EUROPEAN JOURNAL OF BIOMEDICAL AND PHARMACEUTICAL SCIENCES
http://www.ejbps.com

ISSN 2349-8870
Coden USA: EJBPBO
Volume: 13
Issue: 4
Year: 2026

PHARMACOLOGICAL ASSESSMENT OF ANTIPYRETIC EFFECTS OF EXTRACTS, SOLVENT FRACTIONS AND ISOLATED COMPOUNDS FROM *ALSTONIA CONGENSIS* ENGL (APOCYANCEAE) ROOT BARK IN EXPERIMENTAL SWUISS ALBINO MICE

Cimanga K. R.*[a,c], Tshodi E. M.[a], Kikweta M. C.[a], Tona L. G.[b]

[a]Department of Medicinal Chemistry and Pharmacognosy, Faculty of Pharmaceutical Sciences, University of Kinshasa, P.O. Box 212, Kinshasa XI, Democratic Republic of Congo.
[b]Department of Pharmacology and Therapeutic, Faculty of Pharmaceutical Sciences, University of Kinshasa, P.O. Box 212, Kinshasa XI, Democratic Republic of Congo.
[c]Department of Pharmaceutical Sciences, Natural Products & Food Research and Analysis (NatuRAPT), University of Antwerp, Universiteitsplein1, B-2610, Antwerp, Belgium.

***Corresponding Author: Cimanga K. R.**

Department of Medicinal Chemistry and Pharmacognosy, Faculty of Pharmaceutical Sciences, University of Kinshasa, P.O. Box 212, Kinshasa XI, Democratic Republic of Congo.
DOI: https://doi.org/10.5281/zenodo.20034442

How to cite this Article: Cimanga K. R.*, Tshodi E. M., Kikweta M. C., Tona L. G. (2026). Pharmacological Assessment of Antipyretic Effects of Extracts, Solvent Fractions and Isolated Compounds from Alstonia Congensis Engl. (Apocyanceae) Root Bark in Experimental Swuiss Albino mice. European Journal of Biomedical and Pharmaceutical Sciences, 13(4), 243–253.

Article Received on 05/03/2026 **Article Revised on 25/03/2026** **Article Published on 01/04/2026**

ABSTARCT

This study was conducted to evaluate the antipyretic activity or effects of extracts, solvent fractions, crude polysaccharides and isolated compounds from *Alstonia congensis*. Engl. root bark against the fungus Brewer's yeast induced pyrexia in Swiss albino mice. Firstly, it was observed that the subcutaneous injection of Brewer's yeast at dose of 10 ml/kg bw induced significant elevation of rectal temperatures from 37.8±0.08 in 1h to 4039.83°C from 1 to in 5h. Secondly and inversely, it was perceived the oral administration of Aspirin as reference antipyretic drug caused significant lowering of rectal temperature from 37.5±0.4 to 34.6±0.8°C at the same times. Thirdly, in the same way, the oral administration of aqueous Ac-1 and methanol Ac-2 extracts at high dose of 400 mg/kg bodyweight induced also significant reduction of rectal temperatures at all administered dose of 200 and 400 mg/kg bodyweight in dose-dependent manner. Administered at high dose of 400 mg/kg bodyweight, they lowered rectal temperatures to values of 38.2±0.6 and 37.5±0.5°C in 1h and 37.05±0.2 and 36.58±0.7°C I after 5 respectively, compared to test test toxic negative control Brewer's yeast showing high rectal temperatures of 37.8±0.8 and 4039.83±0.5°C in 1 and 5h respectively. Fourthly, it was the same for soluble fractions chloroform, ethylacetate, *n*-butanol and residual aqueous solvent fractions induceding significant decrease of rectal temperatures between 38.4±0.6 and 37.5±0.5°C in 1h and between 36.5±0.6 to 37.5 ±0.5°C after 5h. Crude polysaccharide CPms reacted the same manner by lowering rectal temperatures from 38.4±0.7 to 36.8±0.8°C in 1 and 5h respectively compared to test toxic negative control Brewer's yeast presenting high rectal temperatures of 40.3±0.05°C after 5h. Although sSome increase of rectal temperatures was observed for some samples in some hours without important signification since they falled immediately in normal 36.5-37.5°C and the last rectal temperature induced by each test sample remained significant and determinative as it was all found to be in acceptable physiological limits 36.5-37.5°C. Isolated compounds Boonein, Echintamine, 6,7 Secangustilobine and β-Amyrin also exhibited good antipyretic activity by reducing significantly rectal temperatures induced by the test toxican Brewers's yeast in treated Swiss albino mice from 38.2±0.6 to 37.0±0.60°C in 1h and between 36.5±0.5 to 37.5±0.5°C remaining normal limits compared to text toxican Brewer'sy yeast showing greater rectal temperatures between 37.8±0.08 in 1h and 40.3°C in 5h. These results lowering effects of high rectal temperatures caused by the toxixcn Brewer's yeast, showed clearly that extracts, solvent fractions, crude polysaccharides and isolated compounds from *Alstonia congensis* root bark as well asand Aspirin as reference antipyretic drug used were endowed with appreciable and interesting antipyretic activity, exploitable in traditional medicine mainly for aqueous extract used to treat fever.

KEYWORDS: *Alstonia congensis*, root bark, extracts, solvent fractions, Aspirin, pyrexia, fever, antipyretic activity.

1. INTRODUCTION

Extracts, polysaccharides and solvent fractions in rapport with the empirical uses of mainly aqueous extract of this medicinal plant, have demonstrated that these samples are endowed with different biological activities in previous biological tests *in vitro* and *in vivo* such as antidiarrheal *in vivo* (Nsaka et al., 2012, 2017), antiprotozoal *in vitro* (Lumpu et al., 2013a, 2017), Safety and Tolerability in Wistar rats *in vivo* (Nsaka et al., 2013, 2017), antidiabetic *in vivo* (Cimanga et al., 2016, Nsaka et al., 2017), antispasmodic on isolated guinea-pig ileum (Nsaka et al., 2017, Cimanga e al., 2025), *in vitro* an *in vivo* antiplamodial activity of extracts, soluble fractions and isolated compounds against *Plasmodium falciparum* (Cimanga et al., 2019.

For chemical point, two triterpenoids α-amyrin acetate, lupeol and β-sitosterol were isolated from the root bark of *Alstonia scholaris* (Hemalatha et al., 2008), alkaloids, steroids, saponins, triterpenes, flavonoids, tannins, and glycosides wre isolated from the root bark of A. boonein (Obiagwu et al., 2014). In 2019, Cimanga et al (2019) have isolated a sesquiterpene Boonein, two alkaloids Echitamine and 6,7 Secoangustilobine and a terpenoid β-Amyrin were isolated from Alstonia congensis root bark (Cimanga et al,. 2019).

Pyrexia referred to fever being a regulated increase of body core temperature characterized by an increased thermoregulatory set point, which results from the interaction of the central nervous and immune system (Wali et al., 2018). Normal body temperature is regulated by centers in the hypothalamus that ensures balance between heat and gain loss.

