Searching For Nature Stories 2015

Negative Allelopathy Among *Wedelia triloba L.*

Po Leung Kuk Centenary Li Shiu Chung Memorial College

Team 04

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Content

1 Abstract

Wedelia triloba L., which belongs to family *Compositae*, is commonly found at the road side or on a grass slope throughout the year. They spread at a fast speed once their roots are located extensively. Oddly, there are less and poorly-grow plants in the area where *Wedelia* grow. Thus, people treat them as a kind of weeds. Our objectives are to investigate how *Wedelia* affects the growth of other plants growing near it. In order to do so, we have carried out a series of observations and experiments.

In this study, we examined the effect of aqueous extracts (25%, 50%, 75%) of leaves, stems (with and without) of *Wedelia* on germination and growth of seedlings of green bean. From the data recorded, the result indicates that the leaf and stem parts of *Wedelia* can inhibit the germination and growth of green beans. In another experiment, we investigate the effect of *Wedelia*-planted soil on growing of green bean seeds. After analyzing, the *Wedelia*-planted soil can also inhibit the growth of green beans. Through the test of pH, we also found that *Wedelia* can change the acidic soil to slightly alkaline.

2 Introduction

From our observation, particularly in our field trips to Yuen Long Tai Tong Nature Trail, there are only rare plants on the area where *Wedelia* grow. The relationship between the existence of *Wedelia* and the growth of other plants growing near it raises our interest. We have visited the selected place to collect *Wedelia* and soil samples for investigations.

Brief background information of *Wedelia triloba L.*:
Wedelia triloba L. is a mat forming perennial herb with rounded stems. Leaves are fleshy, usually 2 to 4 inches long and 1 to 5 inches wide, with irregularly toothed margins. Flowers are solitary, one inch in diameter and yellow-orange in color. New plants arise from nodes that root at the soil surface.

Question of investigation:
1. What is the role of *Wedelia* taken in the germination and growth of plants?
2. Which parts of *Wedelia* can inhibit the growth of other plants?
3. Does the *Wedelia*-planted soil inhibit the growth of other plants as well?
4. What is the difference of pH before and after *Wedelia* is planted?

3. Materials and Methods

3.1 Investigating the effects of *Wedelia* to the growth of plants

Aim: To observe whether plants (mainly weeds) can co-exist with *Wedelia.*
To investigate whether other plants cannot co-exist with *Wedelia* is because of *Wedelia*'s large covered area.

1. An observational area was set up in our school's garden with *Bidens pilosa* and *Wedelia* planted.
2. The percentage of covered area of *Wedelia* and *Bidens pilosa* was measured
3. The growth of *Wedelia* and *Bidens pilosa* was observed for a month.
4. The percentage of covered area of *Wedelia and Bidens pilosa* was measured again.

3.2 Investigating the effects of extract of different parts of *Wedelia* on the germination and growth of green beans

Aim: To investigate which parts of *Wedelia* can inhibit the germination and growth of other plants through looking into the effects of extract of different parts of Wedelia.

Purpose of getting stem with and without leaves:
If allelochemicals are produced in leaves, they may pass through the stems to roots. To eliminate the factors brought by leaves, stem without leaves is also investigated.

Prepare of stem without leaves:
Remove the leaves from the stems a week ago before collecting the stems for making extracts.

(A) Germination of green beans

1. Set up nine petri dishes each containing 20 green beans and the same amount of cotton.
2. Prepare 2 portions of each containing 200g of leaves, stems with leaves and stems without leaves.
3. Prepare 600 $cm^2$and 200cm^2 water to 200g of leaves respectively in order to produce 25% and 50% concentration of the extract of *Wedelia* leaves in proportion with water.(For example, 200g/(200+600)cm^3 x 100%=25%)
4. Pour the leaves and the prepared amount of water to a blender and squeeze out the extract of *Wedelia* leaves.
5. Add the extract of *Wedelia* leaves with 0%, 25% and 50% concentration respectively to the petri dishes.
6. Repeat step 3-5 on stems with leaves and stems without leaves respectively.

