

GARCINIA GUMMI-GUTTA (L) ROBS. VAR. GUMMI-GUTTA AS A HERBICIDE

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Article Received on
08 August 2014,

Revised on 05 August 2014,
Accepted on 28 Sept 2014

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ABSTRACT

A study was conducted to evaluate the weedicidal potential of *Garcinia gummi-gutta* leaf leachates on germination of weeds. The effect on seed germination was assayed by Sandwich method. Results revealed that the plant is a strong weedicide. The extract of the plant has also been tested against the mature seedlings grown and brought up in soil media maintained in pots. This has been performed in order to determine the toxicity of the purified compound when used against weeds. Results also revealed that the plant is exerting its allelopathic effect only on the germination stage of seeds whereas a null effect was observed with matured seedlings.

KEY WORDS: allelopathy, secondary metabolites, weeds, allelochemicals, *Garcinia gummi-gutta*, matured seedlings, natural herbicides.

INTRODUCTION

In modern agriculture, most of the natural plant communities are replaced by single crop species or weeds, which can cause ecosystem degradation and biodiversity loss worldwide. Due to the invasive behaviors of these weeds and crops, India is also facing the problem of reduced crop yield and loss of many important plants. A yield decline was recorded up to 40% in agricultural crops while a significant reduction up to 90% has been reported in case of forage production in grassland ^[1]. Thus there arised the need for the elimination of invasive weeds, some microorganisms, viruses, as well as some herbivores. Pesticides and fertilizers not only affect the pests and crops, but also non-pest species, soil, water, food, and also humans ^[2]. The extensive research to elucidate the causes and consequences of the above mentioned threats and also poor establishment and stunted growth of the crops upon growing

the same crop in succeeding years, lead to investigations of possible causes like “Allelopathy”.

Allelopathy is a phenomenon in which the secondary metabolites produced by one plant inhibits the growth and activity of the plants in neighbourhood. A classical definition for allelopathy as given by Rice is that “any direct or indirect effect of one plant on another mediated through the production of chemical compounds that escape into the environment”^[3]. Plants produce an enormous number of chemical substances known as natural products which can affect a wide range of biological activities. These chemicals include primary metabolites and secondary metabolites. Primary metabolites are those which are directly involved in the growth and development of a plant whereas secondary metabolites are the by-products of primary metabolic pathways that do not possess a direct role in the basic metabolism of plants. Secondary metabolites possess a defensive function. Most of them identified as allelochemicals may be important for the defense of plants in situations where several threats or stress factors act simultaneously^[4, 5].

Garcinia gummi-gutta (L.) Robs. Var. *gummi-gutta* formerly known as ‘*Garcinia cambogia*’ is a dicotyledonous tree, belonging to the family Guttiferae [Clusiaceae]. They are commonly known as Kodampuli or Malabar tamarind. The plant is found commonly in evergreen forests of India, Malaysia, and African countries. The Clusiaceae families are a rich source of secondary metabolites. Four major classes of compounds are found in the Clusiaceae family: xanthenes, coumarins, flavonoids, and benzophenones. The plant has been reported to possess various types of secondary metabolites having therapeutic benefits. The fruits of the plant *Garcinia gummi-gutta* are commercially important for its valuable chemical components like hydroxy citric acid, tartaric acid, cambogin, euxanthone, garcinol, reducing sugars and fats. The plant is commercially important as their fruit extracts are used for various treatments such as astringent, demulcent, rheumatism, bowel complaints and purgative^[6]. Ongoing research for anti-malarial drug from *Garcinia* varieties are also reported^[7]. A wide range of studies conducted on the plant gave reports regarding various cytotoxic effects of the plant, of which included antifungal and antibacterial.^[8]

Even though many studies have been conducted by eminent researchers on the plant, *Garcinia gummi-gutta*, only fewer studies have yet been done on its allelopathic effect.. Thus an attempt has been made to determine the allelopathic potential of *Garcinia gummi-gutta*.

The phenomenon of allelopathy can be exploited for the production of natural herbicides which pose little harm to the environment. But before exploiting the phenomenon, it is needful to determine whether the allelopathy exhibited by the plant is harmful to matured seedlings. Thus the effect of the plant leachate on matured seedlings of the receptor crops used has also been performed.

MATERIALS AND METHODS

Plant Material

The plant *Garcinia gummi-gutta* used in the present study were collected from different locations of Kollam district. The collected plant parts were separated from the stem, chopped into small pieces, oven-dried and coarsely powdered by using a pulverizer and were then extracted with methanol. About 25 ml of the crude extract were subjected to purification by means of Normal Phase Column chromatography using silica gel (60-120 mesh size) in step-wise elution with a mixture of chloroform and increasing concentration of methanol. By performing column chromatography, thirty fractions (30 mL each fraction) were collected, tested for activity. To determine the chemistry of the active fractions, colour tests were performed which showed the presence of flavonoids.