Fever occurs where there is a disturbance of this hypothalamic thermostat which leads to the set point of the body temperature being raised. This will cause readjustment to normal set point by temperature regulating system which operates to reduce the body temperature (Kelly, 2006, Wali et al., 2018). Fever is one of the most rampart clinical signs (Karakitsos et al;, 208, Wali et al., 2028. Infection, tissue damages, malignancy or other diseased conditions can cause fever (Chattopadhyay et al., 2015, Wali et al, 2018). Fever is described as the body's natural defense mechanism to create an uncondusive environment among which infectious agents or damaged tissues cannot withstand (Hajare et al., 2000, Wali et al., 2018).

Fever triggers increased muscle tones and chills (Akpa et al., 2014, Wali et al., 2018). Certain endogenous substances including tumor necrosis factor-alpha (TNFα) and prostaglandins can produce fever. Antipyretic drugs are medications that are used to reduce fevers (Wali et al., 2018). They are known to act either centrally on the temperature regulation centers of the hypothalamus or peripherally by inducing vasodilatation and heat dissipation (Adesokan et al., 2008, Wali et al., 2018). They also act by inhibiting the biosynthesis of prostaglandin E2 (Wali et al., 2018) possibly by inhibiting COX-2 expression (Luo et al., 2005, Gege-Adebayo et al., 2013, Wali et al., 2018). Long time usage of these antipyretic drugs produces undesirable side effects including gastrointestinal disorders, renal damage and hepatic diosrders. Fever is a medical condition when the body temperature increases beyond the normal range of 36.5-37.5°C. This condition results from interference in health and is characterized by chills and redness in the skin area (Suproborini, 2018).

Fever is generally caused by an infection induced by microorganisms such as bacteria, viruses, parasites, or even fungi. If not managed appropriately, fever can cause complications in therapy (Azis, 2019). It is characterized by some symptoms such as lack of oxygen, lack of appetite, weakness, headaches, muscle aches, and dehydration (Samiun, 2020). Fever therapy involves the administration of antipyretic medications, such as paracetamol alone or combined du codeine and ibuprofen. Patients frequently use these drugs due to their over-the-counter availability, safety, accessibility, and affordability (Samiun, 2020), but are uneasily accessible to more people in developing countries because of their high prices. The antipyretic effects of this medication are achieved by inhibiting the COX-2 enzyme in the Central Nervous System (CNS), thus preventing arachidonate acid conversion to prostaglandin, which is the mediator of fever or the temperature-inducing agent in the hypothalamus thermostat (Suproborini, 2018, Nurdiani and Dika, 2023).

A large number of synthetic antipyretic drugs are available in local market such as Naproxen, Ibuprofen, Nimesulide, Ketoprofen, Aspirin, Choline salicylate, Novalgine and Paracetamol to treat fever (Anonymous, 2018). All these antipyretic drugs inhibits the COX enzymes causing the no production of prostaglandin which increase the temperature, but they are associated with side effects like the NSAIDs causing gastric lesions some others show the signs of anemia, nausea, clotting problems, kidney problems, throat, skin infections and the liver cytotoxicity (Anonymous 2018).

Despite of the frequently conveyed evidences about the traditional claimed use of *A. ccongensis* root bark to treat fever in decoction form, no any scientific study reporting its antipyretic activity, have been found in literature until now. Reason for which the present study was initiated to evaluate the antipyretic activity of aqueous, methanol 80%, solvent fractions, crude polysaccharides extracts as well isolated compounds against Brewer's yeast-induced pyrexia in animal model.

2. MATERIALS AND METHODS

2. Materials and Methods

2.1. Plant material

Root barks of *Alstonia congensis* Engl. (Apocynaceae) were firstly collected in Kinshasa in July 2013. The plant

was identified at the Institut National d'Etudes et de Recherches en Agronomie (INERA), Department of Biology, Faculty of Sciences and Technoogies, University of Kinshasa. A voucher specimen INERA12765TGRB was deposited in the herbarium of this institute. For the present study, a fresh new batch plant material was collected at the same place in October 2023. The plant material was dried at room temperature and reduced to powder kept in brown bottle hermetically closed before use to avoid contaminations.

Figure 1: *Alstonia congensis* (leaves, stem and flowers) hermetically closed before use to avoid contaminations.

2.2. Preparation of extract and its fractionation

20 g of powdered rootstem barks were mixed with 300 mLl distilled water and boiled on a hot plate at 100°C for 15 minutes. After, cooling, the mixture was filtered on sterile cotton and on filter paper Whatman N° 1 and the resulting filtrate evaporated *in vaccuo* giving a dried extract denoted as AcC-1 (14.37 g). After, an amount of 10 g of AcC-1 extract were dissolved in 200 ml distilled water and filtered as described above, and the resulting filtrate was exhaustively and successively extracted with solvents of different polarities including chloroform, ethylacetate, and *n*-butanol. All fractions including rsisualresidual aqueous phase were treated as described above yielding corresponding dried extract denoted as chloroform (AC-1.1: 1.46 g), ethylacetate (AC-1.2: 2.45 g), *n*-butanol (AC-1.3: 2.75 g) and. The residual aqueous phase was also evaporated in the same way given a dried extract denoted as AC-1.4 (3.59 g) (Cimanga et al., 2020). On the other hand, 20 g of powdered root barks were macered with methanol 80% for 24h. After filtration as described above, the marck was exhaustively percolated with the same solvent until incolore solvent. The macerate and percolate were combined and treated as described above yielding dried extract denoted Ac-2 (13.67g).

2.3. Extraction of alkaloids and terpenoids

Alkaloids were isolated as followed: 1000 g of powdered rootstem barks of *A. congensis* were macerated with 3 L 80% methanol 80% for 24h. After filtration on sterile cotton and paper filter Whatman N°1. Next, the marc was percolated exhaustively with the same solvent. The macerate et percolate were combined and evaporated *in vaccuo* giving dried extract denoted as A-1 (45825.90 g). 420 g of AC-1 extract were dissolved in 300 ml HCl 0.1N and filtered as described above. The resulting filtrate was alkalinized with 160 mL NH3 10% and extracted exhaustively with CHCl3 until negative test with Dragendoff's reagent giving 182.54 g dried extract. 1150 g of CHCl3 extract were chromatographied on silica gel column (4 x 12 cm, Davisil, LG 60 A 40-60 µm, Merck, Germany) eluated with a stepwise gadientgradient of CHCl3/MeOH 100, 1:9, 2:8 to 100% MeOH. Several fractions of 10 ml were collected and anlaysedanalyzed by thin layer chromatography on silica plates (0.25 mm thickness layer, Merck, Germany) using CHlCl3/MeOH 9/1 as mobile phase. Fractions according to chromatographic profile, were combined as followed: F1-9 (5.8760 mg) and, F10-20 (868.32 mg)., F21-34 (1134 mg), F36-38 (20 mg), F39-76 (178.36 mg), F76-96 (98 mg), F97=136 (408.38 mg).

