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Phytochemical Analysis and Isolation of chemical Constituent of the Root Extract of *Aloe Gilbertii Reynolds*

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Abstract: Aloe gilbertii **belonging to the family Liliaceae is used by local people for treatment various disease such as malaria and wound healing. Phytochemical screening of the CH_2Cl_2:CH_3OH (1:1) extract revealed the presence of Alkaloids, Flavonoids and Anthraquinones. Column chromatographic separation of the crude extract led to isolation of one anthraquinone, 1-hydroxy-8-methoxy-3-methylanthracene-9, 10-dione (43). The structure of this compound was identified using spectroscopic techniques (UV, IR and NMR) and comparison with the literature data.**

Keywords: **Liliaceae, *Aloe gilbertii*, Anthraquinone, 1-hydroxy-8-methoxy-3-methylanthracene-9,10-dione.**

1. INTRODUCTION

Medicinal plants have a long history of use in most communities through the world. In Africa the people still consult traditional healers for their health problem. A needless attempt to narrate the history of medicinal plants is widely documented in various pharmacopoeias plant a medical around the world [1].

Medicinal plants are known to provide a rich source of row materials and have been used as traditional treatments for numerous human diseases for thousands of years in Africa, Asia and other part of developing world, particularly those living in rural areas of the developing countries. It’s continued to be used as the primary source of medicine [2].

Medicinal plants have important contributions in the healthcare system of local communities as the main source of medicine for the majority of the rural population. Plants have not only nutritional value but also, in the eyes of the local people, they have medicinal and ritual or magical values [3].The ethno medicinal healing systems vary across cultures. In Ethiopia, there is cultural diversity with various patterns of using the flora [4].

According to the World Health Organization (WHO), more than 3.5 billion people in the developing world rely on medicinal plants as components of their healthcare [5]. The vast majority of people (70-80%) in Africa consult Traditional Medical Practitioners (TMPs) for their health care [6]. Traditional medicine has been brought into focus for meeting the goals of a wider coverage of primary healthcare delivery, not only in Africa but also, in all countries of the world. It is the first choice healthcare treatment for at least 80% of Africans who suffer from high fever and other common ailments [7]. Thus, medicinal plants are widely used in the treatment of numerous human and livestock diseases in different parts of the world.

Ethiopians have used traditional medicines for many centuries, the use of which has become an integral part of the different cultures in Ethiopia. The indigenous peoples of different localities in the country have developed their own specific knowledge of plant resource uses, management and conservation [8].

A systematic study of a crude drug from medicinal plants embraces through consideration of primary and secondary metabolites derived as a result of plant metabolism. The compounds that are responsible for medicinal properties of the drug are usually secondary metabolites such as flavonoid,anthraquinone,alkaloids,saponnins, tannins, terpenoides and their derivatives

1.1. The genus of *Aloe*

The large and heterogenous family, Liliaceae s. laf. Comprises over 250 genera with 3700 specie s [9]. Most species classified under this family, are perennial herbs with rhizomes and bulbs. The Liliaceae has recently been divided into other smaller and more homogenous families namely, the Asparagaceae, Alliaceae, Asphodelaceae, Dracaenaceae, Eriospemiaceae and Hyacinthaceae. The Asphodelaceae consists of the sub-families Asphodeloideae and Alooideae. Accordingly the genera Asphodeline, Asphodelus, Bidbine, Bidbinello, Eremriis, Hemiphlacirs, Jodrellia, Paradisea, Sirnethis and Trachandra are placed in the sub-hily Asphodeloideae while *Aloe*, *Gasteria*, *Hmvorthici*, *Lomatophylum* and *Poellnitzia* are placed in the Alooideae [10]. So *Aloe*s are one of members of the Liliaceae family and are mostly succulents with a whorl of elongated, pointed leaves. Reynolds described 314 species in his classic monographs; there are now over 360 accepted species.

