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## Chemistry and pharmacological aspects of Aridanin, a lead compound from *Tetrapleura tetraptera* (Fabaceae)

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**ABSTRACT:** Aridanin, a triterpenoid *N*-acetylglucoside has been obtained from chemical investigations of the root of *Tetrapleura tetraptera*, a well-known seasoning spice from Cameroonian food. The structure of the compound has been established using its NMR data and by comparison of that data with the one reported in the literature. Further evidence has been compiled from literature and discussed on the occurrence, the extraction, isolation and analytical techniques as well as the reported biological activities of aridanin. Based on its potency and its high yield in *Tetrapleura tetraptera* fruits, the compound can be considered as lead compound in the development of medicine for the control of schistosomiasis and bacterial infections caused by the bacterial strains *E. coli*, *E. smartii* and *E. aeruginosa*. This paper sheds light on the compound aridanin which deserves more attention in the development of a new potent drug or on the formulation of *Tetrapleura tetraptera* for effective use by the local population.

## 1. INTRODUCTION

It is well known that medicinal plants are largely used around the world in healthcare system as alternative treatments for several diseases due to their important chemical constituents that constitute an important reservoir of potential leads in the new drug development (Happi et al., 2021; Happi, Nangmo, et al., 2022; Happi, Ntabo, et al., 2022). Our recent report on naturally occurring dimeric triterpenoids outlined that triterpenoids are one of the most distributed class of compounds throughout natural sources (plants, animals, microorganisms) with more than 20,000 known structures sorted to more than 100 different scaffolds (Happi, Ntabo, et al., 2022). Triterpenoids are formed from cyclization of squalene via the intermediate 2,3-oxidosqualene produced in a reaction catalysed by squalene epoxidase. The biosynthetic pathway can lead to the formation of several classes of triterpenoids among which the oleanane-type triterpenoids also called  $\beta$ -amyrin subgroup characterized by its pentacyclic skeleton displaying a double bond at positions 12,13 ( $\Delta^{12,13}$ ) with two methyls at position 20 (C-29 and C-30) (Dewick, 2009; Happi et al., 2018; Jäger et al., 2009). Frequently, those pentacyclic triterpenes are isolated from natural sources as triterpenoid saponin structures consisting to sugar unit(s) connected to an aglycone (sapogenin) which is the triterpenoid moiety.

For instance, these saponins possess carboxylic acid groups at positions 4 (C-23), 17 (C-28) and 20 (C-30) on the aglycone ring system and the sugar residues are usually attached to the 3-OH (Dewick, 2009). The triterpenoid glycosides or saponins are of great interest because of their considerable potential as pharmaceutical and nutraceutical agents. It is well documented that that class of compounds demonstrates many bioactivities including anti-inflammatory, immunomodulatory, hepatoprotective, neuroprotective, anticarcinogenic, hypoglycemic or anti-oxidant activity (Rao & Gurfinkel, 2000).

The plant *Tetrapleura tetraptera* belonging to Fabaceae family is a famous seasoning spice in Cameroon food and also used in traditional medicine by local population for the treatment of several diseases including malaria (Kemigisha et al., 2018). Furthermore, the plant is an important source of saponins which have been isolated by several researchers over the years (Maillard et al., 1992, 1989). During our investigation of the root of *T. tetraptera*, we have isolated a triterpenoid glycoside called aridanin (**1**) (Figure 1) which was reported for the first time from the same plant by Adesina and Reisch (1985). The compound has proven to be an important lead in the development of new drugs with medicinal interest due to its biological activity both as antibiotic and in limiting the transmission of schistosomiasis. This paper aims to provide evidence on occurrence, chemistry and pharmacology of aridanin in addition to our own chemical

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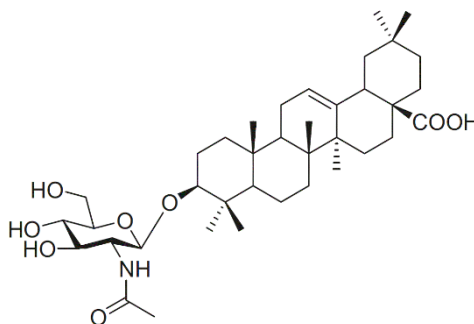