Fraction F10-920 was submitted to preparative chromatography on silica gel plates (1 mm thickness layer, Merck, Germany) with CHCl3/MeOH 9/1 as mobile phase, resulting in the isolation of compound 1 (23.46 mg) chromatographically pure and . Fraction F22-24 were submitted to CCM preparative on silica gel plates (thickness layer 1 mm) with CHCl3/MeOH:NH310%: 9/1/0.5 as mobile phase resulting in the isolation compound 2 (2.434 mg) chromatographically pure. Factions F3910-7620 was submitted to chromatography sur column eluated with CHCl3/MeOH in different proportions and several factions were collected and analysed on CCM as described above. Resulting factions F15-26 (8.3475 mg) were submitted to preparative chromatography on silage plates (thickness layer 1 mm) using CHCl3/MeOH/NH310%: 9/1/0.5 as mobile phase, resulting in the isolation compound 3 (32.16 mg) and 4 (332.714 mg) chromatographically pure. (Harborne, 1998).

2.4. Extraction of polysaccharides

300 of powdered root barksleaves were macerated with 500 ml distilled water for 24 h. After filtration on filter Whatman N°1, the filtrate was concentrated *in vacuum* to 30 ml. To this concentrated aqueous solution, 100 ml of ethanol 95°C was added and left in freezer at 4°C for 48 h. In these conditions, a white precipitate was obtained, washed with ethanol and dried in a hot at 50°C for one day giving dried extract (CPAcM: 125.64 g). Tested with phenol/H2SO4 conc., it showed a colour as a positive test for the presence of polysaccharides (Harborne 1998, Soniamol et al., (2011), Trease and Evans, 2000, Cimanga et al., 2020, 2021).

2.5. Evaluation of antipyretic

2.5.1. Induction of fever or pyrexia

Antipyretic activity was evaluated using methods previously described by Tesema and Makonnen., 2015, Olorukooba et al., 2020 and Tegegne et al., 2023). The normal body temperature of each mice was checked by using digital thermometer at the begining of the study. Pyrexia was induced in all mice by subcutaneously

injecting a 30% w/v yeast extract powder suspension in 0.9% normal saline (*Sacchromyces cervisiae*) or Brewer's yeast at a dose of 10 mL/kg bw below the nape of the neck. Animals were fasted with free access to only water *at libit*um, after injection of Brewer's yeast after 5h.

Rectal temperature of each selected mice was then recorded and pyrexia was confirmed by increase in temperatures between 38 and 40.3°C and these mice were selected for the experiment, while animals showing less than 1°C rise in temperature were excluded.

2.5.2. Treatment of Swiss albino mice with fever or pyrexia

The selected mice subcutaneously injected with Brewer's yeast at dose of 10 mL/kg bw and were randomly divided in following groups of 3 mice each as followed :

-Group I was oallyorally administered 5 mL distilled water as test normal negative control,

- Group II received 5 mlL distilled water and Brewer's yeast suspension by subcutaneous injection of 10 mlL/kg bw corspondingcorresponding to 3g/kg bw of 30% w/v Brewer's yeast to induce pyrexia or fever state as test toxic negative control to induce pyrexia or fever state as toxic negaive control,

- Group III received orallyl Aspirin at oral of 100 mg/kg bw as positive control,

- Groups IV and Va and b were separately and orally administered 200 (a) and 400 (b) mg/kg bw of aqueous Mms-1 and methanol Mms-2 extract respectively.

- Group VI to IX were separately and orally administered the same doses with solvent fractions chloroform, ethylacetate, *n*-butanol and residual aqueous Mms-1.1 to 1.4 solvent fractions respectively.

- Group X was orally administered the same oral doses of the same oral dose of crude polysaccharides CPMmsAc extract.

- Groups XI to IV were adminsisteredadministered separetelyseparately 50 and 100 mg/kg bw of espectiverespective compounds.

The experiment used only mice that showed a temperature increase of ≥0.5°C after yeast injection. One hour after the yeast administration of all samples, the rectal temperature of each mice had been measured again to coincide with the stable or peak phase of fever indicating an appropriate time to test the antipyretic activity of test samples. The rectal temperature of all groups was recorded at 1, 3 and 5h.

3. RESULTS AND DISCUSSION

3.1. Antipyretic activity of Aspirin, extracts, solvent fractions an crude polysaccharides

In yeast-induced pyrexia model, the subcutanous injection of 30% Brewer's yeast suspension significantly increased the rectal temperatures of the mice from 1h to 5 h after administration, owing the release of pro-inflammatory cytokines, which stumulated the synthetis of PGE2 in the vivinityvicinity of the hypothalamic thermoregulatory centers once they reached circulation (Shashank Kumar, 2013, Tegegne et al., 2023) as presented in Table 5.

These temperatures generated by negative text toxic control DW + Brewer's yeast (test toxic negative control) were. prominent from 37.8±0.9 to 40.339.8±0.5°C after respectively 1 and 5h of observationed, compared to test normal negative control consomming only distilled water giving low rectal temperatures ranging between 36.7±0.04 and 37.4±0.06°C and were found to be in normal physiological ranges 36.5-37.5°C.

Administered oral doses of 200 and 400 mg/kg bw of aqueous and methanol 80% extracts Ac-1 and Ac-2, solvent fractions Ac-1.1 to 1.4 and crude polysaccharides CPAc from *A. congensis* root bark decreased remarkably rectal temperatures of treated mice with pyrexia in dose-dependent manner as well as Asprin used as reference antipyretic product acting in the same manner administered at oral dose of only at oral dose of 10 mg/kg bw compared to test toxic negative control (DW + Brewer's yeast generating prominent rectal temperatures (Table 15).

Table 1: Rectal temperatures of treated mice induced by extracts, solvent fractions and crude polysaccharides of treated mice induced by extracts, solvent.

Groups	Dose	Initial temperatures	1h	3h	5h
TN. Control I	DW	36.7±0.04	38.0±0.03	37.0±0.06	36.8±0.03
TToxic N. control BY II	DW+Y	37.8±0.8	39.9±0.9	39.8±0.6	40.3.8±0.5
Aspirin	100	37.5±0.4	37.5±0.2	36.8±0.6	34.6±0.8
Ac-1	200	37.3±0.5	37.2±0.4	38.4±0.5	36.8±0.6
IV	400	37.4±0.4	38.2±0.6	37.4±0.5	37.0±0.2
Ac-1.1	200	36.8±0.5	36.7±0.5	38.1±0.2	36.6±0.10
VI	400	37.5±0.2	37.5±0.6	39.1±0.4	36.7±0.5
Ac-1.2	200	36.5±0.3	39.1±0.7	36.8±0.4	37.5±0.4
VII	400	37.3±0.5	38.4±0.6	37.0±0.2	36.7±0.6
Ac-1.3	200	36.6±0.5	37.5±0.2	37.3±0.4	37.5±0.7
VIII	400	37.4±0.4	39.2±0.7	37.0±0.2	36.8±0.8
Ac-1.4	200	37.6±0.5	38.2±0.6	37.3±0.5	37.5±0.7
IX	400	37.5±0.4	37.2±0.7	38.0±0.2	37.5±0.5
Ac-2	200	36.8±0.6	37.5±0.2	37.4±0.4	36.8±0.8

IV	400	37.5±0.5	37.5±0.5	38.0±0.2	36.5±0.7
CPAc	200	36.7±0.5	37.5±0.2	37.5±0.4	36.9±0.6
X	400	36.9±0.4	38.4±0.7	36.7±0.2	36.8±0.8

See Table 1.N. c TN control : test normal control, TToxic N. BY : test toxic negative control Brewer's yeast, DW: distilled water, Ac-1 : aqueous extract, Ac-1.1 to 1.4:. solvent fractions chloroform, ethylacetate, *n*-butanol and residual aqueous solvent fractions respectively, CPAc : crude polysaccharides.