7. Put the petri dishes under room temperature and observe for one week.
8. Record the number of green beans germinated.

(B) Growth of green beans

1. Immerse the green beans in the water until they germinated.
2. Set up twelve petri dishes each containing 20 germinated green beans and the same amount of cotton.
3. Prepare 3 portions of each containing 300g of leaves, stems with leaves and stems without leaves.
4. Prepare 900 cm^2,300cm^2 and 100 cm^2water to 300g of leaves respectively in order to produce 25% ,50% and 75% concentration of the extract of *Wedelia* leaves in proportion with water.(For example, 300g/(300+900)cm^3 x 100%=25%)
5. Pour the leaves and the prepared amount of water to a blender and squeeze out the extract of *Wedelia* leaves.
6. Add the extract of *Wedelia* leaves with 0%, 25%, 50% and 75% concentration respectively to the petri dishes.
6. Repeat step 4-6 on stems with leaves and stems without leaves respectively.
7. Put the petri dishes under room temperature and observe for one week.
8. Record the number of green beans germinated.

3.3 Investigating the effects of *Wedelia*-planted soil on the growing of green beans

Aim: To investigate whether the *Wedelia*-planted soil plays a role in inhibiting the growth of other plants as well through looking into the effects of *Wedelia*-planted soil on the growing of green beans.

1. Put the soil from a *Wedelia*-planted and without *Wedelia*-planted area in two pot respectively
2. Put 20 green beans in each of the two pots.
3. Observe the germination and growth of green beans for a week.
4. Record the number of green beans germinated and measure the heights of the seedlings.

3.4 Investigating the effects of *Wedelia* on pH of soil

Aim: The alkaline and high acidic condition will lower the solubility of iron, manganese, copper, zinc, etc., which will inhibit the growth of most of the plants. If *Wedelia* has changed the pH of soil, the change in pH may be one of the significant factors inhibiting the growth of other plants.

1. Plant the *Wedelia*, Brassica chinensis L, Brassica parachinensis, Sonchus olercaceus L. and Brassica juncea in five adjacent fields.
2. Measure and record the pH of soil in different sites respectively
3. Measure and record the pH of soil in different sites respectively again and pH of different depth of the soil where *Wedelia* planted after one months.

3.5 Working schedule

Date	Venue	Event
17-01-2015	Yuen Long Tai Tong Nature Trail	Collect *Wedelia* samples
24-01-2015	Yuen Long Tai Tong Nature Trail	Collect *Wedelia*-planted soil, carry out pH test and set up three sites for experiment
01-02-2015	Gardenbackyard of Po Leung Kuk Centenary Li Shiu Chung Memorial College	Farm a field to plant Brassica chinensis L., Brassica parachinensis, Sonchus olercaceus L. and Brassica juncea together with *Wedelia* for oberservation. And test the pH of the site
06-02-2015 to 07-02-2015	Po Leung Kuk Centenary Li Shiu Chung Memorial College	Obtain extracts of different parts of *Wedelia* and add them to the petri dishes together with green beans
08-02-2015 to 15-02-2015	Groupmates' home	Record the result on the effect of extracts of different parts of *Wedelia* on germination and growth of green beans
03-03-2015	Yuen Long Tai Tong Nature Trail	Collect *Wedelia*-planted soil and record the result of pH on the specified three sites
04-03-2015 to 10-03-2015	Groupmates' home	Carry out experiment on the effect of *Wedelia*-planted soil on the growing of green beans and record the result
12-03-2015 to 20-03-2015	Gardenbackyard of Po Leung Kuk Centenary Li Shiu Chung Memorial College	Record the result on the field planting the five plants and analysis the final result recorded

4 Results

4.1 Growing of plants in the *Wedelia* growing area

Observation

We set up an observation area in our school's garden.

From our observation, we found that the area growing *Wedelia* seldom have weeds growing in it (Fig 1.), while other areas are weedy. (Fig 2.)

