# ALLELOPATHY AS AN AID TO WEED CONTROL

By

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#### <u>ABSTRACT</u>

Throughout history weeds have always posed a threat to agricultural crops. Since 1940 when 2,4-D was discovered the most common form of weed control in developed countries has become the used of herbicides. Sadly some varieties of weeds have developed resistance to herbicides, usually herbicides that have single modes of action. There is also an increased pressure on farmers to adopt more environmentally friendly practices in their farming operations. One of the main areas that is under pressure is the application of artificial chemicals to the land. Farmers are being asked to reduce application rates where possible to reduce any chance of contamination of the surrounding environment. This has resulted in farmers looking for alternatives in controlling weed problems within a crops.

Farmers have always known that some crop varieties showed a higher competitiveness than other varieties but in many cased have not understood why. The truth is there are various factors that make plant species and even varieties within plant species competitive they include nutrient demand, root type, growth rate and allelopathy. Allelopathy is the name given to a natural form of weed control. What happens is a plant species releases a toxic chemical referred to as an allelochemical which acts as a herbicide killing or suppressing the growth of neighbouring plants. With allelochemicals being a biochemical they are extremely biodegradable meaning that they have less potential to cause environmental damage.

This dissertation looks at whether allelopathy could be considered as an effective mode of control of chickweed *Stellaria media* in winter wheat. Two sets of experiments were carried out both in controlled environments. The first experiment was simply set up to screen for allelopathic effects on chickweed *Stellaria media* using various plant extracts which included eucalyptus, rice as well as two wheat varieties Consort and Soissons. The second experiment focused on the two wheat varieties looking at which part of a wheat plant (leaves, stem or roots) had the highest allelopathic effect on chickweed *Stellaria media*. It also looked at whether there were varietal differences between Consort and Sossions.

Both experiments showed that it was possible to have some control over chickweed *Stellaria media* by using allelopathy. Sadly allelopathy alone would not be able to be used to control chickweed *Stellaria media* but could be used with a reduced herbicide application rate to achieve acceptable control.

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#### LITERATURE REVIEW

A weed is defined as a wild plant growing where it is not wanted (The Little Oxford Dictionary 1986). In the case of this dissertation the place is in a winter wheat crop and the weed is chickweed *Stellaria media*.

The main objective in plant competitiveness is to establish root systems and required leaf area before any other plant species has the chance to interfere or limit this establishment process. By achieving this, a plant can produce optimum energy levels from the nutrient uptake from the roots and a high rate of photosynthesis from the leaves and stems.

There are many reasons why it is undesirable for weeds to be growing within a crop. These reasons include disease transfer, shadowing and competition for water and nutrients. Competition for water and nutrients is likely to be the most important factor as this affects yields, crop health and quality. It is estimated that ten percent of potential agricultural production on a world scale is lost because of weeds. Pre world war two almost all weed control was carried out by mechanical tillage and cultural practices. But with the demand for increased production rates herbicides were introduced. The discovery of selective herbicides 2,4-D and MCPA during world war two provided an effective new way for the control of broadleaved weeds. In 1986 UK farmers sprayed some 14.5 million ha with herbicides at a cost of £176 million for the chemical alone and £1.98bn in 2003 (British Agrochemicals Association). In more recent years there has been an increased interest in organic farming systems. In an organic system weed control is essentially going back to the traditional methods such as rotations and soil cultivations. If it were not for the development of brush weeders and the like this would mean that there would be a greater demand for labour.