Aloe has a long ethno botanical and medicinal history around world. The genus *Aloe* L. comprises approximately 420 species with centers of diversity in southern and east Africa, the Arabian Peninsula and Madagascar [11], Out of which 46 nutraceuticals Aloe species are indigenous to Ethiopia. The genus is an important source of biologically active compounds with well over 130 phytoconstituents isolated from the group [12]. *Aloe* is one of the oldest medicinal plants known to human since prehistorically times. The name was derived from the Arabic "alloeh" meaning "bitter" because of bitter liquid found in the leaves. It is also known as 'lily of the desert' the plant of immortality and the medicine plant with qualities to serve as alternate medicine. *Aloe* is as old civilization and throughout history. It has been used as a popular folk medicine. It is present in the arid regions of India and is believed to be effective in treating stomach ailments, gastrointestinal problems, skin diseases, constipation for radiation injury, for its anti inflammatory effect, for wound healing and burns, as an antiulcer and diabetes. Currently the plant is widely used in skin care, cosmetics and as nutraceuticals. It is one of the traditionally used as folk remedies.

1.1.1 Botanical description of *Aloe gilbertii*

Aloe gilbertii Usually a succulent shrub, stem erect to 22-200 x 4.9-28 cm long, occasionally stem less. Leaves crowded, 25-67 x 4-13 cm, canaliculated, recurved towards tip, dark green or glaucose, often flushed brown above and below; marginal teeth 5-10 per 10 cm, 3-5 mm high, tips brown, dried sap color brown. Inflorescence up to 124 cm high, compoundly branched; racemes (9-)15-49 or more, cylindrical 6-15 cm long, lax, 2-4(-5) flowers per cm. Bracts ovate, ovate acute, 4-7.5 x 2-3 mm. Pedicels 7-12 mm long. Perianth cylindrical to subclavate trigonously indented, 21-30 x 4.5-8 mm, orange to red; outer segments free for 8-11 mm. Capsule oblong, woody and rough at maturiy. Seed irregular to 3-sided, 5.3-9.5 cm in diameter, testa colour grey.

Aloe gilberti is recently described and is endemic in the flora area. It occurs in a number of places in Sidamo, Shewa, and Gamo Gofa floristic regions. In these regions it is confined in its distribution to rift valley and lowlands. In Sidamo floristic region, the species occur in Alamura Hill, 4-5 km from Awassa town on the road to Dilla (type locality) and on Tabor mountain slopes. It also grows in a large number from Morocho on the road to Billate. In Shewa floristic region the species occurs in different places near Arsi-Negelle, near Langano up to 30 kms northward from Shashemene on the road to Addis Ababa. South of Shashemene it also grows at 5, 20 and 60 kms (near Alaba town). In Gamo Gofa, it occurs at 3 km from Konso on the road to Yavello. In these localities, the species grows in Acacia woodland, often found in hedges and along field margins on rocky places with Adenia venenata [13].

A. gilbertii is a succulent shrub, grouped together with other shrubby (caulescent) *Aloes* such as *A. calidophila* and *A. megalacantha* in the south and eastern Ethiopia respectively. The species is one of the endemic *Aloe* species in Ethiopia; and mainly characterized by erect, ascending or sprawling stems and distinguished from related species by the cylindrical to sub-clavate perianth (flower). The specific epithet '*gilbertii*', was given in honor of one of the collectors of the type specimen, an outstanding botanist who has contributed importantly to the progress of the knowledge of the Ethiopian Flora [14].

Figure 1: Picture of *A. gilbetii* species (taken by Nebiyu Chali, Dec., 2014 from Alamura Hill)

1.1.2. Medicinal use of *Aloe gilbertii*

The different parts of *Aloe gilberti* are used for the treatment of various diseases in traditional or folk remedies throughout the world. The other uses of *A. gilbertii* by Alaba community are as sources of traditional medicine and for fuel. Leaves and root parts have been used by local people mainly for treatment of malaria and wounds. Dried and dead plant body and parts such as stem and leaves are also used as fuel wood. Four other plant species such as *Accacia saligina*, *Agave sesalensis*, *Hypernia species* and *Accacia senegalensis* were mentioned by local community that has been used alone and in the combination with *A. gilbertii* species for degraded land rehabilitation [15].

1.2 Justification of the research

Except recently published by Daniel et al (2014), there are no prior phytochemical studies conducted on *Aloe gilberti.* But there are several research reports on the phytochemistry and biological activity of the genus *Aloe* in general. Considering the wide use of the plant by local community living around Arsi Nagele, Sidama and Alaba treatment of malaria and wounds, this project was designed so as to investigate the chemical constituents of the plant.