Fig 1. *Wedelia* having no weeds growing in it

Fig 2. *Wedelia* and weeds

In order to prove that it is not because of *Wedelia*'s large covered area that other plants cannot grow, we also set up another observation area of 2mX2m to observe the growth of *Bidens pilosa* 鬼針草, which had been proved to have an negative allelopathic effect. (Bais et al., 2003; Hierro and Gallaway, 2003; Prati and Bossdorf, 2004; Dorning and Cipollini, 2006; Cipollini et al., 2008) with *Wedelia* and measure their covered area after one month.

Table 1. The percentage of covered area before and after one month

Plants \ Covered area (%)	*Bidens pilosa*	*Wedelia*
Before	40	60
After	95	5

As shown in Table 1, the *Bidens pilosa* had occupied most of the growing area of *Wedelia* after one month. (Fig 3)

Before **After**

Fig 3. The observation area before and after one month

4.2 The negative allelopathic effect of *Wedelia* on green beans

The inhibitory effect of aqueous extracts of different parts of *Wedelia* on the germinating of green beans

To investigate how the aqueous tissue extracts affected the germination and growth of green beans, we set up two experiment, we put the extract of different concentration of leaves and stems of *Wedelia* in the petri dishes with 20 green beans each and take the record after one week.

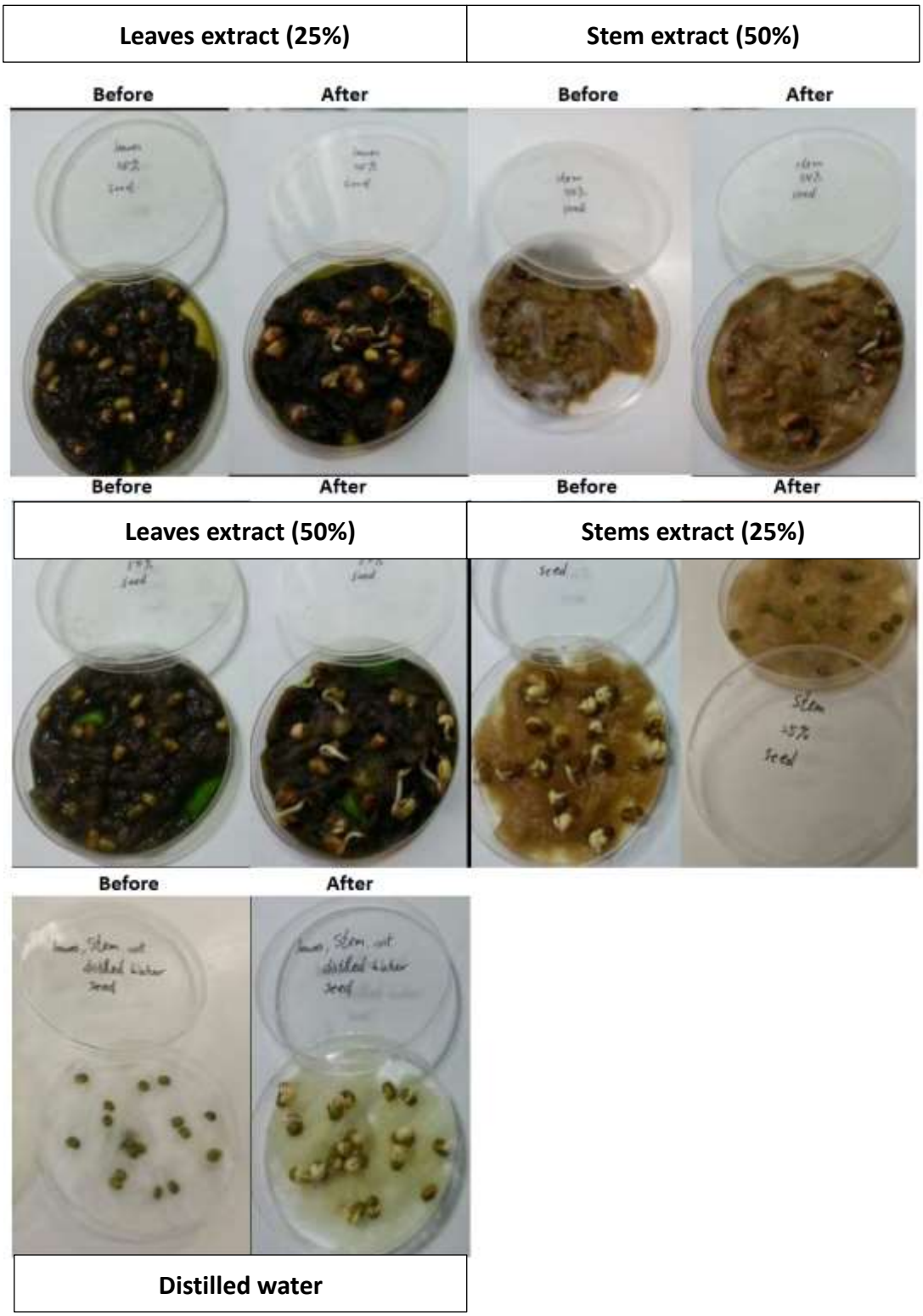
Table 2. The number of germinated green beans