Plant competitiveness is a key factor in the successful growth of a plant species where there are limited resources. There are many external factors that affect plants competitiveness:

- Soil pH
- Diseases • Insects
- Fertiliser Timing

• Fertiliser Placement

• Soil Fertility

• Soil Temperature

- Seed Depth • Seed-Soil Contact • Presence of Stubble
- Phytotoxic Herbicide

• Soil Structure (Pans etc)

• Moisture Conditions

- Crop Density
- Row Spacing

These external factors affecting the competitiveness of a plant by restricting or reducing the development rate of the plant. Any soil based factor reduces the plants ability to form an efficient rooting network resulting in poor nutrient/water uptake, which consequently reduces the rate of photosynthesis. This allows weed species that have already established themselves or with a lower nutrient demand to shad or deprive crops of water and nutrients. Insects can put a plant under stress by introducing disease or by physically damaging the plants ability to develop. Diseases such as Take all Ophiobolus graminis var tritici or Club root Plasmodiophora brassicae restrict a plants ability to take up nutrients through the root systems by causing a restriction or causing poor root formation. Sticking to Take all Ophiobolus graminis var tritici and Club root Plasmodiophora brassicae these diseases can cause long term problems for a crop as spores of Club root *Plasmodiophora brassicae* for an example can survive in the soil for up to twenty years before sassing to be harmful to a crop. Take all Ophiobolus graminis var tritici can become a problem if constant cropping of one species is carried out. The disease is carried over to future years using the green bridge. Effective control of the germinating volunteers and weeds such as couch grass can control Take all Ophiobolus graminis var tritici from building up in a field.

The diagram below illustrates the availability of nutrients at various soil pH levels. As it can be see by figure 0.1.1 it is important that soil pH at the correct level if a crop it to grow properly. The ideal level for cereal crops is around pH 6.5 although some weed species favour other pH levels giving them a competitive advantage over a cereal crop. Figure 0.1.1, Source: aardappel pagina 2004



It is not just external factor that affect a plants competitiveness internal factors also have to be considered.

• Varietal Disease Resistance

• Nutrient Demand

• C3 Or C4

• Type Of Root System

• Leaf Area Index

Different plants and varieties within plant species have different nutrient demands. Nutrient demands depend on various factors such as plans size, production cycle (annual/biannual), growth rate or climate of origin. It is common to find C4 weeds in a C3 crop and vice versa this is because the have different nutrient demands due to the different ways in which they photosynthesise. As a rule of thumb the C4 crops are more efficient users of nitrogen that a C3 crop.

There are two main types of root systems tap roots and fibrous roots.

Fibrous Root

Tap Root



Figure 0.1.2, Source: aardappel pagina 2004



Figure 0.1.3, Source: aardappel pagina 2004

The type of rooting system that a plant has also can affect the competitiveness of a plant over another as if the weed species has a shallow root system compared to the cropped plant. What this allows the crop to draw water and nutrients from a bigger area than the weed species allowing it to in theory draw more nutrients than the weed species allowing it to grow faster.

There is also another factor that can affect competitiveness of a plant ant that is allelopathy. The name allelopathy comes from the Greek words "allelon" meaning "each other" and "pathos" meaning "feeling, suffering, or sensitive" using this as a definition it describes both the positive and negative effects that allelopathy has on plants. There are many other definitions for allelopathy, which better describes the effects of allelopathy such as "the effect(s) of one plant on other plants through the release of chemical compounds in the environment (Rice 1984). Another would be "Allelopathy is the growth suppression of one plant species by another due to the release of toxic compounds"(Lambers et al.1998). Rice (1984) both describes the stimulatory and the suppressing effects of allelopathy while Lambers et al (1998) only describes the suppression effect. It is the Lambers definition that is favoured by ecologists as many ecologists feel that allelopathy should only be used to describe an suppressing effect.

The toxic compounds, which are referred to in the definition of allelopathy, are called allelochemicals. If these allelochemicals are to have an effect on a target plant they have to be released from a donor plant. This process can be carried out in four different ways:

- 1. The run off of leachate (Leaching) or gasses released (Volatilization) from the leaves or stem of the plant.
- 2. The release of Phytotoxic compounds from the green parts of a plant
- 3. The release of Phytotoxic compounds from dead plant parts e.g. leaves
- 4. The release of Phytotoxic compounds (Exudation) from the plants roots



Figure 0.1.4, Source: Sust 2004

Allelopathy works by the release of either Phenolic compounds, flavonoids, terpenoids, alkaloids, steroids, carbohydrates or amino acids. When these compounds are mixed they have a greater allelopathic effect than on their own. It is important to note that not all plants exhibit allelopathic effects.