1.3. Significance of the study

Since the root part of *Aloe gilbertii* is used for the treatment of various diseases, isolation and structural determination is very important to find out biological active compounds. Thus, when isolation of bioactive compounds is accompanied by structural elucidation, it is useful for pharmacological action and allows complete synthesis of novel analogue products. The investigation of secondary metabolites may contribute to a better understanding on the chemical characterization of *Aloe gilbertii*. Moreover, it is also used as a reference for other researchers those are interested on the phytochemical investigation.

1.4 Objectives of the Study

1.4.1 General Objective

The overall objective of this research is extraction, isolation and structural elucidation of the chemical constituents from the roots of *Aloe gilbertii.*

1.4.2 Specific Objectives:

- To isolate compounds from the roots of *Aloe gilbertii*.
- To characterize the structure of the isolated compounds by spectroscopic techniques (IR, UV-Vis and 1D).

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2. EXPERIMENTAL

2.1 Materials and Methods

UV spectrum was measured with GENESY'S spectrometer (200-800 nm) in EtOH at room temperature. IR spectrum was measured with Perkin Elmer BX Infrared spectrometer in the range 4000-400 cm^{-1}. ^{1}H NMR and ^{13}C NMR spectra were recorded on a Bruker Advance 400 MHz spectrometer with TMS as internal standard. Analytical TLC was done on 0.2 mm thick layer silica gel on aluminum plate. The components on TLC were visualized.

2.2 Sample collection and preparation

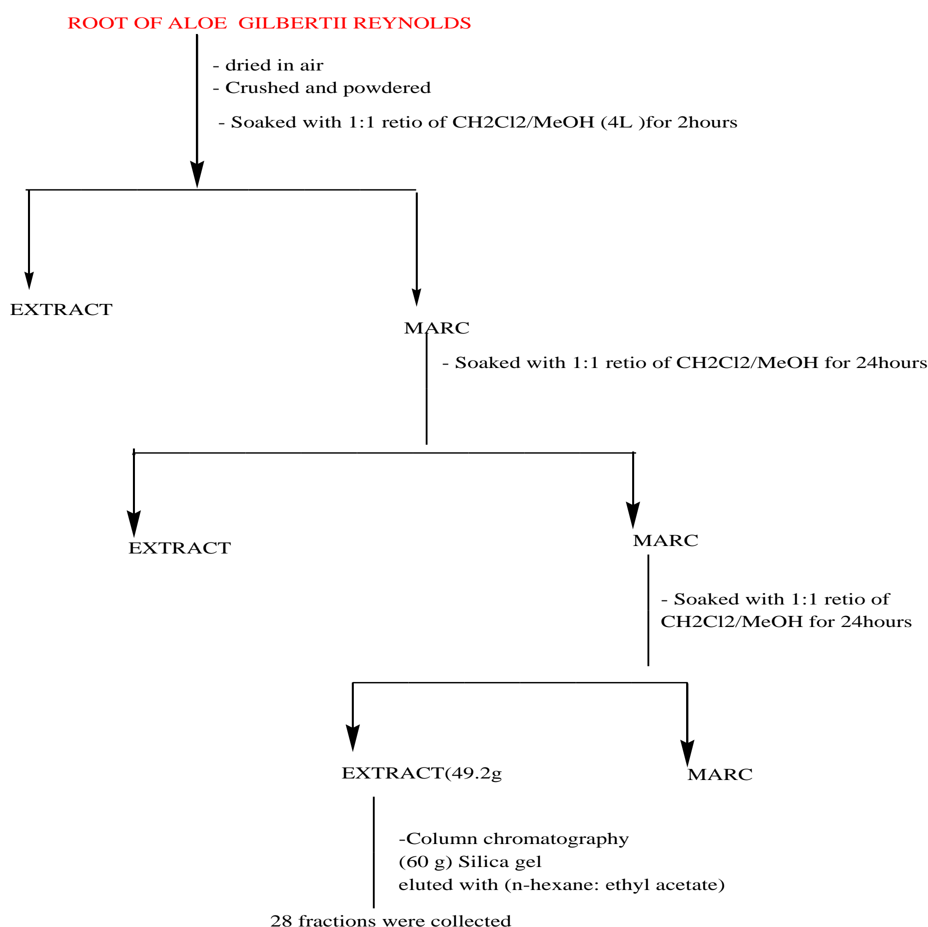
The roots of *Aloe gilbertii Reynolds* were collected in December from Alamura hill, in sidama Zone of SNNPR Ethiopia, about 5km away from Awassa town, 280km away from Addis Ababa. A voucher specimen was identified as *Aloe gilbertii Reynolds* at the National Herbarium of Ethiopia, Department of Biology, Addis Ababa University, and Addis Ababa. The collected plant sample was dried in a shade protected from direct sunlight and then grinded into powder using mortar and pestle. Finally, 500g of the sample was made ready for extraction.