Allelopathic Activity Assay using Sandwich Method

Preparation of the Extracts

The flavonoid containing fractions were pooled together and evaporated to dryness. From that, an aliquot of the extract containing a concentration of 100 mg dry weight dissolved in methanol state was evaporated to dryness and was then dissolved in 5ml DMSO which is an inert solvent and will not cause any lethal effects.

For evaluating the weedcidal effect, we selected five weed varieties viz. *Sesamum radiatum*, *Crotalaria juncea*, *Tridax procumbens*, *Cleome burmanii* and *Cassia tora* as receptor plants. The sandwich method consists of the use of a special sixwell multidish in which dry leaves

from the donor plant are placed above a layer of autoclaved agar cooled to 40 °C to 45 °C.

After gelatinizing, the same volume of agar is overlaid, forming a sandwich medium. Twenty five seeds of selected weeds were arranged on sterile agar medium in multi-well agar plates in Petri-dishes at room temperature. Agar substrates alone are used as untreated controls ^[9]. The seeds in Petri dishes were allowed to germinate for six days. Those seeds with visible radicle were considered germinated ^[10]. The counting of the germination of the

seed was done daily from the second day onwards. The length of the stems and roots of the germinated seeds were measured with a ruler. Termination of the experiment was done after three consecutive recordings had shown no further variation. The whole experiment was repeated four times and the average values were expressed.

Calculation of Inhibitory Effect

The percentage of inhibitory effect on germination and growth parameters of treatment plants to control was calculated as per the formula given ^[11].

$$I = \frac{100 - (E_2 \times 100)}{E_1}$$

Where, I = % inhibition; E_1 = response of control plant; E_2 = response of treatment plant.

Effect of The Plant The Growth of Seedlings

To evaluate the detrimental impact of the extract with normally growing plant cells in the neighbourhood of the susceptible crop plants, selected seeds like *Cicer arietinum* Acc1, *Pisum sativum*, *Cicer arietinum* Acc2, *Arachis hypogea* and *Vigna radiata* collected from commercial shops were sown on soil, watered adequately and allowed to grow until it reached a length of about 10-12 cm shoot length. Then for the preparation of leaf extract of the plant, the methanolic extract obtained were concentrated to dryness and the dried mass containing about 4.54g was then dissolved in 100 ml distilled water. It was then sprayed to the soil as well as to the top of the plant and observed for 8-10 days. Parameters like root length, shoot length, dry weight and fresh weight of the seedlings have been recorded. A Control was maintained by spraying distilled water. The whole experiment was repeated four times and the average values were expressed.

RESULTS AND DISCUSSIONS

The compound has been tested against six weed species namely *Sesamum radiatum*, *Crotalaria juncea*, *Tridax procumbens*, *Cleome burmanii* and *Cassia tora*. 25 seeds of each species were sown in each petri-plate. Since previous studies demonstrated that a 100% concentration of *Garcinia gummi-gutta* leaf extract is possessing maximum allelopathic activity, only single concentration of the compound has been applied. Results are shown in Table.1. Here it was observed that of the five species tested, *Crotalaria juncea*, *Cassia tora* and *Tridax procumbens* were found inhibited maximally showing a 100% germination inhibition, whereas an eighty percentage inhibition has been found in *Sesamum radiatum* and

Cleome burmanii. Inhibitory effect of methanolic extract from the leaf of Spanish jasmine on germination and seedling growth of barnyardgrass and wild pea weeds has been studied ^[12]. In the cases that exhibited a reduction in seed germination, there occurred the extrusion of radicle but it got wilted on the second day after germination. Here the seedlings lost their ability to develop normally as a result of reduced radicle elongation and root necrosis. But for control, the radicle extruded continued to grow into a complete plant. The length of root and shoot has been measured and compared with Test plants. The study also revealed that the shoot and root length of those seeds that exhibited inhibited growth were severely impeded as compared to Control. This provides evidence that the allelochemicals from *Garcinia gummi-gutta* had caused reduced weed seed emergence and development of the receptor weedy species used. The capacity of leaf extracts of Mango for killing or suppressing weed growth supports the work ^[13].