Figure 1. Structure of aridanin

**Table 1**  
Distribution and yield of aridanin isolated from natural sources

Family	Plant	Organ, weight (kg)	Amount of aridanin, yield (w/w)	
		Root, 4.2 kg	21.3 mg (0.0005%)	Current work
Fabaceae	<i>Tetrapleura tetraptera</i>	Root, 300 g	14.7 mg (0.005%)	Noté et al., 2009
		Fruits, 1 kg	1.42 g (0.14%)	adesina1985
Sapindaceae	<i>Paullinia pinnata</i>	Bark, 2.02 kg	170 mg (0.008%)	Lunga 2014a,b
Euphorbiaceae	<i>Manniophyton fulvum</i>	Twigs, 7.1 kg	2.1 mg (0.00003%)	Mbeunkeu et al., 2018
Sapotaceae	<i>Manilkara obovata</i>	Bark, 2.65 kg	8 mg (0.0003%)	Akosung et al., 2021
Meliaceae	<i>Leplaea mayombensis</i>	Seed, 4.3 kg	27.3 mg (0.0006%)	Djeukeu et al., 2021
Moraceae	<i>Ficus exasperata</i>	Root, 4.7 kg	200 mg (0.004%)	Tameye et al., 2020
Ebenaceae	<i>Diospyros conocarpa</i>	Bark, 8.5 kg	30 mg (0.0003%)	Feusso et al., 2016

investigation.

## 2. NATURAL OCCURENCE OF ARIDANIN

The compound aridanin (**1**) has been detected or isolated from several natural sources belonging to the plant kingdom. From the literature survey on the chemistry of aridanin, seven plants from seven plant families have been reported as its main sources. The information in Table 1 shows that the compound has been isolated majorly from fruits of *Tetrapleura tetraptera* with a yield of 0.14% followed by the bark of *Paullinia pinnata*, the root of *T. tetrapleura*, and the root of *Ficus exasperata* with the yield of 0.008%, 0.005% and 0.004%, respectively (Lunga, Qin, et al., 2014; Lunga, Tamokou, et al., 2014; Noté et al., 2009; Tameye et al., 2020).

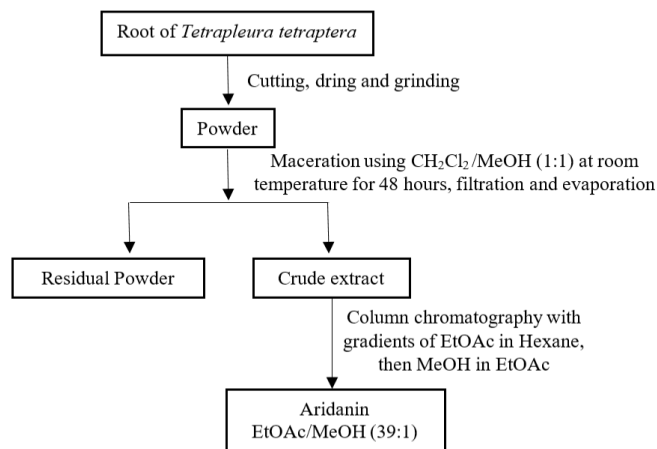
From these data, *Tetrapleura tetraptera* (Fabaceae) represents a rich source of aridanin from which the compound can be obtained with an important yield from its fruits, root or bark. Additionally, the plant from the genus *Paullinia* (Sapindaceae) and *Ficus* (Moraceae) can be also good sources of producing aridanin.

## 3. EXTRACTION AND ISOLATION TECHNIQUES OF ARIDANIN

Aridanin (**1**) is a specialized metabolite isolated from the plant kingdom. Over the two last decades, the compound has been obtained during the purification of plant extracts by several researchers using conventional extraction and separation tools. Basically, the plant material is extracted either by simple maceration in an organic solvent, mostly methanol or the

mixture of dichloromethane-methanol (1:1), or the plant is extracted by soxhlet using methanol to obtain the crude extract which will be further fractionated to obtain sub-fractions with different polarities. Generally, the polar fraction with polarity located between ethylacetate and ethylacetate-methanol (9:1) contains aridanin. Further purification of that sub-fraction using silica gel column chromatography eluting with a gradient of methanol in ethyl acetate or by sephadex column using an isocratic system of chloroform in methanol (2:8 or 3:7) can allow the obtention of the pure aridanin after some additional purification or recrystallization procedures (Akosung et al., 2021; Lunga, Qin, et al., 2014; Mbeunkeu et al., 2018).

During our investigations on *Tetrapleura tetraptera*, the roots were collected at Bafang in the West region of Cameroon in September 2020, identified at the National Herbarium of Cameroon in Yaounde, and then air-dried and grounded to obtain a powder (4.2 kg). Aridanin (**1**) was extracted and isolated following the protocol in Figure 2. Briefly, the powder was extracted at room temperature with methanol for 72 h to obtain 180.1 g of crude extract after the evaporation of the solvent. The partition of the crude extract using flash chromatography with EtOAc, EtOAc/MeOH 3:1 and MeOH gave three subfractions A–C. The subfraction B (EtOAc/MeOH 3:1) was purified with silica gel column chromatography eluting with increasing polarity of methanol in EtOAc to afford aridanin (**1**) (21.3 mg).