2.3 Coding system

In the coding system of AGR, A stands for the genus name *Aloe*, GR stands for species name *gilbertii* Reynolds, and the number behind AGR indicated the position of the compound starting from the highest to the lowest R_f value. Thus, AGR-1 stands for the first compound.

2.4 Extraction

500 g of the powdered root of *Aloe gilbertii* Reynolds was first soaked with 4L 1:1 ratio of dichloromethane/methanol for 2hr and then with the same solvent ratio for two successive extractions (two times every 24 hr). The mixture was filtered and then the extract was concentrated on rotary evaporator under reduced pressure at $40^{0}C$ to yield 49.2g of black solid as shown in scheme 1.



Scheme 1. General outline for the extraction of *Aloe gilbertii reynolds*

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2.5 Isolation

Slurry was prepared by mixing 200g of silica gel with 400 mL of *n*-hexane and packed into column chromatography. And 30g of the dried dichloromethane/methanol extract was adsorbed on 60g of silica gel using *n*-hexane. Then, the adsorbed sample was applied on the top of the packed column chromatography and eluted with different solvent systems by increasing their polarities. The elution process was started by pure hexane (100%), hexane/ethyl acetate and then pure ethyl acetate (100%) respectively. This successive elution was resulted in 28 fractions as shown in table 1. Out of the 28 fractions (fractions 1-10),(fraction 13,16-17,19,20,23,27-28) were discarded because their TLC results showed unclear spots and more than four spots respectively in different solvent systems.

Fractions 12, 18, and 24-26 each showed the same two pure spots on their TLC results in hexane/ethyl acetate (7:3), hexane/ethyl acetate (1:1) and hexane/ethyl acetate (4:1) solvent systems. Then, these fractions were mixed and concentrated on rotary evaporator and subjected to a small column chromatography for further fractionation using different solvent systems. And a total of 21 fractions were collected as shown in table 2.

Table 1: Solvent systems and column chromatography fractions collected from crude extract

Fraction	Solvent	Ratio	Volume (ml)	Remark
1	*n*-hexane	Pure	50	
2	*n*-hexane/ethyl acetate	99:1	50	
3	*n*-hexane/ethyl acetate	98:2	50	
4	*n*-hexane/ethyl acetate	97:3	50	
5	*n*-hexane/ethyl acetate	96:4	50	
6	*n*-hexane/ethyl acetate	95:5	50	
7	*n*-hexane/ethyl acetate	94:6	50	
8	*n*-hexane/ethyl acetate	93:7	50	
9	*n*-hexane/ethyl acetate	92:8	50	
10	*n*-hexane/ethyl acetate	91:9	50	
11	*n*-hexane/ethyl acetate	90:10	50	
12	*n*-hexane/ethyl acetate	85:15	50	
13	*n*-hexane/ethyl acetate	80:20	50	
14	*n*-hexane/ethyl acetate	75:25	50	
15	*n*-hexane/ethyl acetate	70:30	50	
16	*n*-hexane/ethyl acetate	65:35	50	
17	*n*-hexane/ethyl acetate	60:40	50	
18	*n*-hexane/ethyl acetate	55:45	50	
19	*n*-hexane/ethyl acetate	50:50	50	
20	*n*-hexane/ethyl acetate	45:55	50	
21	*n*-hexane/ethyl acetate	40:60	50	
22	*n*-hexane/ethyl acetate	35:65	50	
23	*n*-hexane/ethyl acetate	30:70	50	
24	*n*-hexane/ethyl acetate	25:75	50	
25	*n*-hexane/ethyl acetate	20:80	50	
26	*n*-hexane/ethyl acetate	15:85	50	
27	*n*-hexane/ethyl acetate	10:90	50	
28	*n*-hexane/ethyl acetate	5:95	50	
29	Ethyl acetate	Pure	50	