It has long been thought that their maybe differences in allelopathic effects between varieties within plant species similar to the known varietal differences such as disease and pest resistance. One of the first studies on varietal differences was carried out in cucumbers *Cucumis sativus*. This experiment that involved five hundred and twenty six

varieties of cucumber showed twenty six varieties caused fifty to eighty seven percent growth suppression (Putnam and Duke 1974). Later a field study showed that allelopathic effects could only be achieved in certain conditions, which makes field trials difficult to carry out which is why most scientists prefer lab methods.

Allelopathy affects germinating seeds in one of two ways by reducing seed germination levels and reducing seedling growth. There is no common mode of action or physical target for allelochemicals. Some of the known modes of action are disruption of cell division, the damaging of cell membranes causing poor nutrient/water uptake reducing photosynthesis rates and production of pollen (James J. Ferguson, Bala Rathinasabapathi 2003)

Allelopathy has potentially got a future use in the control of weeds this could be in many forms for an example a variety of plant which is known to have allelopathic qualities could be planted to smother a crop, in a rotational sequence or when left as a residue or mulch especially in minimum tillage systems in an attempt to control subsequent weed growth. This method would not achieve high levels of weed control a better method would be to apply allelopathic compounds with synthetic herbicides. This would give the herbicide a greater affect especially in the presence of herbicide resistant weeds allowing application rates of synthetic herbicides to be reduced, which would be both, cost saving and beneficial to the environment.

#### **INTRODUCTION**

It was estimated that ten percent of the total world production is lost by weeds (Hill 1977). In areas of poor weed control such as Ethiopia this percentage can be as high as 60 percent. It is estimated that applying herbicides cost British farmers 1.98bn in 2003 (Agricultural Biotechnology Council 2002). This is quite a considerable amount of money when you consider that there are 303,000 agricultural holdings in the UK of which 1781 are located in Scotland (DEFRA)

Competition affects all forms of life. In more cases than not it becomes survival of the fittest. This maintains high vigour within species. Competition within plant species is usually over the following:

- Water can be limited by weed uptake
- Light can be limited by shading or by reduced GAI caused by competition over nutrients
- Nutrients limited by weed uptake

These factors are all important for plant development. For instance water, light and nutrients are required in photosynthesis. The process of photosynthesis is an important process as this is what generates the energy for the plant to grow and develop. The overall reaction of photosynthesis can be written as:

 $6H_2O + 6CO_2$  \_\_\_\_\_  $C_6H_{12}O_6 + 6O_2$ 

Six molecules of water plus six molecules of carbon dioxide produce one molecule of sugar plus six molecules of oxygen (W H Freeman). The water for this reaction enters the plant through the roots and is transported to the leaves through a series of specialised plant cells called the xylem. The carbon dioxide enters the plant through structures located on the underside of the leaves called the stomata. The stoma is an opening that opens and closes allowing the plant to respire without losing water. The second part of photosynthesis is the interception of light. It is here that nutrients such as nitrogen are important in order to produce an optimum Green Area Index (GAI) of five to six (Sylvester-Bradley R. Scott R. K. Clare R. W 1997). Nitrogen encourages plants to

tiller and aids leaf formation allowing a plant to capture light energy. It is understood that an increase from GAI two to GAI three captures fifteen percent more light, but only three percent is gained from GAI six to seven (Sylvester-Bradley R. Scott R. K. Clare R. W 1997). The light intercepted by the leaves excite the electrons in the chlorophyll which then produce what is called ATP and NADPH which are used by the plant for energy to grow. Any reduction of any of the three main components can result in a lowered rate of photosynthesis resulting in lowered plant energy levels.

