

Toxicity of Aquatic Extract of *Acacia Farnesiana*, *Alhagi maurorum* and *Myrtus Communis* Leaf, Stem and Seed against *Physella Acuta* Intermediate Host of Parasites

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ABSTRACT

The recent study aimed to screen suitable plant extracts to control the snail especially which have medical importance. The Molluscicidal activity of leaves, stems, and seeds from *Acacia farnesiana*, *Alhagi maurorum* and *Myrtus communis* against the snail *Physella acuta* was studied. The plant *M. communis* was more toxic than the *A. farnesiana* and *A. maurorum* to the snail *P. acuta*. The stem extract of *M. communis* and *A. farnesiana* was more toxic than leaf and seed. The seed of *A. maurorum* was more toxic than the stem and leaf. LC50 of leaf from *A. farnesiana*, *A. maurorum* and *M. communis* were recorded as 24.1, 38.8, and 6.5 part per million (ppm) respectively. LC50 of the stem were (13, 43.8, and 2.7 ppm) respectively. LC50 of seed were recorded as (15.3, 21.7, and 20.2 ppm) respectively. The study concluded that the plant extracts utilized were have molluscicidal activity and can used to kill the snails.

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INTRODUCTION

The freshwater snail *P. acuta*, is an important intermediate host of many parasites, including *Echinostoma revolutum* can infect birds and mammals, including humans. Also, humans are incidental hosts of the parasite *Parastrongylus cantonensis* (Benelli et al., 2015). The acute bladder snail, *Physella acuta* (Mollusca: Physidae) (synonym *physa acuta*) is globally considered as a highly invasive snail. This species is host to many trematodes causing foodborne diseases in humans (Jayachandran et al., 2022). *P. acuta* is one of the common types distributed in the middle and south of Iraq (Al-Waaly, et al. 2014). The highest frequency of *P. acuta* was recoded in Thi-Qar province, Iraq (Jawad et al., 2022). *P. acuta* snails were studied in Iraq with different subjects such as parasitic infection (Mohammad, 2015), heavy metals accumulation (Al-Warid et al., 2020; Jawad et al., 2022).

Many synthetic compounds have been tested for

their ability to control the snails but none of them was proven to be as entirely satisfactory yet. Therefore, natural sources of novel agents came into play and are considered now attractive alternatives to seek for novel drug leads (Ribeiro et al. 2021; Xing et al. 2021). Screening for less hazardous molluscicides, low cost, and safe to control the aquatic intermediate hosts is still under global consideration (WHO, 2017). Primary target sources are medicinal plants because they are generally a rich source of renewable bioactive organic chemicals, and they may grow well in endemic areas, providing income for local farmers. Furthermore, plant derived molluscicides may have several advantages like low costs, high target specificity, water solubility, high biodegradability, and low toxicity towards normal organs in the human hosts (Singh et al., 2010).

Aquatic extracts of plant origin were used for different purposes in the control of snail subjects. *Acacia farnesiana* (or *Vachellia farnesiana*), and

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previously *Mimosa farnesiana*, known as sweet acacia (family, Fabaceae) is one of these plants (Ismail et al., 2016). *Alhagi maurorum* is one of the plants that possessed antimicrobial activities (Al-Snafi, 2016). A study showed the ability to use the aqueous extracts of leaves of *Myrtus communis* in the biological control of *Lymnaea Auricularia* (Jarallah, 2012). Plant extracts are containing potential substances that give them the ability to kill the snails as containing of *M. communis* more than 50 active ingredients including monoterpene, sesquiterpene, oxygenated sesquiterpenes, tannins and flavonoids (Aleem & Anis, 2021). *Alhagi* species revealed the presence of many substances as fatty acids, sterols, coumarins, alkaloids, vitamins, flavonoids as tamarixtin 3-O-dirhamnoside, isorhamnetin 3-O-glucosylneohesperidoside, isorhamnetine 3-O-robinoside, isorhamnetin 3-Orotinoside, quercetin 3-O rhamnoside, kaempferol 3-O-galactoside, quercetin 3, 7-diglycoside, isorhamnetin 3-rutinoside, daidzein 7,4-dihydroxyisoflavone, calycisin 3-hydroxyformononetin, isorhamnetin and tamarixtin glycones (Ahmad et al., 2015). In addition, the antioxidant, antimicrobial and cytotoxicity activities of *Acacia farnesiana* leaves ethanolic extract were improved (Ramli et al., 2011).