Table.1. Weedicidal Effect of the Compound Isolated From *Garcinia Gummi-Gutta*

S. No.	Seeds	Percentage Inhibition of Germination	Root Length(cm)		Shoot Length(cm)	
			C	T	C	T
1	Sesamum radiatum	80.25±2.87	0.45±0.06	0	1.48±0.12	0.48±0.11
2	Crotalaria juncea	96.5±3.12	0.875±0.15	0.112±0.03	2.4±0.16	0.4±0.12
3	Tridax procumbens	99.5±1.10	0.725±0.15	0.11±0.02	3.38±0.13	0.44±0.05
4	Cleome burmanii	80.5±1.29	0.483±0.04	0	2.02±0.06	0.56±0.12
5	Cassia occidentalis	98.75±1.50	0.783±0.11	0.21±0.11	1.87±0.16	0.4±0.12

Fig.1. Effect of the Flavonoids Isolated From The Leaf Extracts of *Garcinia Gummi-Gutta* on Weeds.

1. Sesamum Radiatum



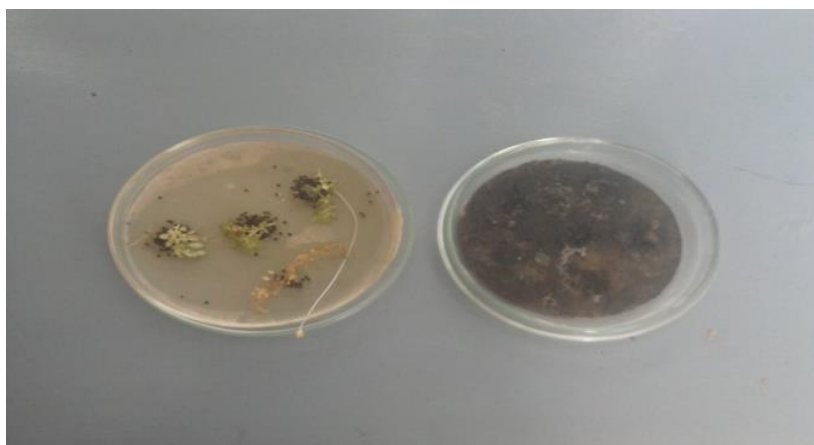
Control Test

2. *Crotalaria Juncea*



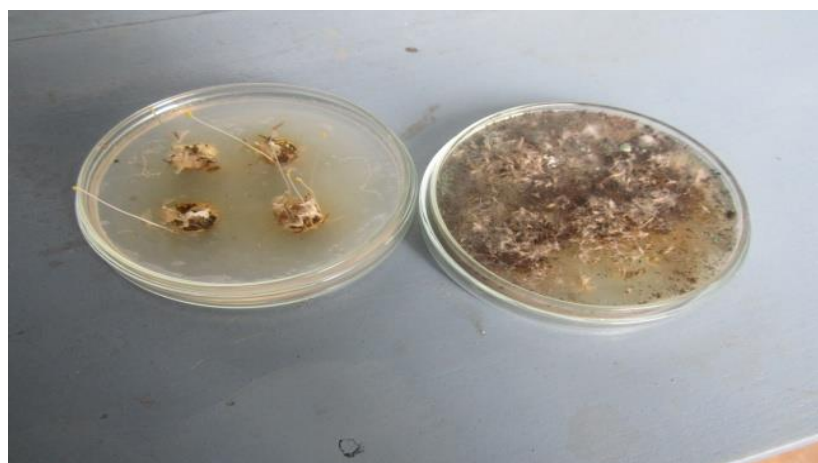
Control Test

3. *Tridax Procumbens*



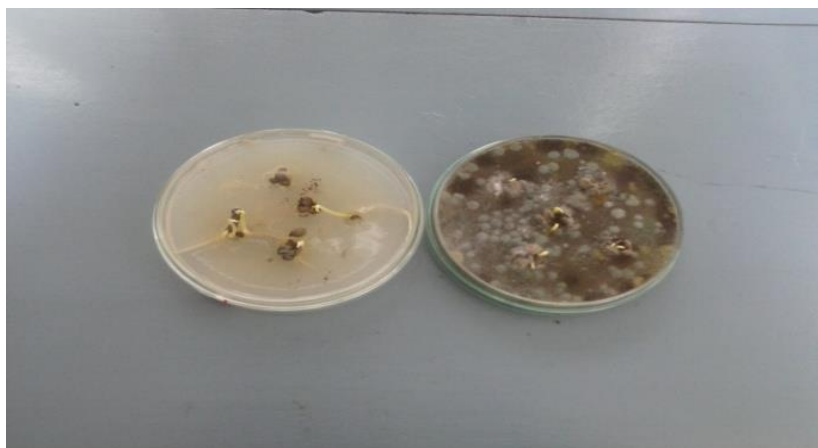
Control Test

4. *Cleome Burmanii*



Control Test

5. Cassia Occidentalis



Control Test

The present investigation clearly confirms that the allelochemicals present in the plant *Garcinia gummi-gutta* can negatively affect the neighboring or succession plants. The effectiveness of the plant on germination and growth of receptor plants suggests that the leaves of *Garcinia gummi-gutta* may act as a source of allelochemicals after being released into soil or after decomposition. Therefore we can suggest that the plant is possessing allelopathic potential. It can also be suggested that it will not be suitable to grow those agricultural crops close to or in fields where the plant *Garcinia gummi-gutta* were grown previously. A study conducted on the effect of leachate from *Accacia xanthophloea* on seed germination and root and shoot elongation gives an indication on how some herbaceous and woody plants would perform when intercropped with this species ^[14]. A reduced germination and radicle length of white mustard [*Sinapis alba* L], when it was grown alongside germinating barley seeds also supports the idea ^[15]. Thus the farmers should be provided adequate information about the allelopathic effects of the plant.

Impact of the Plant Extract on the Growth and Survival of Seedlings

For evaluating the impact of the plant extract on matured crop seedlings, parameters like root length, shoot length, dry weight and fresh weight of the seedlings have been observed (Table.2). Results revealed that the extract is not posing any harm to plants that have already germinated. The receptor plants were found growing normally like that of the control. There occurred a negligible variation in the root length, shoot length, dry weight and fresh weight. The present observation suggests that the plant when used as a herbicide or a weedicide. Thus we can assume that the active principle present in the plant *Garcinia gummi-gutta* when applied as a weedicide / herbicide will not affect the growth of adjacent crops which are