In fact, aqueous Ac-1 and methanol Ac-2 extracts administered at high oral dose of 400 mg/kg bw displayed significantly lowering rectal temperatures of treated mice with pyrexia to value temperatures ranging from 38.2±0.6 to 37.5±0.5°C in 1h and continued to decrease gradually to 37.50±0.2 to 36.58±0.7°C after 5h respectively, while Aspirin as reference antipyretic product exerted the same effect by lowering these temperatures to value of 37.5±0.5 and 34.6±0.8 °C after 1 and 5h of treatment respectively, compared to test toxic negative control (DW + Brewer's yeast) showing prominent rectal temperatures of 37.8±0.8 and 4039.83±0.5 at the same hours.

Solvent fractions Ac-1.1 to 1.4 chlorofoem, ethyalacetate, n-butanol and residual aqueous solvent fractions responded in the same order by breaking down significantly these rectal temperatures of treated mice with pyrexia bringing back them to values between 37.5±0.2 and 38.4±0.4°C in 1h and continued their fall or abatement to values of 36.5±0.6 and 37.5±0.5 after 5h, while crude polysaccharides caused remarkable decrease of these temperatures to value of 38.4±0.7°C and 36.8±0.8°C after 1 and 5h respectively.

This increase antipyretic effect of extracts and solvent fractions from other medicinal plants, was also previously observed in other studies without comment (Tesema et al, 2015, Adbidoye et al., 2017, Olorukooba et al., 2020, Ghauri et al., 2021, Geresu et al;, 2022,Tegegne et al., 2022, Alkandahri et al., 2022, 2024) and could be explained by the nature of the active principles responsible for antipyretic activity and our results were on *A. congensis* root barks samples, were in good accordance with these previous reported results on antipyretic effects studied of extracts and solvent fractions from other medicinal species.

Furthtermore, higher activity was observed with high doses that might contain more active and synergistic principles responsible for this biological activity than lower doses as also previously telled by Tegegne et al., (2023).

During this experiment, it was aslo regarded or examined that some test samples increased rectal temperatures to 38-39°C than the normal ranges as 36.5-37.5°. These cases were also reported in other studies on antipyretic activity of other medicinal plant species without comments (Teggne et al. 2022, Tripathy et al., 2023, Gunarti et al., 2025) as already mentioned above. Fortunately, by simple observation of in these cases noticed in the pesentpresent study, these high temperatures were brought back quickly to stick on or post normal temperatures and the last recorded temepaturestemperatures after 5h were significative and determinative confirming that these extracts, solvent fractions and crude polysaccharides extract from *Alstonia congensis* root bark as well as Aspirin used as reference product were endorsedwed with good antipyretic properties since these rectal temperatures at the end of experiment, stayed in acceptable physiological ranges 36.5-37.5°C.

Figure 4 carried over the effects of test toxic negative control Brewer's yeast (TTNc BY), AsprinAspirin, aqueous Ac-1 extract to and methanol 80% Ac-2 extracts on rectal temeraturestemperatures of treated mice with pyrexia. Results pointed or evinced that the test normal negative control administered 5 ml distilled water generated temperatures from 38. 0 to 36.8°C considered to be in acceptable physiological limits 36.5-37.5°C although an inscrease rectal temperature was observed at 38.2°C for aqueous Ac-1 extract without important significance after 1h since it declined immediately in following hours in acceptable limits.

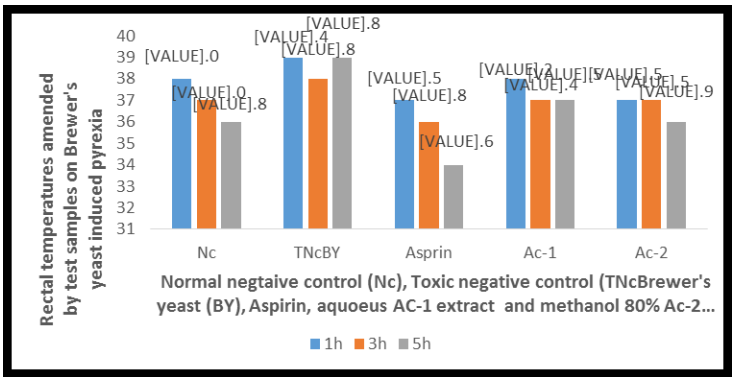


Figure 4: Effects of test normal negative control (Nc), test toxic negative control (TTNc BrewerY), Aspirin, aqueous Ac-1 extract and methanol 80% Ac-2 extract on rectal temperatures of treated mice with pyrexia.

In acceptable physiological limits 36.5-37.5°C although an increase rectal temperature was observed at 38.0°C without important significance after 1h since it declined immediately in following hours in acceptable limits (Fig. 4). Its effects were opposite to those manifested by test toxic control Brewer's yeast (TTNc BY) showing marquant rectal temperatures ranged betwenbetween 38.8 and 4039.83°C, high out in physiological ranges and compared to test normal negative control presenting rectela values in accaeptable physiological limits (Fig. 4).

The oral administration of Aspirin as reference antipyreytic product adminsiteredadministered at oral dose of 100 mg/kg bw, occasioned remarkably reduction of these temperatures to value of 34.6°±0.8°C under the normal limits after 5h, established its strong antipyretic effect, while orale administration of both aqueous Ac-1 and methanol 80% Ac-2 extracts administered at high oral dose of 400 mg/kg bw, brought significantly these rectal temperatures to respective low values of 37.05±0.2 and 36.85±0.7°C respecctively after 5h compared to negative control (Brewer's yeast (Nc +Y)) showing rectal high rectal temperature of 4039.83±0.5°C after the same hour.

This significative lowering effects produced by these samples on rectal temperatures compared to test toxic control Brewer' yeast, clearly demonstrated without ambiguity that Aspirin, aqueous Ac-1 and methanol 80% Ac-2 extracts respectively, exerted good and interesting antipyretic effect exploitable mainly for aqueous extract Ac-1 infor the treatment of fever in tradional medicine.

Figure 5 notified the effects of test normal control (TNC), test toxic control Brewer's yeast, Aspirin, solvent fractions Ac-1.1 to 1.4 and crude polysaccharides extract on high rectal temperatures induced by Brewer's yeast injection in mice.

Subsequently, the oral administration of chloroform, ethylacetate, n-butnaol and residual aqueous solvent fractions Ac-1.1 to 1.4 solvent fractions, displayed significant lowering of rectal temperatures between 36.5±0.6 and 37.5°C after 5h when administered at high oral dose of 400 mg/kg bw confronted to test toxic negative control (Brewer's yeast (TNc BY)) showing rectal high rectal temperature of 4039.83±0.5°C at the same hour.

Crude polysaccharides CPAc administered at hight oral dose of 400 mg/kg bw, caused also the same effect on rectal temperatures between of 36.8±0.8°C after 5h confronted to negative control Brewer's yeast presenting high rectal temperatures at the same hour (Fig. 5).