The aim of the current study was to evaluate molluscicidal activity of three plants extracts from each plant three parts include (leaf, stem, and seed) against *P. acuta* snails. Any plant extract wants to be used as a molluscicide, it should be effective at low doses, inexpensive, biodegradable and nontoxic to humans (Singh, et al., 1996; Rao & Singh, 2000). For

Table 1

Concentration of different preparations of leaves, stem, and seeds of *A. farnesiana*, *A. maurorum*, and *M. communis* used in toxicity trials against *P. acuta*

Plant species	Plant part	Test concentrations (ppm)
<i>A. farnesiana</i>	Leaf	0,10,15,20,25
	Stem	0,10,15,20,25
	Seed	0,10,15,20,25
<i>A. maurorum</i>	Leaf	0,25,50,75,100
	Stem	0,25,50,75,100
	Seed	0,4,6,8,10
<i>M. communis</i>	Leaf	0,6,12,5,25,50
	Stem	0,4,6,8,10
	Seed	0,14,16,18,20

Three replicates were used for each read. The mean of snail individuals' number was (8) individuals, for different concentrations. The Standard exposure periods of time were (24, 48, 72, and 96 hours). Dead snails were removed from the experimental Peaker and put in clean water for (24 hours) as a recovery period. Snails unenabled to recovery were considered as mortal. The percentages of observed mortalities

control of water snails, the WHO indicates that the products used should be effective at concentrations lower than 100 ppm (WHO, 1993). Phytochemicals have been shown to have molluscicidal, larvicidal, suicidal, antimicrobial, antiparasitic and ovicidal activities among others (Syombua et al., 2013).

MATERIAL & METHODS

Collection of Snail Samples

Six hundred and sixteen adult *P. acuta* freshwater snails were collected from the internal canal of Baghdad University, Baghdad Capital, Iraq, from March to May of the 2022 year. The collected samples were transported to the laboratory of Microbiology in Tropical Biological Research Unit at the College of Science, University of Baghdad. Samples were acclimatized to laboratory conditions in three class containers measured as (15× 10 × 10 cm), aerated and fed with *Alfa alfa plant* extract for 2 weeks period.

Molluscicidal Activity

Three parts (leaf, stem, and seed) of the three local plants used in recent study (*A. farnesiana*, *A. maurorum* and *M. communis*) were chosen to test the molluscicidal activity. Plant samples were collected according to (Meena, 2020). Plant extract powder of each part for each plant was dissolved in (1 ml) DW. A stock solution was prepared as 1gm and series of concentrations were prepared to calculate the mortality of the snails. The series of concentrations were chosen according to primary experiments (Table 1).

were recorded and the median lethal concentration was calculated by Probit analysis (Stephan, 1977).

Statistical Analysis

Toxicity results were analysed by Probit analysis (Finney, 1971) using BioStat software v2008 and p-value for concentrations comparisons were calculated by SPSS v. 25 for Windows (SPSS Inc. 2008).

Results & Discussion

than the *A. farnesiana* and *A. maurorum* to the snail *P. acuta* (Table 2).

The plant *M. communis* was found to be more toxic

Table 2

Probit analysis among *P. acuta* snail treatments of various extracts concentrations after 96-hour exposure

Probit analysis	A. farnesiana			A. maurorum			M. communis		
	Leaf	Stem	Seed	Leaf	Stem	Seed	Leaf	Stem	Seed
LC10	10.7	-19.2	-1.7	-17.3	-15.1	7.9	2.02	-0.7	8.5
LC16	13.6	-12.1	2.01	-5.03	-2.17	11	3.02	0.01	11.1
LC50	24.1	13	15.3	38.8	43.8	21.7	6.5	2.7	20.2
LC84	34.6	38.2	28.6	82.7	89.9	32.3	10.1	5.4	29.3
LC100	39.8	50.8	35.2	104.6	112.9	37.7	11.9	6.8	33.8
SE-LC50	2.4	3.1	2.8	6.3	16.2	2.1	0.8	1.3	1.3
LCL-LC50	19.1	6.8	9.6	26.2	9.1	17.3	4.7	-0.4	17.6
UCL-LC50	29.1	19.2	20.9	51.4	78.5	26	8.3	5.9	22.8
Beta	0.09	0.03	0.07	0.02	0.02	0.09	0.28	0.36	0.1
Alfa	2.69	4.48	3.84	4.11	4.04	2.97	3.14	3.99	2.77

LC=lethal concentration, SE= standard error, LCL=low concentration level, UCL=upper concentration level

The highest mortality of the snail caused by leaves of *A. farnesiana* was observed in the concentration 25ppm. The highest mortality of the snail caused by stems of *A. farnesiana* was observed in the concentration 25ppm.

The highest mortality of the snail caused by seeds of *A. farnesiana* was observed in the concentration 25ppm (Table3).