Source of extract \ Conc. of extract (%)		0	25	50
Leaves	No. of germinated green beans	19/20	18/20	17/20
	Rate of inhibition (%)		5.3	10.5
Stems (with leaves)	No. of germinated green beans	19/20	16/20	14/20
	Rate of inhibition (%)		15.8	26.3
Stems (without leaves)	No. of germinated green beans	19/20	16/20	15/20
	Rate of inhibition (%)		15.8	21.1

*Calculation: Rate of inhibition= $\frac{19 - \text{No. of germinated green beans}}{19} \times 100\%$

From Table 2, the inhibitory effect of the stems of *Wedelia* on green beans is better than that of the leaves. The higher the concentration the more potent the effect is. While the inhibitory effect of the stems with leaves on green beans is better than that without leaves. (Fig 4.)

Fig 4. No. of germinated beans before and after one week



The inhibitory effect of aqueous extract of different parts of *Wedelia* on the growing of green beans

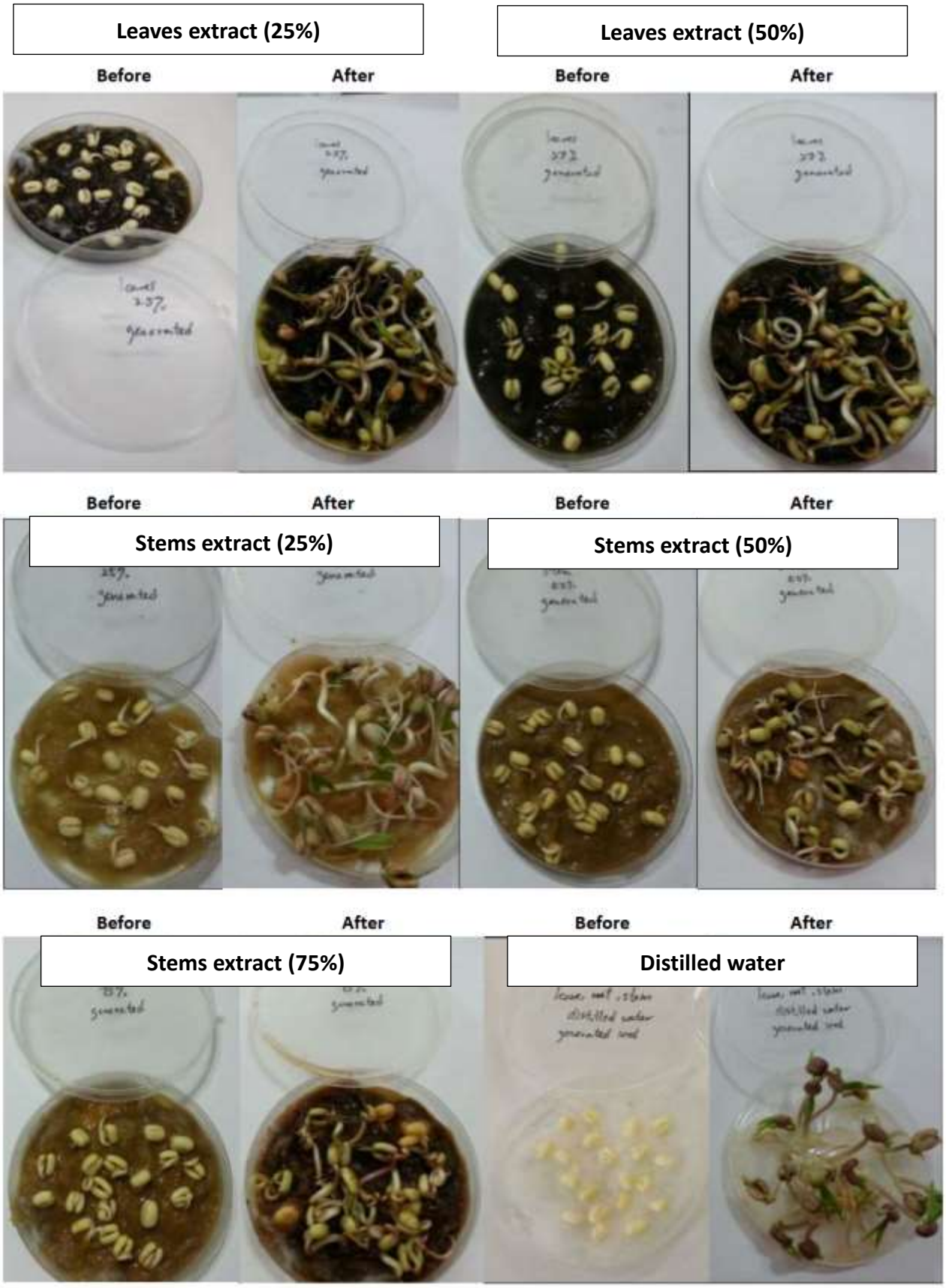
Table 3. The heights of germinated green beans