already grown. It should be applied after a time when the sown crops have grown and reached up to a length 2-3cm above the soil. A study conducted on the allelopathic effect of Sorghum on Wheat varieties where a single spray of 5% sorgaab (water extract of mature stalk of *Sorghum bicolor* L. Moench plants obtained after soaking in water for 24 h and sprayed as a natural herbicide) solution when applied for 30 days after sowing increased wheat yields by 14% and suppressed weed biomass by 20–40% supports the work ^[16].

Table 2. Impact of the Plant Extract on the Growth of Seedlings

Sl. No.	Seeds	Root length (cm)		Shoot length (cm)		Fresh weight (mg)		Dry weight (mg)	
		C	T	C	T	C	T	C	T
1	S ₁	7.17±0.16	7.2±0.03	7.78±0.75	7.45±0.16	2.91±0.15	3.11±0.28	1.25±0.44	1.22±0.31
2	S ₂	4.25±1.1	4.32±0.14	8.33±0.23	8.46±0.15	1.54±0.54	1.55±0.21	1.12±0.15	1.34±0.11
3	S ₃	3.85±0.58	3.7±0.16	6.28±0.19	6.03±0.21	1.85±0.36	1.82±0.45	1.26±0.22	1.26±0.26
4	S ₄	3.68±0.25	3.5±0.22	7.45±0.3	7.48±0.09	1.97±0.19	2.01±0.13	1.78±0.36	1.73±0.48
5	S ₅	6.93±0.2	6.5±0.19	15.38±0.81	14.82±0.19	0.91±0.16	0.88±0.19	0.43±0.11	0.47±0.13

Here, receptor seeds are represented by the letter 'S'. S₁ = *Cicer arietinum* Acc1, S₂= *Pisum sativum*, S₃= *Cicer arietinum* Acc2, S₄= *Arachis hypogea*, S₅= *Vigna radiata*. Values are expressed as mean±SD.

The excessive use of synthetic pesticides to get rid of noxious pests has resulted in several environmental hazards. Various efforts are being made worldwide to replace these chemicals with natural alternatives that are safer and do not cause much toxicological effects on the environment. Eventhough synthetic herbicides continue to be a key component in most weed management strategies, in the recent past some progressive studies have focused towards natural herbicides from plants. Unlike the synthetic herbicides, the use of plant allelochemicals is environmentally safe and can also reduce the cost of weed control. Manipulations of allelopathy for crop improvement can help conserving species diversity, weed management, environment protection through ecofriendly use of allelopathy/allelochemicals for the control of weeds, pests and plant diseases. Biotechnological approaches like tissue or cell culture, DNA recombinant technology, microbial fermentation and several other upcoming molecular-genetic techniques like Polymerase Chain Reaction (PCR) and Random Amplified Polymorphic DNA (RADP), synthesis of herbicide and pesticide based on natural product chemistry etc. can be employed for developing crop production. This can make the crop plants resistable to the harmful effects of weeds, nematodes, pests and insects ^[17, 18].

Here, since we identified that the methanolic extract of the plant *Garcinia gummi-gutta* is possessing allelopathic property, it is needful to explore the nature of the compound, and also, to determine whether it is the action of a single compound or a group of compounds. If so, further steps has to be taken to isolate, purify and characterize the bioactive compound/s and to test its weedcidal effect and various other broad-spectrum activities.

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