Table 3

Mortality of snail's exposure to leaves, stems, and seeds of *A. farnesiana*

Part Extract	N	con	24hr	48hr	72hr	96hr	total
leaves	9	0	0	0	0	0	0
leaves	9	10	1	0	0	0	1
leaves	9	15	0	1	0	0	1
leaves	9	20	1	0	1	0	2
leaves	9	25	1	1	2	2	6
stems	26	0	2	3	0	3	8
stems	26	10	8	2	0	1	11
stems	26	15	6	3	3	3	15
stems	26	20	9	6	0	0	15
stems	26	25	9	5	2	1	18
seed	9	0	0	1	0	1	2
seed	9	10	0	1	0	1	2
seed	9	15	0	1	1	1	3
seed	9	20	1	5	0	0	6
seed	9	25	1	5	2	0	8

The highest mortality of the snail caused by leaves of *A. maurorum* was observed in the concentration 100ppm. The highest mortality of the snail caused by stems of *A. maurorum* was observed in the

concentration 100ppm. The highest mortality of the snail caused by seeds of *A. maurorum* was observed in the concentration 10ppm (Table 4).

Table 4

Mortality of snail's exposure to leaves, stems, and seeds of *A. maurorum*

part extract	N	con	24hr	48hr	72hr	96hr	total
leaves	24	0	0	1	1	2	4
leaves	24	25	4	6	1	1	12
leaves	24	50	6	4	1	0	11
leaves	24	75	5	5	4	5	19
leaves	24	100	11	10	0	2	23
stems	4	0	1	0	0	0	1
stems	4	25	0	1	0	0	1
stems	4	50	0	2	0	0	2
stems	4	75	0	1	1	1	3
stems	4	100	2	1	0	1	4
seed	24	0	0	0	0	0	0
seed	24	4	4	0	0	0	1
seed	24	6	4	0	0	0	1
seed	24	8	4	0	0	0	2
seed	24	10	4	0	0	0	4

The highest mortality of the snail caused by leaves of *M. communis* was observed in the concentration 100ppm. The highest mortality of the snail caused by stems of *M. communis* was observed in the

concentration 100ppm. The highest mortality of the snail caused by seeds of *M. communis* was observed in the concentration 10ppm (Table 5).

Table 5

Mortality of snail's exposure to leaves, stems, and seeds of *M. communis*

part extract	N	con	24hr	48hr	72hr	96hr	total
leaves	4	0	0	0	0	0	0
leaves	4	6	4	0	0	0	1
leaves	4	12.5	4	0	0	0	2
leaves	4	25	4	0	0	0	2
leaves	4	50	4	0	0	0	4
steam	8	0	0	0	0	0	0
steam	8	4	4	4	0	0	1
steam	8	6	4	4	0	0	3
steam	8	8	4	4	0	0	5
steam	8	10	4	4	0	0	8
seed	24	0	0	0	0	0	0
seed	24	14	1	1	1	1	4
seed	24	16	1	2	1	1	5
seed	24	18	2	2	2	4	10
seed	24	20	4	4	4	3	15

The stem extract of *M. communis* and *A. farnesiana* was more toxic than leaf and seed. While in *A.*

maurorum the seed was more toxic than stem and leaf (Figure1).