Source of extract \ Conc. of extract (%)		0	25	50	75
Leaves	Average height of the seedlings (cm)	9.24	7.84	7.81	7.49
	Rate of inhibition (%)		15.2	15.5	18.9
Stems (with leaves)	Average height of the seedlings (cm)	9.24	7.55	7.43	7.28
	Rate of inhibition (%)		18.3	19.6	21.2
Stems (without leaves)	Average height of the seedlings (cm)	9.24	7.72	7.63	7.43
	Rate of inhibition (%)		16.5	17.4	19.6

* Calculation: Rate of inhibition= $\frac{9.24 - \text{Average height of the seedlings}}{9.24} \times 100\%$

As shown in Table 3, heights of the seedlings grow shorter when the concentration becomes higher. The higher the concentration, the more potent the effect is. (Fig 5.)

Fig 5. The height of germinated green beans before and after one week



The effect of *Wedelia-planted* soil on the growing of green beans

In this experiment, we put 20 green beans in each of the pot of soil geting from the *Wedelia* planted area and area without *Wedelia* planted than take the record after one week.

Table4 . The growing of green beans in soil with and without *Wedelia*-planted

	No. of germinated beans	Average height of the seedlings (cm)
Soil with *Wedelia* planted	14	5.26
Soil without *Wedelia* planted	16	9.35

As table 4 shows, the soil from the *Wedelia* planted area has an inhibitory effect on the growing of green beans. (Fig 6 , 7)

Fig 6. The growing of beans before and after one week

Fig 7. The height of beans after one week

4.3 The test of pH of soil

The pH of soil of different sites with *Wedelia*

We visited the Tai Tong Nature Trail in Yuen Long and set up three sites where *Wedelia* growing there to test the pH of the soil. (Fig 8.)

Fig 8. Tai Tong Nature Trail

Site 1:
Near the car park
Site 2:
Around the starting point of the trail
Site 3:
Around the midway of the trail

Table 5. The pH of soil of the three sites

	Site 1	Site 2	Site 3
pH	6.5	7.5	7

From table 5, the pH of soil for the growth of *Wedelia* is mostly neutral or even slightly alkaline.