While space is also an important factor in a plants development as in the case of roots if there is not enough room for each individual plant to establish an adequate rooting system the plant will not be able to achieve the required nutrient uptake. And in the case of development above the soil if there is not enough space for the plant to maximise its leaf area light interception will be reduced consequently reducing the rate of photosynthesis. It is for these reasons that weed control is essential within a crop if acceptable/good crop yields are to be achieved.

The presence of weeds can also lead the crop to come under attack from fungal growths. An example of this would be mildew *Blumeria graminis*. The increased humidity caused reduced airflow through the stem systems of a crop favours the growth of fungi such as mildew *Blumeria graminis*. Depending on the severity of the infection in an area of cropped land crop yields can be considerably reduced.

Different varieties of crops and weed species have differing levels of competitiveness. This fact is often used in an attempt to reduce the cost of weed control and produce good crops while maintaining low costs. Similar to the principles used in selecting disease or pest resistant crops.

There are many different ways in which farmers have controlled weed species. Some of these techniques have been developed over hundreds of years and some methods have been developed by beliefs as in the case of Biodynamic farmers. The most common method of weed control is the use of herbicide sprays. With the ever increasing amounts of weeds developing herbicide resistance new forms of control always have to be developed. In the UK Black grass *Alopecurus myosuroides* which herbicide resistance has been a problem since 1982 with over one thousand confirmed cases

between 1982 and 2002. (Dr S Moss, Mr J Orson 1993) It is not just Black grass *Alopecurus myosuroides* that has developed herbicide resistance in the UK Italian ryegrass *Lolium multiflorum* and wild oats both Common wild oats *Avena fatua* and winter wild oats *avena sterilis* have shown levels of herbicide resistance over the past decade. This is not purely a UK problem the USA has the highest levels of herbicide, fungicide and pesticide resistance in the world. There is much debate over why this is much of this debate revolves around these three schools of thought:

- 1. Reduced doses cause more slightly resistant isolates of fungi, pests or weeds to survive and hence exert a push towards a more resistant population
- 2. Full rate doses pose an increased selection pressure and kill the most sensitive in the target population leaving only the most resistant to survive and multiply
- 3. Quality of control rather than dose is critical a well timed reduced dose will create the same selection pressure for resistance as a full dose.

Farmers are constantly looking for alternative methods as weeds become resistant and there are increased restrictions on where and how herbicides can be applied. Alternative weed controls include:

- Integrated Crop Management (ICM)
- Brush weeders
- Flame weeders
- Hand weeding
- Crop rotation

Many of these forms of weed control can be expensive and time/labour demanding and in some cases require several treatments.

Genetically Modified Organisms (GMO) have potential in improving control of weeds. There are currently concerns in the UK over the supply of GMO's and there impact on the surrounding environment especially when tests have shown that GMO's can contaminate a fifteen mile radius from where the GMO is grown. This is largely spread by bees cross pollinating. This concern is quite different in the USA and Canada where GMO oilseed (Canola) is widely grown. It could be said this is down to how the general public have been informed and educated (Pers. Com. 2003). The British government are currently trying to weigh up the benefits and the draw backs of GMO's in the attempt to decide on whether to allow the growth of GMO's on British farms. (Anon 2003)

A natural phenomenon that has potential for the control of weeds is known as Allelopathy. Allelopathy is the name given to biochemical interactions between different plants. This term usually implies that one plant produces one or more chemical that have an inhibitory effect on nearby plants, but allelopathy may also include stimulatory effects (Muller. C.H 1965). Allelopathy currently is not readily recognised as a form of weed control and only is utilised in some organic systems. There would be many benefits if allelopathy could be used to control weed populations in crops. As previously mentioned herbicides cost British farmers 1.98bn in 2003 and this cost is always rising. In recent years grain prices have fallen dramatically resulting in poor profit margins for British farmers. There are also environmental considerations in reducing the amount of artificial chemicals being applied to the land, as this reduces the risk chemicals entering local ecosystems. Herbicide pollution can reduce plant life that other species depend on having reduced food sources etc and can consequently have a knock on effect on other species.