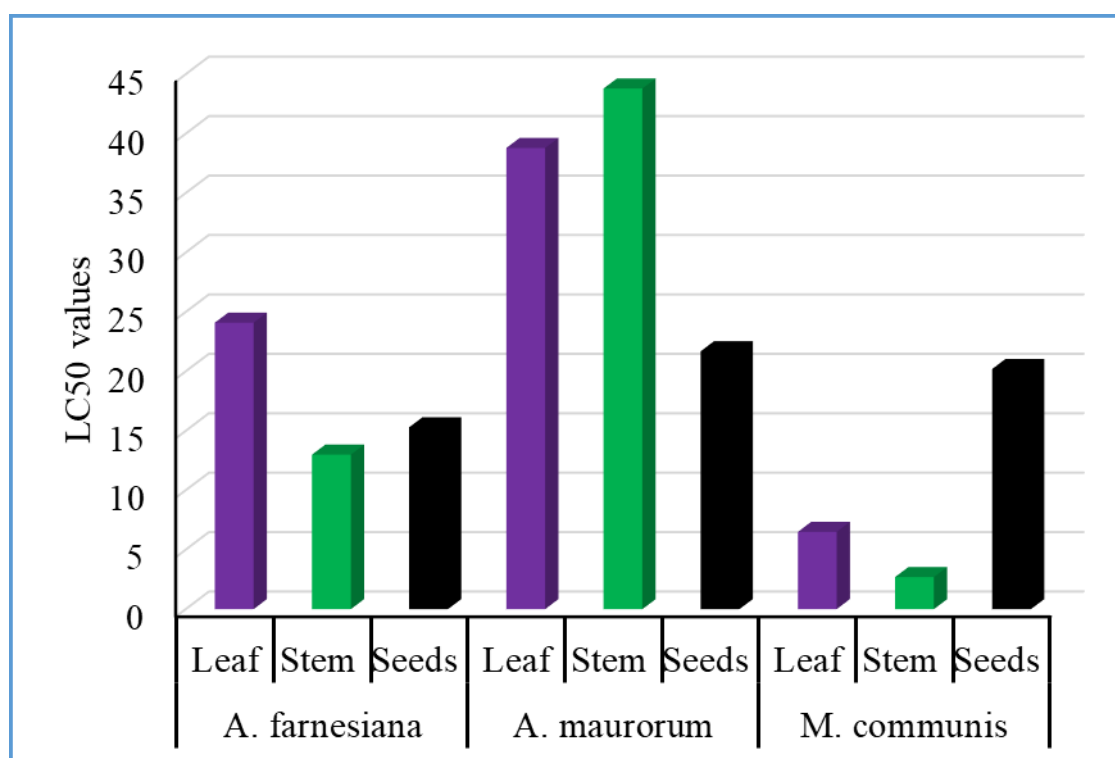


Fig. 1. Comparison among LC50 values of *A. farnesiana*, *A. maurorum* and *M. communis* leaf, stem, and seed against *P. acuta*

The results of a recent study found that the plant *M. communis* was found to be more toxic than the *A. farnesiana* and *A. maurorum* to the snail *P. acuta*. These results agreed with other results improved that the crude water extract and a flavonoid fraction of *M. communis* induced molluscicide effect against the aquatic snail *Biomphalaria glabrata* snail (Deruaz

et al., 1993). Previous studies explain the high activity of the leaves extract of *Myrtus communis* may be due to its containing of many chemical compounds which have biological activity such as myrtol, tannins, bitter principle and phenol compound (uniphenol and manyphenol) and gallic acid (Elisha et. al., 1988; Kery, et. al., 1985). Furthermore, a study

suggests that the mortality in snail was due to *Myrtus communis* characteristics and it contains of medical components as polyphenols, myrtucommulone (MC), semimyrtucommulone (S-MC), 1,8-cineole, α -pinene, myrtenyl acetate, limonene, linalool and α -terpinolene (Alipour et al., 2014). Using of plant extract as molluscicidal to control the snails is continuous in literature. In our pervious study, we used *Nicotinana tobaccum* leaves (Solanaceae) as a molluscicide against the snail *Bulinus truncatus* the intermediate host of urinary Schistosome haematobium in Iraq (Hanoon et. al., 2004).

LC50 values of leaf extracts for the plants *A. farnesiana*, *A. maurorum* and *M. communis* were recorded as (24.1, 38.8, and 6.5) respectively. LC50

values of stem extracts for the plants *A. farnesiana*, *A. maurorum* and *M. communis* were recorded as (13, 43.8, and 2.7) respectively. LC50 values of seed extracts for the plants *A. farnesiana*, *A. maurorum* and *M. communis* were recorded as (15.3, 21.7, and 20.2) respectively (Table 2). Significant differences were noticed among leaf and seed concentrations of *A. farnesiana* plant in their toxic to the snail *P. acuta*, but no significant differences were noticed among stem concentrations of *A. farnesiana* (Table 6). Also, the results of recent study improved that the stem extract of *M. communis* was more toxic than leaf and seed. This may be due to the chemicals found in more concentration in stem than in leaf and seed as 1,8-Cineole, E-b-Ocimene, Linalool, and Oxygenated monoterpenes (Wannes et al., 2010).

Table 6

Comparison of concentrations of leaf, stem, and seed extracts of *A. farnesiana* against *P. acuta*.

Plant species and parts	A. farnesiana		
	Leaf	Stem	Seed
Regression equation	$y=-5.28+3.76*x$	$y=-1.75+1.58*x$	$y=-5.94+4.97*x$
R ²	0.688	0.909	0.913
p-value	0.018	0.061	0.003

Significant differences were noticed among leaf and stem concentrations of *A. maurorum* plant in their toxic to the snail *P. acuta*, but no significant differences were

noticed among seed concentrations of *A. maurorum* (Table 7).

Table 7

Comparison of concentrations of leaf, stem, and seed extracts of *A. maurorum* against *P. acuta*.