Table 2: Solvent systems used in further fractionation of fractions 12,18,24,26 and27

Fractions	Solvent system	Ratio	Volume (ml)	Remark
1	*n*-hexane	Pure	20	
2	*n*-hexane/ethyl acetate	19:1	20	
3	*n*-hexane/ethyl acetate	18:2	20	
4	*n*-hexane/ethyl acetate	17:3	20	
5	*n*-hexane/ethyl acetate	16:4	20	
6	*n*-hexane/ethyl acetate	15:5	20	
7	*n*-hexane/ethyl acetate	14:6	20	
8	*n*-hexane/ethyl acetate	13:7	20	
9	*n*-hexane/ethyl acetate	12:8	20	
10	*n*-hexane/ethyl acetate	11:9	20	AGR-1
11	*n*-hexane/ethyl acetate	1:1	20	
12	*n*-hexane/ethyl acetate	9:11	20	
13	*n*-hexane/ethyl acetate	8:12	20	
14	*n*-hexane/ethyl acetate	7:13	20	
15	*n*-hexane/ethyl acetate	6:14	20	
16	*n*-hexane/ethyl acetate	5:15	20	
17	*n*-hexane/ethyl acetate	4;16	20	
18	*n*-hexane/ethyl acetate	3:17	20	
19	*n*-hexane/ethyl acetate	2:18	20	
20	*n*-hexane/ethyl acetate	1:19	20	
21	ethyl acetate	Pure	20	

According to the TLC results of further fractionation, except fraction 10 all the other fractions were discarded because they did not show pure single spots on TLC .But, fraction 10 was shown a pure single and bright orange spots on TLC having 0.57 Rf value in *n*-hexane/ethyl acetate (7:3) solvent system. Then, it was concentrated on rotary evaporator under reduced pressure at 40 ^{0}C and left in hood for about 48 hours. Finally about 10 mg of yellow solid was obtained and coded as AGR-1. The UV, IR, and 1D NMR (^{1}H NMR) spectra were recorded for compound AGR-1 but ^{13}C and dept were not because of scarcity of sample.

2.6 Phytochemical screening

Chemical tests were carried out for the detection of common secondary metabolites in the crude dichloromethane/methanol (1:1) extracts and powdered root of *Aloe gilbertii* Reynolds using standard procedures described.

2.6.1 Test for Alkaloids

0.5g of plant powder was defatted with 5% ethyl ether for 15min. The defatted sample was extracted for 20min with 5 ml of aqueous HCl on water bath. The mixture was centrifuged for 10 min at 300rpm. 1 ml of the filtrate was treated with few drops of Mayer's reagent and 1 ml with Dragendroff's reagent and turbidity was inspected**.**

2.6.2 Test for Tannins

Small quantity of the crude extract was mixed with water and heated on water bath. The mixture was filtered and small amount of solid $FeCl_3$ was added to the filtrate. Dark-green solution was inspected.

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2.6.3 Test for Antraquinones

0.5g of the crude extract was boiled with concentrated hydrochloric acid for few minutes in water bath and filtered. The filtrate was allowed to cool and equal volume of $CHCl_3$ was added to it. Few drops of ammonia were added to the mixture and heated in water bath. Formation of rose-pink color was inspected.

2.6.4 Test for Saponins

0.2g of the crude extract was shaken with 5 ml of distilled water for 30 minutes and then heated to boil. Appearance of creaming miss of small bubbles (frothing) was inspected.

2.6.5 Test for Terpenoids

0.5g of plant powder was dissolved in 5 mL of methanol. 2 mL of the extract was treated with 1 mL of 2,4-dinitrophenyl hydrazine dissolved in 100 mL of 2M HCl. A yellow-orange coloration was observed as an indication of terpenoids.