**Figure 2.** Extraction and isolation procedure of aridanin

#### 4. ANALYTICAL TECHNIQUES OF ARIDANIN

As commonly reported in natural products chemistry, the compounds are isolated via several chromatographic techniques and characterized based on their spectroscopic and spectrometric data. The triterpenoid aridanin is usually obtained as a white powder from silica gel chromatography at the solvent polarity of ethylacetate-methanol from 2–3% or from sephadex column eluting with dichloromethane/methanol (3/7) (Akosung et al., 2021; Djeukeu et al., 2021; Feusso et al., 2016; Mbeunkeu et al., 2018). It is a lead compound that was firstly and widely isolated from *Tetrapleura tetraptera* fruit and the literature indicated that it melts around 276–280 °C, it is not soluble in water and chloroform but easily dissolves in pyridine, methanol or ethanol (Adesina & Reisch, 1985).

As part of this literature survey on aridanin, we have investigated the root of *T. tetraptera* which led to the isolation of several compounds including the lead aridanin (**1**) obtained as a white powder with a yield of 0.0005% (21.3 mg of aridanin from 4.2 kg of the root). The recorded melting point confirmed the previously reported data of Adesina and Reisch (1985), while its <sup>13</sup>C NMR data (Table 2) was in agreement with the previously reported data in the literature. A comparison of the <sup>13</sup>C NMR confirmed that the structure contains an oleanolic acid moiety indicated by the very close chemical shifts for the carbon atoms in the triterpene core scaffold of aridanin. In addition, the sugar unit displayed close chemical shift values with one in the literature independantly of the frequency or the solvent of the NMR analyses (Table 2). These data are helpful for a quick comparison of spectroscopic data of aridanin in order to confirm the structure of the compounds that might be isolated in future research works.

#### 5. PHARMACOLOGICAL POTENCIES OF ARIDANIN

##### 5.1. Molluscicidal and antischistosomal activities

The works of Adewunmi and collaborators were mainly carried out on the evaluation of the potency of *Tetrapleura tetraptera* extracts and compounds against *Schistosoma man-*

*soni* (the parasite responsible for intestinal schistosomiasis in humans) and its intermediate host *Biomphalaria glabrata* which is a species of freshwater pulmonate snail.

Aridanin was applied at sublethal concentrations (0.25–0.125 ppm) on the glycogen and protein content of *Biomphalaria glabrata*. The results showed that it causes a reduction in the glycogen content of *B. glabrata* but not significantly until 4 weeks of continuous exposure. Therefore, it was assumed that the molluscicides may exert their primary molluscicide action on the carbohydrate metabolism of the snail (Adewunmi et al., 1988). Furthermore, the exposure of 3-day-old *B. glabrata* eggs with aridanin caused a stunning effect on the pre-hatch snails which were less susceptible than juvenile and adult snails. Consequently, aridanin did not exert a significant action as an ovicidal agent in the control of snail intermediate hosts of schistosomiasis (Adewunmi, 1991). At a low concentration (0.25 µg/ml), aridanin decreased the cercariae production by snails already shedding cercariae and therefore the compound can reduce the transmission of schistosomiasis at different stages of the schistosome development (Adewunmi & Furu, 1989).

##### 5.2. Antibacterial activity

Aridanin was tested for its antibacterial activity against a panel of bacterial strains and displayed strong antibacterial potencies, comparable to ciprofloxacin in some cases with MIC value range of 0.78–6.25 µg/ml with the highest potencies recorded against *E. coli*, *E. smartii* and *E. aeroginense* with MIC of 0.781 µg/ml (Lunga, Qin, et al., 2014; Lunga, Tamokou, et al., 2014).

##### 5.3. Cytotoxic activity

Some biological activity of aridanin for its cytotoxicity has been done and reported in the literature. Hence, the works of Tameye et al. (2020) showed that aridanin induced a very strong cytotoxic activity on L5178Y mouse lymphoma cells line, with a viability percentage of 3.5% at 10 µg/mL. Furthermore, within the same year, Mbaveng et al. (2020) performed the evaluation of the cytotoxicity of aridanin on 18 cancer cell lines using the resazurin reduction assay as well as its mechanisms of action. From the results obtained, it is reported that aridanin inhibited the proliferation of all the 18 cancer cell lines with IC<sub>50</sub> values changing from 3.18 µM against CCRF-CEM cells to 9.56 µM against HepG2 cells. More interestingly, the compound displayed an IC<sub>50</sub> value lower than the reference doxorubicin against the multidrug-resistant CEM/ADR5000 cells and the melanoma cell lines (MaMel-80a, Mel-2a, MV3, and SKMel-505) and therefore represent a potent candidate in the development of new potent drugs (Mbaveng et al., 2020)

