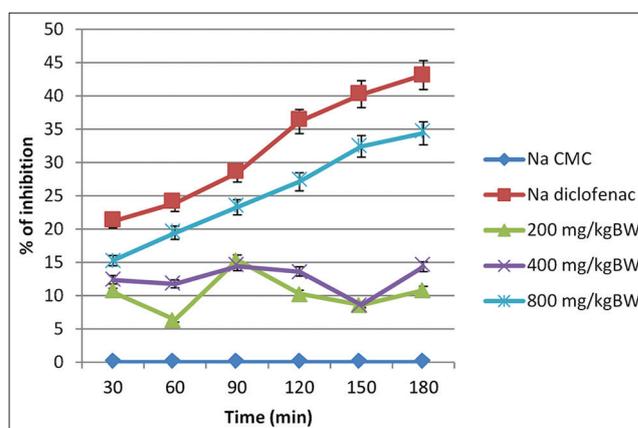
Drugs from plant sources have been used for the treatment of various diseases since ancient times. Herbal drugs are inexpensive and effective with fewer side effects [19]. Nowadays, the use of herbal drugs to cure inflammation and pain is gaining popularity due to their effectiveness,

**Table 1: Phytochemical screening result of ethanolic extract from Asam kandis fruits**

No	Screening	Result
1	Alkaloids	Negative
2	Flavonoids	Positive
3	Glycosides	Positive
4	Tannins	Negative
5	Saponins	Negative
6	Triterpenoid/steroids	Positive



**Fig. 1: Effect of various doses of Asam kandis fruit ethanolic extract in carrageenan-induced paw edema in rats (change in mean paw volume)**

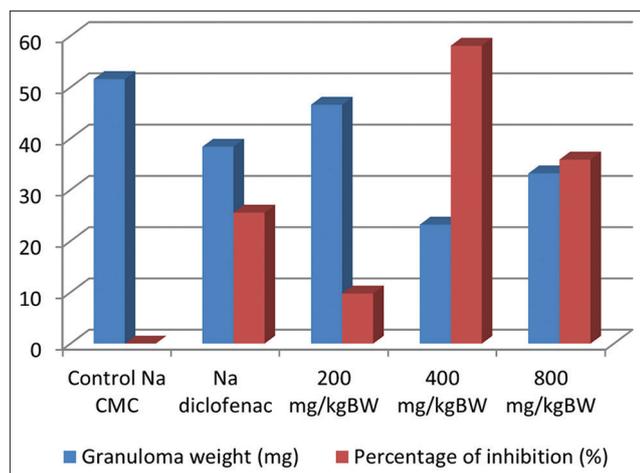


**Fig. 2: Effect of various doses of Asam kandis fruit ethanolic extract in carrageenan-induced paw edema in rats (% of inhibition)**

fewer side effects, low cost, and availability. NSAIDs act by inhibiting cyclooxygenase and the production of prostaglandins. Diclofenac offers relief from inflammation by suppressing the production of prostaglandins and bradykinin. The presence of flavonoids has been reported in *Garcinia* species, and flavonoids are known to inhibit prostaglandin synthetase. Previous studies have shown similar relationships between flavonoids and anti-inflammatory effects [20-29]. Therefore, it is possible that the anti-inflammatory action of Asam kandis fruit extract may be related to the inhibition of prostaglandin synthesis. The phytochemical profile of Asam kandis fruit extract may be explored further to identify the active constituents responsible for its anti-inflammatory activity.

### CONCLUSIONS

The ethanolic extract of Asam kandis fruit (*G. xanthochymus* Hook. f. ex T. Anderson) ethanolic extract at dose of 800 mg/kg BW had the most



**Fig. 3: Effect of various doses of Asam kandis fruit ethanolic extract in cotton pellet granuloma method**

potent anti-inflammatory activity throughout the observation period. These studies can be concluded that Asam kandis fruit (*G. xanthochymus* Hook. f. ex T. Anderson) has the potential to be developed as an anti-inflammatory agent.

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#### AUTHORS CONTRIBUTIONS

All the author have contributed equally.

#### CONFLICT OF INTERESTS

Declared none.

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