2.6.6 Test for Flavonoids

A portion of plant powder was heated with 10 mL of ethyl acetate over a steam bath for 3 min. The mixture was filtered and 4mL of the filtrate was shaken with 1 ml of dilute ammonia solution and a yellow coloration was inspected.

3. RESULTS AND DISCUSSION

3.1 Preliminary phytochemical screening test on methanol/dichloromethane e extract

The results from the phytochemical screening test of the crude dichloromethane/methanol (1:1) extract using different chemicals reagents showed the presence of flavanoids, anthraquinone and Alkaloids (Table 3).

Table 3: Results of phytochemical screening tests of the crude methanol/dichloromethane extract and powder of root of *Aloe gilbertii* Reynolds

Plant Constituent	Reagent used	Present (+) / Absent(-)
Alkaloids	Mayer's reagent + Dragendroff's reagent	+
Tannins	$FeCl_3$	_
Anthraquinones	$HCl + CHCl_3 + NH_3$	+
Saponins	Warming in water bath	_
Terpenoides	2,4-Dinitrophenyl hydrazine	_
Flavonoides	Dilute ammonia solution	+

Chromatographic separation of the dichlormethane/methanol (1:1) extract of roots of *Aloe gilbertii Reynolds* crude yielded one compound, AGR-1. Structural elucidation of the compound was determined based on the spectroscopic data obtained from UV, IR, ^{1}HNMR and comparison with data from literature for similar compound.

3.2 Partial characterization of compound AGR-1

Compound AGR-1 is a yellow solid with Rf value of 0.57 in *n*-hexane/ethylacetae (7:3) solvent system. The UV-Vis spectrum (Appendix-1) absorption band at λmax (in EtOH)

440nm indicate characteristics absorption chromophore center for anthraquinone. The IR spectrum (Appendix-2) showed broad and weak absorption approximately 3400 cm^{-1} indicated the presence of hyrodyl group. The strong absorption band at approximately 2920and 2850cm^{-1} revealed the presence of aliphatic C-H. The strong stretching vibration at 1615 cm^{-1} showed the presence of aromatic functionality in the molecule. The absorption bands at 1715 cm-1 and 1665 $cm\text{-}^{1}$ indicate the absorption of the u*n*-chelated and chelated carbonyl carbon respectively [39].

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The ^{1}H NMR spectrums (Appendi-3) revealed a singlet signal at δ 12.90 integrated to one proton indicate the presence of peri effects with carbonyl carbon attributed to chelated hydroxyl proton resonance attached on C-1 of the aromatic ring. A triplet peak at δ7.64 coupled with a doublet proton at δ δ7.64 indicates the presence of an ABX aromatic pattern on one ring of antraquinone skeleton. Two protons showed a weak coupling not well resolved multiplicity (close to broad singlet) at δ7.32 and 7.28 indicates the aromatic protons on C_4 and C_2 of the other ring of the anthraquinone skeleton. A singlet peak at δ 4.08 and 2.99ppm integrated to three protons indicates the presence of methoxy proton with *peri* effect possibly connected at C-$_8$ position of ring C of anthraquinone and a methyl directly connected to an sp^2 carbon possibly connected to C-3 of ring A of anthraquinone skeleton.

Table 4: Comparison of the observed 1H NMR (400 MHz, CDCl3) spectroscopic data of Compound AGR-1 with the reported value of Chrysophanol (42)

Position	Observed data (in ppm)	Reported data (δ in ppm)
1-OH	12.9 s	12.00 *s*
2-H	7.283 *brs*	7.09 *br s*
3-CH_3	2.90 *s*	2.48 s
4-H	7.32 *brs*	7.29, *dd*, *J*=1.2
5-H	7.78 *dd* (*J*=7.5, 1.2)	7.9 Hz
6-H	7.66 *t*	7.67, *t*
7-H	7.78 *dd* (*J*=8.0, 1.2)	7.81 dd, *J*=1.1, 7.5 Hz
8-OH	-	12.03 *s*
8-OCH_3	4.10 *s*	

The structure of both observed and reported compound was almost approximately the same depending on ^{1}H NMR spectra except substituent on C_8 in which hydroxyl was substituted by methoxy group in our case. The chemical shift of the methoxy group was deshielded to 4.1 due to the *peri* effect with the carbonyl carbon. Thus, based on the observed and reported literature data of ^{1}H NMR compound AGR-1 is 1-hydroxy-8-methoxy-3-methylanthracene-9,10-dione(43) as shown below.