##### 5.4. Trypanocidal and antiviral activities

Aridanin isolated from *Diospyros conocarpa* was tested against *Trypanosoma brucei brucei* strain and for its inhibitory activity of HIV-1 integrase. The results demonstrated that aridanin has a potent trypanocidal potency with IC<sub>50</sub> value of 0.1 µM while

**Table 2**  
Comparison of  $^{13}\text{C}$  NMR data of aridanin and oleanolic acid

Carbon	Oleanolic acid <sup>a)</sup>	Aridanin <sup>b)</sup>	Aridanin <sup>a)</sup>	Aridanin <sup>c)</sup>
1	38.8	38.6	38.5	38.6
2	27.9	26.3	26.2	26.4
3	78.0	89.1	89.1	89.2
4	39.3	39.2	39.1	39.3
5	55.7	55.7	55.7	55.8
6	18.7	18.4	18.4	18.6
7	33.2	33.1	33.2	33.2
8	39.6	39.5	39.6	39.8
9	48.0	47.8	47.9	48.0
10	37.3	36.8	36.9	37.0
11	23.7	23.7	23.6	23.7
12	122.5	122.8	122.3	122.6
13	144.7	145.0	144.7	144.8
14	42.1	42.2	42.1	42.2
15	28.2	28.2	28.1	28.3
16	23.7	23.8	23.7	23.8
17	46.6	46.7	46.6	46.7
18	41.9	41.9	41.9	42.0
19	46.4	46.5	46.4	46.5
20	30.8	30.9	30.9	31.0
21	34.1	34.2	34.1	34.3
22	33.2	33.3	33.2	33.3
23	28.2	28.2	28.1	28.3
24	16.4	16.8	16.9	17.0
25	15.4	15.6	15.4	15.4
26	17.3	17.4	17.3	17.4
27	26.1	26.2	26.1	26.2
28	180.1	179.9	180.1	180.1
29	33.2	33.3	33.2	33.3
30	23.7	23.7	23.7	23.7
1'	-	104.8	104.7	104.8
2'	-	58.1	58.0	58.2
3'	-	76.2	76.0	76.1
4'	-	72.7	72.6	72.8
5'	-	78.1	78.0	78.2
6'	-	62.9	62.9	63.1
N-Ac	-	170.3 23.6	170.2 23.6	170.1 23.4

a) Recorded at 22.6 MHz in pyridin-d<sub>5</sub> (Adesina & Reisch, 1985)

b) Our data recorded at 125 MHz in CD<sub>3</sub>OD

c) Recorded at 75 MHz in pyridin-d<sub>5</sub> (Fouokeng et al., 2019)

the compound moderately inhibited the HIV-1 integrase with an IC<sub>50</sub> of 18.32  $\mu\text{M}$  (Fouokeng et al., 2019).

## 6. CONCLUSION

During our investigation of the root of *T. tetraptera*, we have isolated aridanin, a triterpenoid *N*-acetylglucoside previously reported from the same plant and different other ones belonging to different families. Based on the recorded NMR data, we confirmed the structure of the isolated compound by comparison with the data reported in the literature. Aridanin contains a  $\beta$ -amyrin moiety identifiable by the characteristic  $^{13}\text{C}$  NMR peaks of oleanolic acid in its pentacyclic chain. Furthermore, we found that the fruits

of *Tetrapleura tetraptera* might be an important source of aridanin due to its high yield calculated (0.14%). Further insights have been given on the different isolation and analytical techniques of aridanin from medicinal plants. The compound demonstrated diverse pharmacological potential especially in antibacterial, molluscicidal and antischistosomal. In some cases, it showed a potency comparable to the reference ciprofloxacin in antibacterial assay while aridanin was established to possess an important activity in the control of schistosomiasis both against the parasite *Schistosoma mansoni* and its intermediate host *Biomphalaria glabrata*. Due to its high yield of aridanin, its main lead compound, the fruits of *T. tetraptera* can serve as raw material for the formulation of potent ameliorated traditional



medicine for the local population. However, more in vitro and in vivo biological investigations as well as the studies on the mechanism of action of aridanin are necessary to confirm their therapeutic potential in new drug discovery.

## CONFLICTS OF INTEREST

The authors disclose that they have no competing interests.

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## A. APPENDIX. SUPPLEMENTARY INFORMATION

Supplementary information to this article can be found online at <https://doi.org/10.53365/nrfhh/152273>.

## AUTHOR CONTRIBUTIONS

Research concept and design : Gervais Mouthé Happi; Collection of data : Gervais Mouthé Happi, Virginia Kien Ntabo; Data analysis and interpretation: Klev Gaïtan Sikam, Livine Zemo Meikeu; Writing the article : Gervais Mouthé Happi, Gaïtan Klev Sikam, Virginia Kien Ntabo, Livine Zemo Meikeu; Critical revision and final approval of the article : Jean Duplex Wansi.

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