Plant species and parts	A. maurorum		
	Leaf	Stem	Seed
Regression equation	$y=-4.14+2.72*x$	$y=-4.58+2.7*x$	$y=-2.99+1.87*x$
R ²	0.695	0.978	0.782
p-value	0.000	0.030	0.132

Significant differences were noticed among leaf, stem, and seed concentrations of *M. communis* plant in

their toxic to the snail *P. acuta* (Table 8).

Table 8

Comparison of concentrations of leaf, stem, and seed extracts of *M. communis* against *P. acuta*.

Plant species and parts	M. communis		
	Leaves	Stem	Seeds
Regression equation	$y=-1.42+1.1*x$	$y=-4.09+4.87*x$	$y=-10.88+8.53*x$
R ²	0.764	1.000	0.931
p-value	0.050	0.001	0.000

The coefficient of determination value of *A. farnesiana* leaf extracts was 0.688, and the mortality in snail individuals explained as 68% due to the variation in concentrations of leaf extracts (Figure 2). The coefficient of determination value of *A. farnesiana* stem extracts was 0.909, the mortality in snail individuals was explained as 90% due to the variation in concentrations of stem extracts (Figure 3). The coefficient of determination value of *A. farnesiana* seed extracts was 0.913, the mortality in snail individuals

explained as 91% due to the variation in concentrations of seed extracts (Figure 4). *A. farnesiana* aquatic extracts also found to has molluscicidal activity against the snail *P. acuta*. These findings were in agree with other findings documented that the genus *Acacia* is rich in polyphenols and tannin has molluscicidal activity (Ismail et al., 2016; Thotathil et al., 2022).

The molluscicide activity of *A. maurorum* (Camel thorn) plant extract was noticed in current work against the snail *P. acuta*. This plant is belonging

to the fabaceae family that produces several vital secondary metabolites like phenolic and flavonoid compounds (Duke 2007, Sultan et al., 2011). Some studies were investigated on *Alhagi* species and it indicated many secondary metabolism compounds like alkaloids, fatty acids, coumarins, and sterols (Kalhor et al., 1997, Awaad et al., 2011 and Verma et al., 2013). Molluscicide activity of aqueous extract of *A. maurorum* due to it contain of compounds as

phenols, 2,3-Dimethylpene-1,4-dicarbonitrile and 1,3-Butanediol, 1,3-butylene glycol, 4-Fluoroveratrole, fluorobenzene, 3,4-methoxy, and tannin compounds (Mostafa et al., 2019). The plant *A. maurorum* seed was found to be more toxic than stem and leaf to *P. acuta* snail. Seed of *A. maurorum* was reported to contain unsaturated fatty acid 88% and a lot of microelements (Lei, 2008).

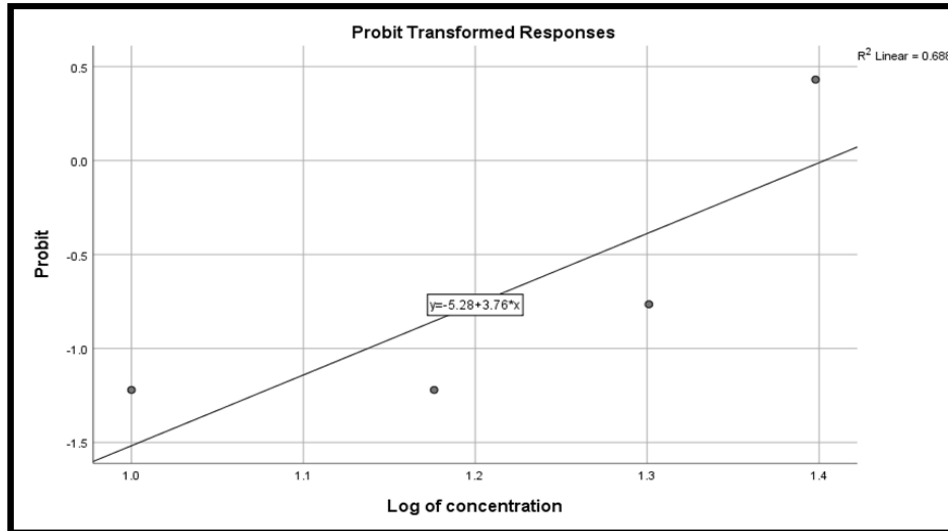


Fig. 2. Regression analysis of *A. farnesiana* leaf extract against the snail *P. acuta*.