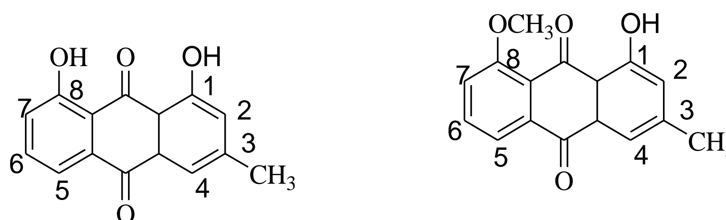

Figure 2: Structure of 1-hydroxy-8-methoxy-3-methylanthracene-9,10-dione and comparison with chrysopanol

3.3 Spectral data of 1-hydroxy-8-methoxy-3-methylanthracene-9, 10-dione

Compound AGR-1 is yellow solid, R_f 0.57 in *n*-hexane-EtoAc (7:3), IR (KBr) Vcm^{-1}: 3400, 2920,2850, 3063,1715, 1665, 1615, 1450,1400,1300,1200, 'H-NMR ($CDCl_3$) at δ 2.9 (3H, s, CH_3), 7.28 (, brs, H-2) 7.80 (2H,*dd* and H-7), 7.65(1H, *brs,* H-4), 7.64 (H, *t*, H-6), 7.80 (2H, *dd* H-5),12.9 (1H, *s*,1 -OH), and 4.08 (1H, *s*,8 –OCH_3).

4. CONCLUSION AND RECOMMENDATION

4.1 Conclusion

In order to promote Ethiopian herbal drugs and traditional use of medicinal plants, there is an urgent need to evaluate the therapeutic potentials of the drugs as per the WHO guidelines. Bioactive extracts should be validated and standardized on the basis of phytochemical constituents.

Results of the phytochemical investigation on the roots of *Aloe gilbertii* revealed the presence of anthraquinone, flavonoids and alkaloids, and absence of terpenoids, tannins, and saponins. Column chromatographic purification of the CH_2Cl_2:CH_3OH (1:1) yielded an anthraquinone characterized to be 1-hydroxy-8-methoxy-3-methylanthracene-9,10-dione(**43**) given semi-systematic name 8-methoxychrysophanol.

Previous phytochemical reports on the genus revealed that the genus has various medicinal uses and hence a couple of phytochemical screening and detail phytochemical analysis have been conducted on the various parts of the plant including the gel and roots. Nevertheless, to the best of our knowledge there were no prior phytochemical screening and phytochemical investigation done of the roots of *Aloe gilbertii* Reynolds. Thus, this works will be used as a reference material to initiate further research works on the roots of the plant so as to support the medicinal uses of the plant traditionally.

4.2 Recommendation

➢ The present study used gravity column using ethyl acetate and *n*-hexane as an eluent. However, as evidenced by the presence of more compound spots in the TLC profile, more phytochemical analysis needs to be carried out on the polar extracts of the plant with the help of reverse phase RP-HPLC (C8 or C18 column) using water/methanol or water/acetonitrile as mobile phase**.** This will help to identify more polar phenolic compounds which are not identified in the present study.

➢ The present study started with small amount of the plant material and could not achieve to isolated and identify some of the minor anthraquinones as supported by the literature reports, Literature reports suggest for the genus as rich source of anthraquinones, Hence, future phytochemical work is recommended starting with higher amount of the plant material and also needs further optimization of purification scale and approach for better result.

➢ *In vitro* bioassay and structural activity relationship (SAR) and has to be done on the crude as well as isolated compounds on various strains of microorganisms so as to establish the traditional uses of the plant.