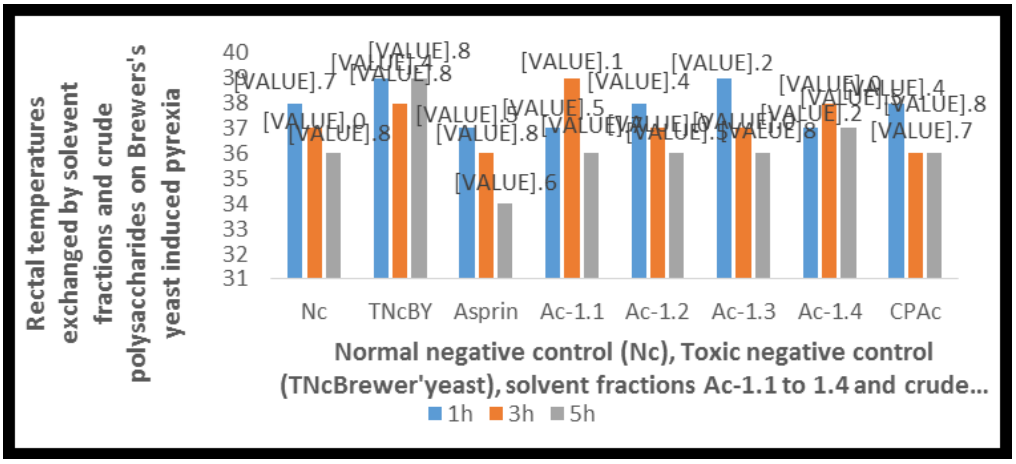


Figure 5. Effects of test normal control (Nc), test toxic control Brewer's yeast (TTNc BY, Asprin, aqueous and solvent fractions Ac-1.1 to 1.4 and crude polysaccharides CPAc extract on rectal temperatures induced by Brewer's yeast injection with pyrexia.

NevertelessNevertheless, aqueous Ac-1 extract, solvent fractions chloroform Ac-1.1, ethylacetate Ac-1.2, *n*-butanol Ac-1.3 and residual aqueous Ac-1.4 solvent fractions and crude polysaccharides CPAc extract generated high rectal temperatures in some hours between 38.2 and 39.6°C without significant importance or consequence since they falled immediately in normal ranges in future hours as already mentioned above (Fig. 5).

AditionallyAdditionally, the most principal mechanism for lowering body temperatures induced by Brewer's yeast-induced was based on the inhibition of the synthesis of protanglandinprostaglandin production within the brain, especially PGE2, secondary to inhibiting the enzymes responsible for prostanglandinprostaglandin synthesis, possibly due to the inhibition of cyclooxygenase enzyme stimulation of the system's antipyretic substance (Khan et al., 2014, Tegegne et al., 2023).

On mechanisms of action, it was pointed out that antipyretic drugs like acetylsalicylic acid (Aspirin) and non-steroidal anti-inflammatory drugs (NSAIDs)

reduced body temperatures by inhibiting the synthesis of prostangladinsprostaglandins (E-type) in hypothalamus. Paracetamol gave antipyretic effect by inhibinginhibiting the cycloxygenasecyclooxygenase (COX) iso-enzyme activity in brain (Jyothi et al., 2024, Nhung et al., 2023). Thus, it was supposed and speculated that extracts, solvent fractions and crude polysaccharides extract from *A. congensis* root bark would act also in the same manner to generate their antipyretic effect in this animal model.

Additionally, some mechanisms of actions for some secondary metabolites were also described for the inhibition of pyrexia or fever induced by the toxicant Brewer's yeast:.

In fact, alkaloids acted as antipyretic agents primarily by reducing fever through anti-inflammatory pathways, including inhibiting cyclooxygenase (COX) enzymes, suppressing proinflammatory cytokines (e.g., IL-, TNF-, IL-6), and stimulating the pituitary-adrenal cortex axis. These plant-derived compounds regulate body temperature by blocking fever-induced mediators rather than affecting normal body temperature.

Key Mechanisms of alkaloids as antipyretics included

- Inhibition of Inflammatory Mediators: Alkaloids inhibit the release of proinflammatory cytokines such as interleukins (IL-1, IL-6) and tumor necrosis factor (TNF-).
- COX Enzyme suppression: Similar to nonsteroidal anti-inflammatory drugs (NSAIDs), certain alkaloids inhibit cyclooxygenase (COX) enzymes, preventing the synthesis of prostaglandins from arachidonic acid in the central nervous system, which reduced fever.
- Regulation of immune responses: Alkaloids like stephanine and berberine regulated the NF-B pathway, reducing inflammation and fever.
- Activation of neuroendocrine pathway: Some alkaloids, such as those found in Chinese materia medica, stimulated the pituitary-adrenal cortex axis to release adrenal cortex hormones, which act to reduce fever.
- Analgesic/Anti-nociceptive Effects: Diterpenoid alkaloids may block nerve receptors or inhibit P2X3 receptors to reduce pain-related fever.
- Inhibition of the production of pro-inflammatory cytokines such as interleukins 1β and 6 (IL-1β, IL-6), interferon (IF-α) and tumor necrosis factor (TNF).
- Acting on the brain which sets the thermoregulatory center at a lower-temperature regulatory area of the hypothalamus acting on the brain which sets the thermoregulatory center at a lower-temperature regulatory area of the hypothalamus.

Examples of Antipyretic alkaloids

- Berberine: An isoquinoline alkaloid with strong anti-inflammatory properties.
- Stephanine: Known for inhibiting inflammatory edema and cytokines in LPS-induced models.
- Diterpenoid Alkaloids: Compounds like lappaconitine and aconitine act on descending inhibitory systems.

These compounds were considered promising, safer alternatives to conventional NSAIDs for treating fever (Kolawole and Dapper, 2016, Ahmad et al., 2017, Yimer er al;, 2021, Asante et al., 2023, Kamoru et al., 2023, Nawaz et al. 2025.

Fflavonoids, steroids and tannins were known to be the major inhibitors of PGE2 synthase, cycloogenase cyclooxygenase and lypoxygenase enzymes helping in the inhibition of pyrexia. Flavonoids had been also found to interfere with prostanglandins and related compounds had been shown to inhibit arachidonic acid peroxidation resulting in lower prostanglandin levels and thus resulted in the decline of fever (Tegegne et al., 2023). Another possible mechanism of action of medicinal plant extracts and solvent fractions was the mediation of superficial blood vessel dilatation, resulting in improved or ameliorated heat loss from the resetting of the hypothalamic thermostat and the supressionsuppression of mediators induced pyrexia can be considered with some importance

Flavonoids acted as antipyretic (fever-reducing) agents primarily by mitigating the inflammatory response that triggers fever, rather than acting as direct antipyretics like paracetamol. They function by inhibiting key enzymes, reducing pro-inflammatory cytokines, acting as antioxidants, and modulating signaling pathways in the central nervous system.