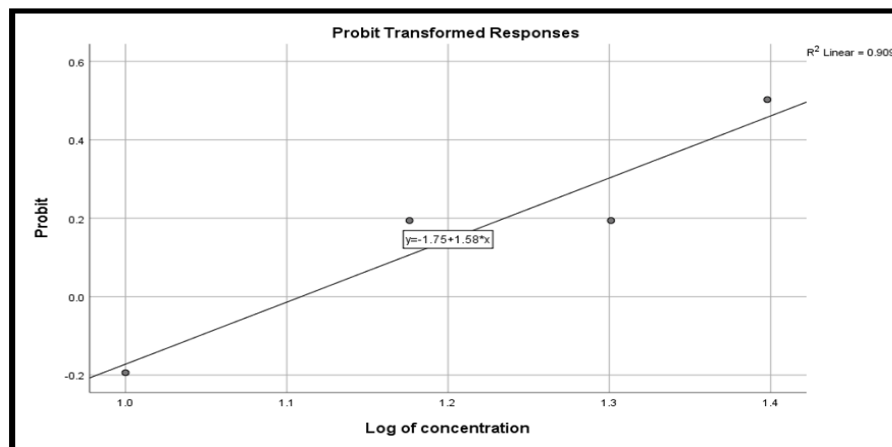


Fig. 3. Regression analysis of *A. farnesiana* stem extract against the snail *P. acuta*.

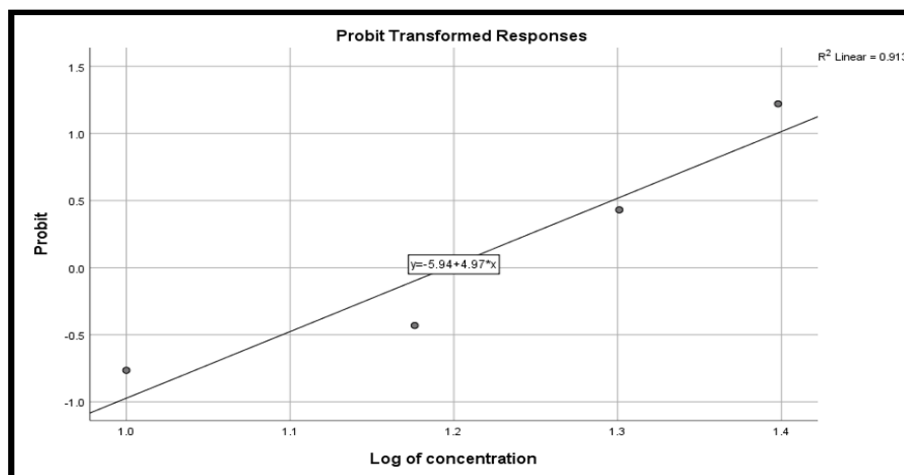


Fig. 4. Regression analysis of *A. farnesiana* seed extract against the snail *P. acuta*.

The coefficient of determination value of *A. maurorum* leaf extracts was 0.695, and the mortality in snail individuals explained as 69% due to the variation in concentrations of leaf extracts (Figure 5). The coefficient of determination value of *A. maurorum* stem extracts was 0.978, the mortality in

snail individuals explained as 97% due to the variation in concentrations of stem extracts (Figure 6). The coefficient of determination value of *A. maurorum* seed extracts was 0.782, the mortality in snail individuals explained as 78% due to the variation in concentrations of seed extracts (Figure 7).

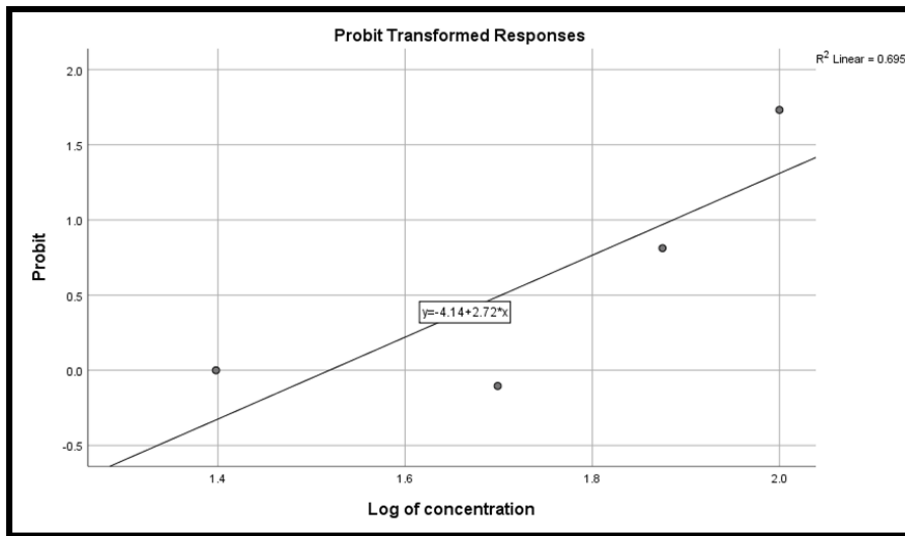


Fig. 5. Regression analysis of *A. maurorum* leaf extract against the snail *P. acuta*.