The change of pH of soil before and after *Wedelia* and the four other plants are planted

In order to show how *Wedelia* affect th pH of the soil, we plant the *Wedelia*, *Brassica chinensis L, Brassica parachinensis, Sonchus olercaceus L.* and *Brassica juncea* in five adjacent fields and take the record after one months.

Table 6. The change of pH before and after one month

Plants / pH	*Wedelia*	Brassica chinensis L 小白菜	Brassica parachinensis 菜心	Sonchus olercaceus L. 油麥菜	Brassica juncea 芥菜
Before	6.5	6.5	6.5	6.5	6.5
After	7.5	6.5	6.5	7	6.5

As shown in Table 6, the *Wedelia* has increased the pH of the soil, from slightly acidic to slightly alkaline, while the pH of the other three plants did not have obvious change, Sonchus olercaceus L. only change from slightly acidic to neutral. (Fig 9.)

Fig 9. The fields planting the five plants

The pH of different depth of the soil where *Wedelia* planted

To investigate to what extent can the *Wedelia* affect the pH of the soil, we take the soil samples with *Wedelia* planted of different depth and take the record.

Table 7. The pH of different depth of the soil

	Topsoil	10cm deep	20cm deep
pH	7.5	7	6.5

As shown in Table 7, the pH of the topsoil is the highest, the deeper the soil, the less its pH will be affected by the *Wedelia.*

5 Discussion

5.1 Interpretation of results

5.1.1 Growing of plants in the *Wedelia* growing area

From our observation, the area growing *Wedelia* seldom has weeds growing with it, while the areas beside where *Wedelia* are growing (but areas that *Wedelia* has not spread to) are weedy.

However, *Wedelia* is a kind of creeper; in order to prove that it is not because of its large covered area that other plants cannot grow, we have made another observation: An area originally with about 40% of *Bidens pilosa* and 60% of *Wedelia* growing in it, after one month, the number of *Wedelia* shrinks, with about 5% *Wedelia* and 95% *Bidens pilosa*. This shows other plants, like *Bidens pilosa*, which had been proved to have an negative allelopathic effect. (Bais et al., 2003; Hierro and Gallaway, 2003; Prati and Bossdorf, 2004; Dorning and Cipollini, 2006; Cipollini et al., 2008) can still survive under *Wedelia*'s llarge coverd area. Also, *Indigofera spicata* 穗花木藍 is another kind of creeper, but there are still some weeds co-existing with it.(Fig 10.) From the above, we can deduce that the inhibitory effect of *Wedelia* to other plants is not due to its large covered area, the success of it is its allelopathic effects

Fig 10. Weeds co-existing with Indigofera spicata

5.1.2 The negative allelopathic effect of *Wedelia* on green beans

As shown in the experiment, both the stems (with and without leaves) and leaves of *Wedelia* can inhibit the growth and germination of the green beans. And the inhibitory effect of the stems with leaves is better than the stems without leaves. These show that both the stems and leaves of *Wedelia* can produce allelochemicals. From another experiment, we found that the Wedelia-planted soil can also inhibit the growth and germination of the green beans. We can deduce that the allelochemicals are released the roots of *Wedelia* to the soil.

5.1.3 The test of pH of soil

The pH of soil of different sites with *Wedelia*

The change of pH before and after *Wedelia* and the four other plants are planted

The samples of soil of *Wedelia* collected from the three sites in Tai Tong Nature Trail were tested to be slightly alkaline. Another experiment also shows that *Wedelia* turns the slightly acidic soil (the relatively suitable growing condition for most of the plants) to slightly alkaline. The alkaline condition will lower the solubility of iron, manganese, copper, zinc, etc., which will inhibit the growth of most of the plants.

The pH of different depth of the soil where *Wedelia* planted

In the area growing *Wedelia*, the pH of the topsoil is the highest, and pH of the soil gradually increase as it goes deeper. The reason for this maybe *Wedelia* has shallow roots, as its roots are mostly distributed in the topsoil, and because of this, the deeper soil (≧20cm) is not easily affected by the roots of *Wedelia*. As the seeds of the weeds are usually germinated in the topsoil, the negative allelopathic effect of *Wedelia* on them is obvious. While the roots of the woody plants penetrate deeply into the soil, they are not easily affected by the *Wedelia* and therefore we can see *Wedelia* growing under the trees. (Fig 11.)