Their key mechanisms included

- Inhibition of pro-inflammatory Enzymes: Flavonoids inhibit cyclooxygenase-2 (COX-2) and 5-lipoxygenase (5-LOX), which are responsible for the production of prostaglandins (specifically PGE_2) and leukotrienes. PGE_2 is a critical mediator in the hypothalamus that induces fever.
- Modulation of cytokines: They reduce the synthesis and release of pro-inflammatory cytokines, including Interleukin-1 beta ($IL-1\beta$), Interleukin-6 $IL-$and Tumor Necrosis Factor-alpha ($TNF-\alpha$).
- Inhibition of signaling pathways: Flavonoids inhibit the nuclear factor kappa-light-chain-enhancer of activated B cells ($NF-\kappa-\kappa B$) and Mitogen-Activated Protein Kinase ($MAPK$) signaling pathways, which regulate the expression of pro-inflammatory genes.
- Antioxidant activity: As potent antioxidants, they scavenge reactive oxygen species (ROS) and reduce oxidative stress, which contributes to the inflammation-induced fever.

- Inducible nitric oxide synthase (iNOS) inhibition: They inhibit iNOS, reducing the production of nitric oxide (NO) that can contribute to inflammatory responses.

The key flavonoids with antipyretic activity included

- Quercetin: Known for potent inhibition of COX-2 and 5-LOX, reducing PGE_2.
- Baicalein/Baicalin: Reduces fever by inhibiting the $NF-\kappa B$ pathway and reducing pro-inflammatory cytokines.
- Luteolin & Apigenin: Inhibit $NF{-}\kappa B$ and $MAPK$ pathways, decreasing pro-inflammatory cytokine production.
- Rutin: Inhibits $NF-\kappa B$ and activates the pathway.

The antipyretic/anti-inflammatory activity of flavonoids depends on their structure. The presence of a C2-C3 double bond in the C-ring, a 4-oxo group, and hydroxyl groups at specific positions (particularly $C3'$,$C4'$, in the B-ring and $C5$, $C7$ in the A-ring) were crucial for their inhibitory effects on inflammatory mediators ((Khan et al., 2014, Al-Khayri et al., 2022, Mahmud et al., 2023, Nurdiani and Dika, 2023, Tegegne et al., 2023).

In general, during this evaluation of antipyretic activity of extracts, solvent fractions and crude polysaccharides from *Alstonia congensis* root bark and AsprinAspirin reference product, it was assumed that all test samples were capable to reduce significantly rectal temperturestemperatures caused by the toxican Brewer's yeast in all treated mice with pyrexia with different magnititudes compared to test toxic negative control (DW + Bremers's yeast). But, the last recorded rectal tempertures were very significant and determinative since they were found in acceptable physilogical limits indicating in the some time that these test samples exerted antipyretic activity considered as good and appreciable as already mentioned above.

This study showed clearly that that extracts, solvent fractions and crude polysaccharides from this medicinal plant were endowed with antipyretic effect which can be exploited in traditional medicine and supporting and justifying the use mainly of aqueous extract Ac-1 extract for the treatment of fever in traditional medicine based on its empirical use scientifically demonstrated in the present study as the affordability, accessibility, safety, tolerability and relative lack of toxicity showing that medicinal plant species weare gradually becoming an available and important substitute or alternative agents to known synthetic antipyretic drugs because they contained various secondary metabolites endowed with various biological activities among antipyretic in which, they can react alone or in synergistic manner to increase thise activity as previously also reported by Walter et al., 2016 and Jyothi et al., 2024, and medicinal plants were known to be devoid with tocictoxic effects.

Neverteless, several medicinal plants comprised wide diversity of active secondary metabolites acting either alone or in synergistic manner with few or no toxic effects, in the treating of a variety of human illenesses including fever or pyrexia (Blomqvist and Engblon, 2018).

The presence of active metabolites such as alkaloids, flavonoids, steroids, terpenoids, polysaccharides, tannins and saponins in *Alstonia congensis* root bark (Nsaka et al., 2013, 2017) played a considerable and significant role in the manifestation of antipyretic activity of the extracts and solvent fractions. As it was known, steroids and terpenoids exerted their antipyretic effect through inhibiting the activity of prostanglandinprostaglandin synthetase, enzyme that stimulated the production and release of PGs, the primary mediator in fever induction. Flavonoïdes inhibited elevation body températurestemperatures by suppression médiators like PGs responsibe for fever through its effect towards the release of arachidonic acid (AA) by interfering with the eicosanoid biosynthetis pathway involved in fever production (Mbirinda et aL ;, 2016, Rakib et al., 2020, Yimer et al., 2021).

In general, our findings for the evaluation of antipyretic activity of extracts, solvent fractions and crude polysaccharides from *A. congensis* root bark were in good concodanceconcordance with other resutls previously reported in other studies on the same biological activity conducted with extracts and solvent fractions from various other medicinal plant species (Anowi et al., 2015, Yimer et al., 2021, Tegegne et al., 2023, Tripathy and Mishra, 22023, Jyothi et al., 2023, Gunarti et al., 2025).

3.2. Antipyretic activity of isolated constituent from *A. congensis* root bark

All isolated compounds were identified by NMR and mass spectrophotometric methods and measuring their opical rotation. Thus, compound 1 was identified as β-Amyrin, since its 1H and 13CNMR data were in good agreement with the assignments reported for β-Amyrin, a common terpene by Mahato and Kundu, (1994) with opitical rotation of +18.5, c 0.5 MeOH, m/z : 171.10 and a Na+- adduct at m/z193.08. Compound 2 and 3 were identified as Booenin and Echitamine respectvelyrespectively showing complete 1H and 13CNMR spectral data in total agreement with those reported by Marini-bettolo et al., (1983) and Keawpraduub et al., 1994) respectively. Their strucuresstructures were confirmed by accurate mass measurements. Compound 4 was identified as 6,7 Seco-angustilobine showing recorded ^{1}H and $^{13}CNMR$ spectral data in total agreement with those reported by Yamauchi et al., (1990) and by accurate mass measurement with a protonated molecular ion at m/z 341.30.

This was the first report of Boonein from *A. congensis*. Both alkaloids have been isolated before from the leaves, root or stem bark of *A. congensis* (Monsiuer and Van Bever, 1955, Caron et al., 1989) and in other *Alstonia* species like *A. glauscens* (Keawpradup et al., 1994), *A. boonei* (Adotey et al., 2012) and *A. scholaris* (Yamauchi et al., 1990, Kam et al., 1997). β-Amyrin was an ubiquitous triterpene isolated from many other medicinal plant species (Mahato and Kundu, 1994) as well as from *A. congensis* (Monsieur and Van Bever, 1955) and *A. boonei* (Okoye et al ;, 2014).