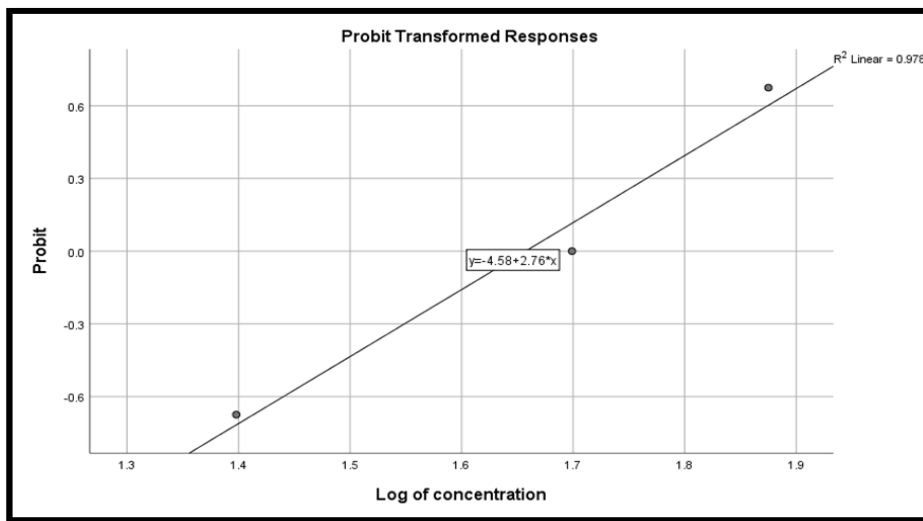


Fig. 6. Regression analysis of *A. maurorum* stem extract against the snail *P. acuta*.

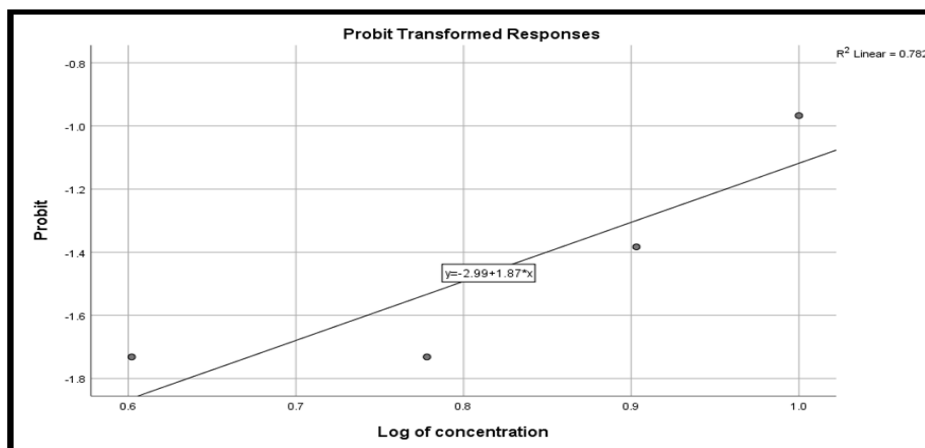


Fig. 7. Regression analysis of *A. maurorum* seed extract against the snail *P. acuta*.

The coefficient of determination value of *M. communis* leaf extracts was 0.764, the mortality in snail individuals explained as 76% due to the variation in concentrations of leaf extracts (Figure 8). The coefficient of determination value of *M. communis* stem extracts was 1.000, the mortality in snail individuals explained as 100% due to the variation in concentrations of stem extracts (Figure 9). The coefficient of determination value of *M. communis* seed extracts was 0.931, the mortality in snail individuals explained as 93% due to the variation in concentrations of seed extracts (Figure

10). The aquatic snail *Biomphalaria glabrata*, which transmits schistosomiasis, was found to be susceptible to molluscicidal activity when a crude water extract and a flavonoid fraction of *Myrtus communis* were combined (Deruaz et al., 1993). Because *Myrtus communis* contains numerous chemical compounds with biological activity, including myrtol, tannins, bitter principle, phenol compound (uniphenol and manyphenol), and galic acid, its leaves extract has a high level of activity (Jarallah, 2012).