Fig 11. *Wedelia* growing under a tree

5.2 Significance

Allelopathy plays an important role in ecosystem and agroecosystems. Allelochemicals that suppress or eliminate competing plant species near the source plant have been extensively studied because of their agricultural potential as herbicides Rice (Oryza sativa L) is an important annual plant cultivated for grain or as a cover crop in many countries and it is also used for weed suppression in agro-economic systems through its release of allelochemicals.
As *Wedelia* creeps and roots at nodes to form a low-growing mat, making a dense ground cover, it is an excellent vegetation on slops that especially good for soil retention and erosion control. Although *Wedelia* are very attractive with nearly constant and prolific blooming, it can be mowed to keep low and manicured. Moreover, because of its negative allelopathic effect, most herbaceous plants can hardly grow. *Wedelia* acts as an herbicide that can save the money, time and labour allocated to weeding.

5.3 Limitations

Limited equipment

As for the equipment, we used the pH paper for the experiments testing pH. However, it is quite difficult to determine the difference in colour change by naked eyes, especially when the samples are mainly weak acid and alkali. It would be much better if we had a pH meter.

Lacking time

One of our experiments investigating whether *Wedelia* will remove the herbaceous plants (four different kinds were being investigated) when they are growing in the same field failed due to a lack of time, as it takes time for *Wedelia* to produce and release the allelochemicals that can affect a large area of soil.

Limited targeted species

As we only investigated the allelopathic effect of *Wedelia* on one species i.e. green beans, the result is not general enough.

5.4 Improvement

As we have done only one set of experiment for investigating the inhibitory effect of *Wedelia* on green beans, we should repeat the experiment few more times in order to reduce the measurement error.

Also, in our study we cannot shows the presence of allelochemicals in *Wedelia* due to lacks of technology and resources.

We use the function of dieing the polyphenols to know that the Wedelia trilobata can produce Allelochemicals called as the polyphemols. (龔冠寧,2003) The mesophycell produce the polyphemols from chloroplast of cortex,the polyphemols transport to root and release from root exudation. The plant will absorb polyphemols of the soil, and will affect the growth of the plant.

6 Conclusion

From the experiment of investigating the inhibitory effect of aqueous extracts of different parts of *W. chinensis* on the germinating of green beans, it is found that the inhibitory effect of the stems of *W. chinensis* on green beans is better than that of the leaves, while the inhibitory effect of the stems with leaves on green beans is better than that without leaves.

From studying the inhibitory effect of aqueous extracts of different parts of *W. chinensis* on the growing of green beans, it is suggested that the heights of the seedlings grow shorter when the concentration of *W. chinensis*' extract becomes higher.

Concerning the effect of *W. chinensis-planted* soil on the growing of green beans, the final figures indicate that the soil from the *W. chinensis* planted area has an inhibitory effect on the growing of green beans.

Regarding the change of pH of surrounding soil and plants before and after *W. chinensis* is planted, we discovered that the *W. chinensis* has increased the pH of the soil, from slightly acidic to slightly alkaline, while the pH of the surrounding plants did not have obvious change. Furthermore, the deeper the soil, the less its pH will be affected by the *W. chinensis.*

To conclude, W. chinensis limit the growth of other plants in most condition. The main reason is that both the stems and leaves of *W. chinensis* can produce allelochemicals which are released through its roots. These allelochemicals turns the slightly acidic soil to slightly alkaline. The alkaline condition will lower the solubility of iron, manganese, copper, zinc, etc., which will inhibit the growth of most of the plants.

However, as *W. chinensis* has shallow roots, which are mostly distributed in the topsoil, the negative allelopathic effect of *W. chinensis* can only show when the seeds of the weeds are usually germinated in the topsoil. In some cases, if the roots of the woody plants penetrate deeply into the soil, they are not easily affected by the *W. chinensis*, thus the negative allelopathic effect of *W. chinensis* is not obvious.

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