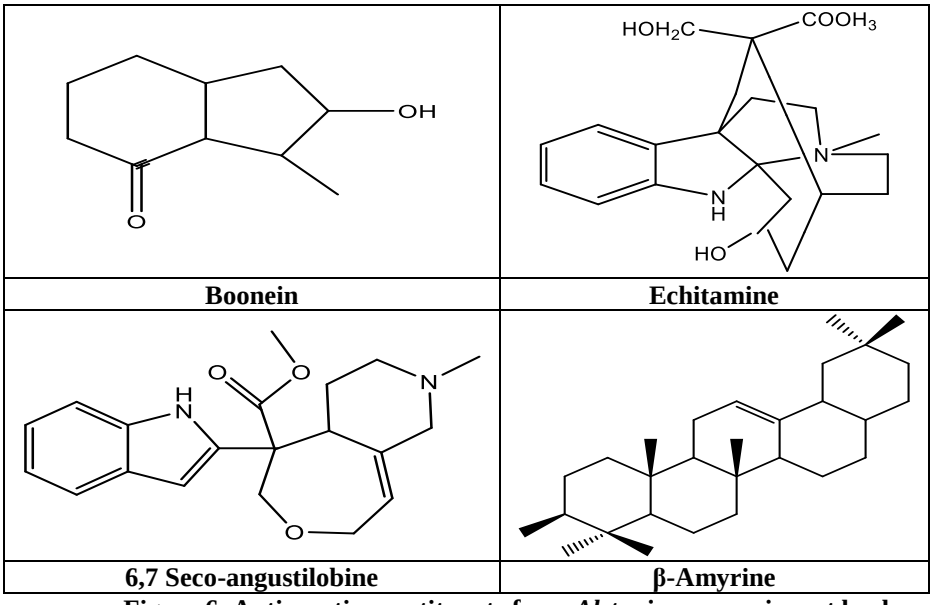
Isolated constituents from *A. congensis* root bark were tested *in vivo* for their potential antipyretic activity against the toxican Brewer's yeats induced high rectal temperatures in animals. Results revealed that these isolated compounds exhibited good and interesting antipyretic activity by decreasing significantly rectal temepraturestemperatures in dose-dependent manner (Table 2). Administered at high oral dose of 100 mg/kg bw, Boonein displayed rectal temperatures to 38.2±0.6 in 1h and continued to be reduced remarkably to 37.5±0.6°C after 5h.

Table 2: Rectal temperatures induced by isolated compounds against Brewer's yeast treated Swiss albino mice.

Group	Dose	Initial temperature	1h	3h	5h
N. control I	DW	36.7±0.04	38.0±0.03	39.0±0.06	36.8±0.03
Toxic N. control BY II	DW+Y	37.8±0.8	39.9±0.9	38.1±0.6	39.8±0.5
Aspirin III	100	37.5±0.4	37.5±0.2	36.8±0.6	34.6±0.8
Boonein	50	37.5±0.5	37.3±0.5	38.6±0.4	37.0±0.6
XI	100	37.2±0.4	38.2±0.6	37.4±0.6	37.5±0.6
Echitamine	50	36.7±0.6	37.3±0.5	38.5±0.4	36.8±0.7
XII	100	36.5±0.2	37.0±0.6	38.1±0.5	37.3±0.5
6,7 Seco-angustilobine	50	36.7±0.5	39.0±0.5	36.7±0.4	37.5±0.7
XIII	100	36.5.±0.5	37.6±0.6	36.5±0.2	36.5±0.6
β- Amyrin	50	37.4±0.5	37.5±0.2	37.1±0.4	37.5±0.7
XIV	100	37.2±0.4	38.2±0.7	36.7±0.5	36.8±0.6

N. contol: normal negative control, Toxic N. control BY : toxic negative control Brewer's yeast.

Administered at high oral dose of 100 mg/kg bw, Boonein displayed rectal temperatures to 38.2±0.6 in 1h and continued to be reduced remarkably to 37.5±0.6°C after 5h. It was the same for the alkaloids Echitamine and 6,7 Seo-angustilobine exhibiting antipyretic activity exerting the same effect by producing significant decliine of rectal temperatures to values of 37.0±0.6 and 38.6±06°C on 1h and 37.3±0.5 and 36.5±0.6°C after 5h respectively. The terpenoid β-Amyrin acted the same manner in notifying rectal temperatures of 38.2±0.7 and 36.8±0.6°C at the same times.



Boonein | **Echitamine**

6,7 Seco-angustilobine | **β-Amyrine**

Figure 6; Antipyretic constituents from *Alstonia congensis* root bark.

It was the same for the alkaloids Echitamine and 6,7 Seo-angustilobine exhibiting antipyretic activity exerting the same effect by producing significant decline of rectal temperatures to values of 37.0±0.6 and 38.6±06°C on 1h and 37.3±0.5 and 36.5±0.6°C after 5h respectively. The terpenoid β-Amyrin acted the same manner in notifying rectal temperatures of 38.2±0.7 and 36.8±0.6°C at the same times.

Unfortunatelly, all isolated compounds displayed high rectal temperturestemperatures at different oral doses in same hours having no important consequence since these high rectal temperatures falled immediately in normal limits in follwingfollowing fours.

In general, as already mentioned above, the last temperaures presented by these isolated compounds remained important, significant and determinative since they remained in acceptable physiological limits of 36.5-37.5°C showing well their antipyretic effect by lowering exerted by these isolated compounds on high rectal temperatures caused by the toxicant Brewer's yeast injection. These last results demonstrated clearly without ambiguity that these isolated compounds from *A. congensis* root bark were endowed with good and interesting antipyretic effects and would act by the same mechanims of action already evoked above.

4. Histological examination

Futhermore, the hispatrological examination of the vitlalvital organs such as liver, heart, lungs, kidney and pancreas revealed that liver, kidney and pancreas showed inflating or swelling. while heart and lungs presented a contracting or narrowing. Fortunatelly, the treatment with aqueous Ac-1 extract of *Alstonia congensis* at the high oral doses of 400 mg/kg bw, was noteworthy in a dose-dependent manner reduced or prevented considerably these severe anomalies and which had disappeared with the time treatment and all vital organs recovered rapidly their normal architecture compared to test normal negative control. Hence, this aqueous extracts Ac-1 from *Alstonia congensis* showed obviously the protection of these vital organs due probably to its antioxidant activity already evaluated (Cimanga et al., 2025).

5. CONCLUSION

The present study reported the for the first time the antipyretic effects *in vivo* of aqueous and methanol 80% extracts, solvent fractions chloroform, ethylacetate, *n*-butanol and residual aqueous soluble fractions as well as crude polysaccharides and isolated compounds from *A. congensis* root bark. Results revealed that all test samples exhibited good and interesting antipyretic activities. The present study provided scientific bases or evidences in favor of the pharmacological use of *Alstonia congensis* root bark in traditional medicine in for aqueous extract (decoction) as an effective alternative in the treatment of fever. The study's overall findings suggested that extracts, solvent fractions, crude polysaccharides extracts and isolated coumpounds from *A. congensis* root bark could be considered as a new source for the development of new plant-based antipyretic agents since these tested samples demonstrated significant and interesting antipyretic activity on Brewer's yeast-induced pyrexia in animal model.