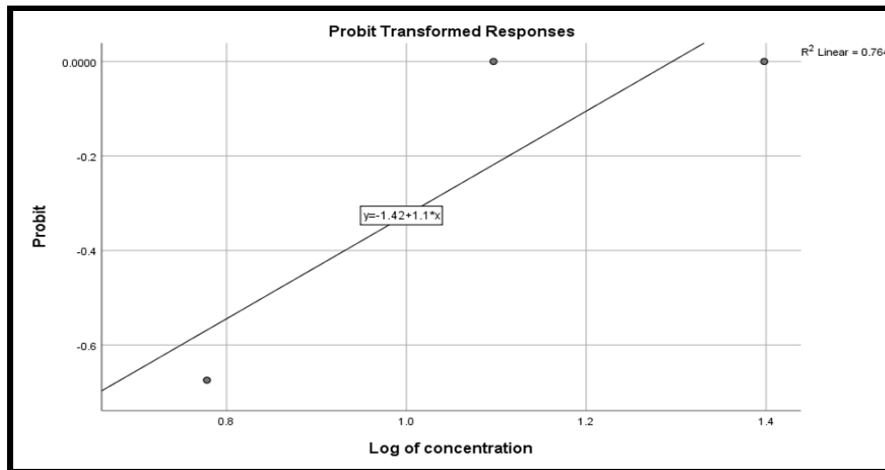


Fig. 8. Regression analysis of *M. communis* leaf extract against the snail *P. acuta*.

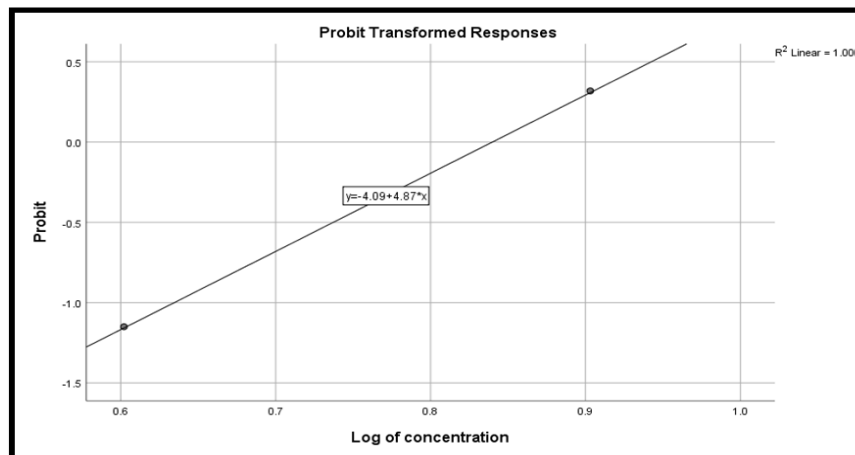


Fig. 9. Regression analysis of *M. communis* stem extract against the snail *P. acuta*.

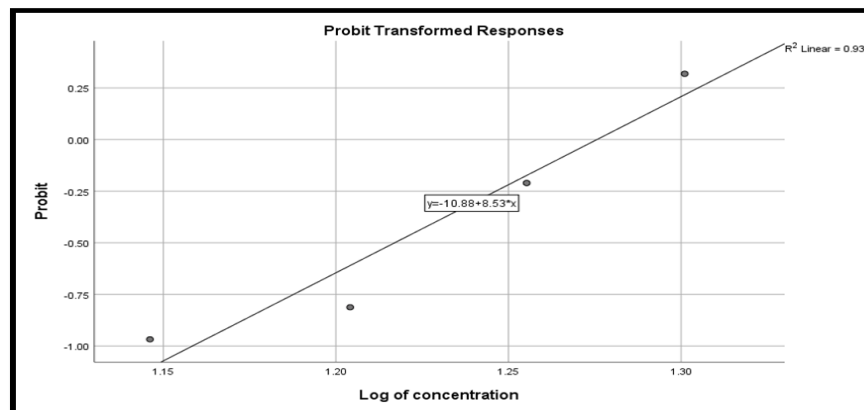


Fig. 10. Regression analysis of *M. communis* seed extract against the snail *P. acuta*.

CONCLUSION

The plant *M. communis* was more toxic than the *A. farnesiana* and *A. maurorum* to the snail *P. acuta*. The stem extract of *M. communis* and *A. farnesiana* was more toxic than leaf and seed. The seed of *A. maurorum* was more toxic than the stem and leaf. LC50 of leaf from *A. farnesiana*, *A. maurorum* and *M. communis* were recorded as 24.1, 38.8, and 6.5 part per million (ppm) respectively. LC50 of the stem were (13, 43.8,

and 2.7 ppm) respectively. LC50 of seed was recorded as (15.3, 21.7, and 20.2 ppm) respectively. The study concluded that the plant extracts utilized were have molluscicidal activity and can used to kill the snails. The recommendations of this study are testing of other plant extracts and studying their toxicity to other species of than snails.

Competing Interest

The authors had no competing interests.

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