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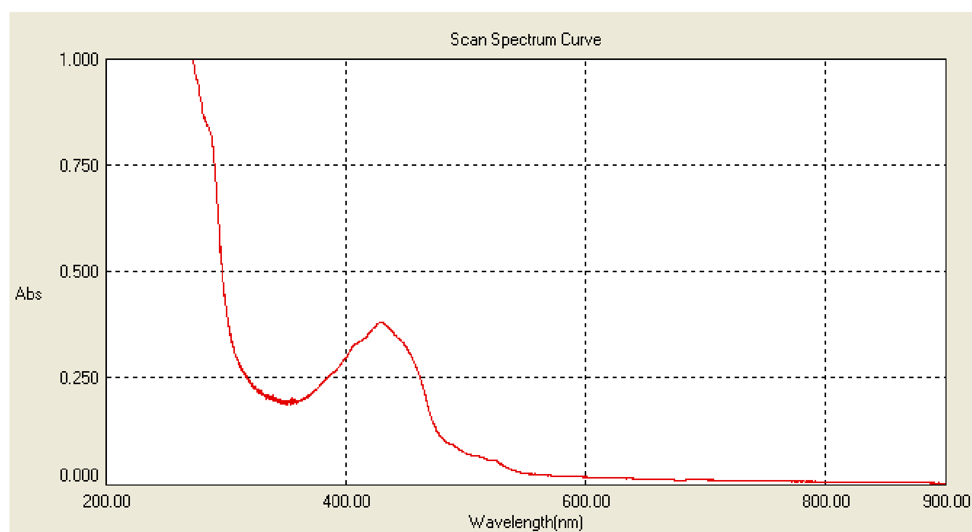
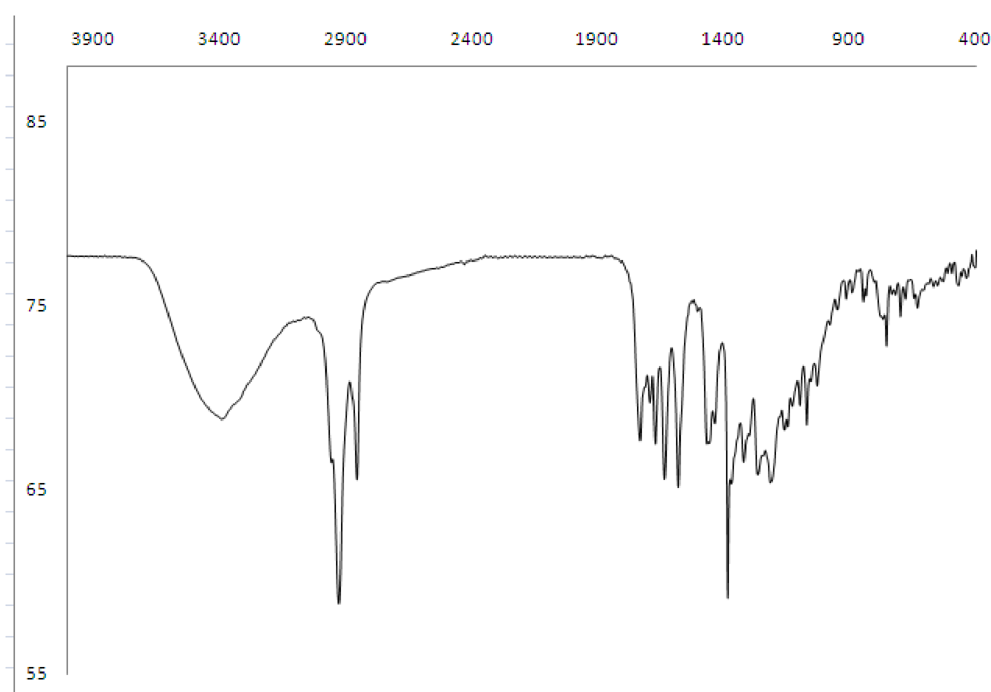
APPENDIX – A

Appendix 1: UV-Vis spectrum of 1-hydroxy-8-methoxy-3-methylanthracene-9,10-dione



Appendix 2: IR spectrum 1-hydroxy-8-methoxy-3-methylanthracene-9,10-dione



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Appendix 3: ^{1}H NMR spectrum of 1-hydroxy-8-methoxy-3-methylanthracene-9,10-dione