REFERENCEE

1. Adbidoye EO, Aplakah I. Antinociceptive and antipyretic properties of ethanol extract of *Oryza bathii* (Olaceae) in Wistar rats. Sosoko J Vet Sci., 2017; 15(2): 43-48.
2. Adesokan A, Yakub MT, Owoyele BV, Akanji MA, Soladoye AO, Lawal K.,. Effect of administration of aqueous and ethanolic extracts of *Enantia chlorantha* stem bark on brewer's yeast induced pyresia in rats." Afr. J. Biochem. Res., 2008; 2: 165-169.
3. Ahmad I, Khan H, Anwar-Ul-Hassan Gilani AUH, Mohammad Amjad Kamal MA. Potential of Plant Alkaloids as Antipyretic Drugs of Future. Curr Drug Metab, 2017; 18(2): 138-144.
4. Al-Khayri JM, Sahana GR, Nagella P, Biljo VJ, Alessa FM, Al-Mssallem MQ. Flavonoids as Potential Anti-Inflammatory Molecules: A Review. Molecules, 2022; *27*(9): 2901. https://doi.org/10.3390/molecules27092901MQ
5. Alkandahri MY, Arfania M, Abriyani E, Ridwanuloh G et al.,. Evaluation of antioxidant and antipyretic effects of ethanolic extract of *Cep-cCepam* leaves (*Castanopsis costata* (Blume) A;DC. J Adv Pharm Educ Res., 2022; 12(3): 107-112.
6. Alkandahri MY, Sadino A, Pamungkas BT, Oktoba Z, Arfania M, Yuniarsih N et al,. Pharmacological evaluation of anti-inflammatory, antipyretic, analgesic and antioxidant activities of *Castanopsis costataca* leaf fractions (water, ethyl acetate and n-hexane fractions): the potential medicinal plants from North Sumatra, Indonesia. Res Pharm Sci., 2024; 19(3): 251-266.
7. Akpa C, Kehinde AO, Ojewuyi OB, Olayemi J Antipyretic activity of abutilon mauritianum (Jacq.) roots in wistar rats. Int J Pharma Scid Res., 2014; 5: 42-46.
8. Anowi CC, Onyekaba TU, Awemu G, Fadeyi O. Antipyretic, phytochemical and toxicological investigations of ethanol extract of the leaves of *Baphia pubescens* Hook. F (FAMILY/leguminosae). Int J pharm Phytoparmacol Res., 2015; 5(2): 2-8.
9. Anonymous. Analgesic & antipyretic drugs, 2018. http://www.vxldrugs.com/analgesicantipyretic-drugs.html.
10. Asante D, Henneh IT, Acheampong DO, Kyei F, Adokoh CK, Ofori, EG, Ameyaw EO. Antiinflammatory, anti-nociceptive and antipyretic activity of young and old leaves of *Vernonia amygdalina*. Biomed Pharmacother, 2019; 11: 1187-1203. https://doi.org/10.1016/j.biopha.2018.12.147.

11. Azis, A. Kunyit (*Curcuma domestica* Val) Sebagai Obat Antipiretik', J Ilmu Kedokteran dan Kesehatan, 2019; 6(2): 116–120.
12. Blomqvist A, Engblon D. Neutral Mechanisms of Inflammation-Induced Fever. Neuroscientist, 2018; 24(4): 381-399.
13. Caron C, Graftieaux A, Massiot G, Le Men-Olivier L, Delaude C. Alkaloids from *Alstonia congensis*. Phytocemistry Phytochemistry, 1989; 28: 1241-1244.
14. Chattopadhyay D, Arunachalam G. Ghosh L, Rajendran K, Mandal AB, Bhattacharya SK. Antipyretic activity of *Alstonia macrophylla* Wall ex A. DC: An ethnomedicine of Andaman." Islands. J Pharm. and Pharma. Sci., 2015; 8: 558-564.
15. Cimanga KR., Nsaka Lumpu, S.., Tshodi Ehata, M.., Kikweta Munduku C.., Kambu Kabangu, O., Apers, S.., Vlietinck, A.J., Pieters, L.. Assessment of antibiabetic activity of the 80% methanol extract of the stem bark of *Alstonia congensis* Engl. (Apocynaceae) in Wistar rats. World Journal of Pharmacy and Pharmaceutical Sciences, 2016; 5: 112-134. doi: 10.1016/j.jep.2012.04.038
16. Cimanga KR, Tshodi EM, Nsaka LS, Lami NJ, Vlietinck AJ, Pieters L. *In vitr*o antioxidant activity of extracts, fractions and polysaccharides, and *in vivo* sub-chronic and sub-acute toxicity of lyophilized aqueous extract from *Brucea sumatrana* Roxb. (Simaroubaceae) leaves. World J Pharm Pharm Sci., 2020; 9(8): 159-189.
17. Cimanga KR, Nzinga, Vlietinck AJ, Pieters L. Antidiabetic activity of aqueous extract and its fractions, methanol 8% extract, flavonoids and polysaccharides from *Morinda morindoides* (Baker) Milne-Redhead (Rubiaceae) leaves in experimental animals. Eur J Biomed Pharm Sci., 2021; 8: 183-208.
18. Cimanga KR, Kikweta MC, Tshodi EM, Tona LG, Tuenter R, Vlietinck AJ, Pieterse L. Pharmacological assessment of antispasmodic, antioxidant and free radical scavenging activities of extracts, soluble fractions ands its isolated constituents from *Alstonia congensis* Engl. (Apocynaceae) root bark. European Journal of Pharmaceutical and Medical Rerseach, 2025; 12(4): 695-713.
19. Gege-Adebayo GI, Bassi AS, Igbokwe VU, Shafe MO.. "Antipyretic effect of *Ocimum gratissinum* on brewer's yeast induced fever in Wistar rats." J. Med. Med. Sci., 4: 247-2.
20. Ghauri MA, Iqbal l, Raza A, Hayat U, Atif N, Javeed A. *Iin vivio* anti-inflammatory, antipyretic, analgesic activity and *in vitro* anti-proliferative activity of aqueous methanolic extract of *Euphorbia granulata* Forssk. Futur J pharm Sci., 2021; 7(1): 1-10.
21. Gunarti NS, Alkandahri MY, Wahyuningsih ES, Agustina P, Mursal LLP, Hidayah H, Nurviana V. Evaluation of antipyretic and antioxudantantioxidant activities of ten indigenous medicinal plants of Tirtajaya, Karawang, Regency, West Java, Indonesia. Ind J Pharm Edu Res, 2025; 59(1): 252-263.
22. Hajare SW, Tandan SK, Sarma J, Lal J, Telang, A. G.,. Analgesic and antipyretic activities of *Dalbergia sissoo* leaves." Indian J. Pharmacol, 2000; 32: 357-360.
23. Harborne JB. Phytochemical Methods. A Guide to Modern Techniques of Plant Analysis. Chapman & Hall Ltd. London UK, 1998.
24. Hemalatha K, Satyanarayana D, Subrahmanyam EVS,. Phytochemical Constituents of Root Bark of. Asian J Chem, 2008; 20(7): 5405-5408.
25. Khan IA, Azziz A, Manzoor Z et al.,. Stsudy Study on antipyretic activity of *Rumex vesicarius* leaves extract in albino rabiitsrabbits. Vet Worl, 2014; 7: 1. doi:10.14202/vetworld.2014.44-48.
26. Kolawole T, Dapper DV. Anti-pyretic and anti-inflammatory effects of the methanolic extract of the rind of *Citrullus lanatus* on albino Wistar rats, J Med Plants Res., 10(9): 108-112.
27. Kam et al. Alkaloids from *Alstonia scholaris*. Phytocemistry, 1997; 45: 1303-1305.
28. Jyothi Ch, Hemamalini K, snehaSneha K, Malika P, Keerthy Reddy K, Roja B, Ruksana Md. Phytochemical analysis and antipyretic activity of extract of *Aegele marmelo*s leaves on different models. World J pharm Res, 2024; 13(20): 743-754.