This dissertation investigates whether allelopathy can be used to control weeds in a winter wheat crop. Firstly I shall be looking at the effects of allelopathy and which plant species exert allopathic effects on Chickweed *Stellaria media* which was the chosen weed species due to its completeness and because it is a common weed species in UK fields. The two wheat varieties Soissons and Consort were selected at random.

#### METHODOLOGY

### Experiment 1

The propose of this experiment was to screen for allelopathic effects on chickweed seeds using a range of plant extracts. The methodology for this experiment was modified from M C Robson. D Robinson. A M Litterick. C Watson, M Leitch (August 2000) investigations into allelopathic interactions of white lupins.

An incubator was set to eighteen degrees centigrade, forty eight hours before the experiment was due to start. This was to allow the incubator could achieve a stable environment before the experiment was carried out. Once the incubator had achieved a stable environment nine extracts were prepared. See Figure 1.1.1



Figure 1.1.1

The nine plant extracts, which were selected for this experiment, were:

- 1. Green pine needles
- 2. Senesced pine needles
- 3. Eucalyptus leaves
- 4. Couch rhizomes
- 5. Wheat (Consort) roots, leaves and stems
- 6. Wheat (Soissons) roots, leaves and stems
- 7. Rice seed
- 8. Eucalyptus leaves liquidised
- 9. Green Pine Needles liquidised

The extracts were prepared as follows. The green needles and the eucalyptus leaves were split in to two groups Liquidised and whole this was to see if whole leaves exerted an allelopathic effect. As well as the green needles and the eucalyptus leaves both wheat varieties, senesced pine needles, couch rhizomes, and rice were individually liquidised in a domestic blender and mixed with distilled water. The individual extracts were then bottled and stored in the fridge to reduce any fermentation.

The next stage was to prepare thirty petri dishes with filter paper, which is where the chickweed seeds were to be germinated. All this involved was inserting two pieces of pre cut filter paper into the base of the petri dishes.

The dishes were then separated into three groups of ten these were later to become the three replicas in the experiment. Each one of the dishes from each of the groups had five millilitres of an individual extract applied to it and labelled accordingly. The last dish of the three groups of petri dishes is to become the control this only had five millilitres of distilled water applied.

Twenty five chickweed *Stellaria media* seeds were manually counted out and applied to the surface of the filter paper located in each petri dish see figure 1.1.2 and figure 1.1.3.



Figure 1.1.2



Figure 1.1.3

The lids were then places on each of the petri dishes. The three groups of petri dishes were then arranges in a randomised block on each shelf of the incubator and a layer of plastic was placed over the shelf to help to retain the moisture in the petri dishes which may escape from the sides of the petri dishes see figure 1.1.4.



Figure 1.1.4

Each shelf was located below the three sets of fluorescent lamps in an attempt to achieve even light interception across the shelves see figure 1.1.5 Each of the petri dishes were monitored over two weeks. They were assessed on number of seeds that germinated and the length of the shoots (figure 1.1.6) so that growth rate could be calculated.





Figure 1.1.5

Figure 1.1.6

Note: Chickweed *Stellaria media* seed was bought from Herbiseed of New Farm, Mire Lane, West End, Twyford, England on the 14<sup>th</sup> of November 2003.

#### Experiment 2

The propose of this experiment was to look at the difference in allopathic effects between two winter wheat species (Consort and Sossion) on Chickweed *Stellaria media*. And to find which part of the plant had the greater affect. The methodology for this experiment was modified from M C Robson. D Robinson. A M Litterick. C Watson, M Leitch (August 2000) investigations into allelopathic interactions of white lupins.

An incubator was set to eighteen degrees centigrade, forty eight hours before the experiment was due to start. This was to allow the incubator could achieve a stable environment before the experiment was carried out. Once the incubator had achieved a stable environment nine extracts were prepared. See Figure 2.1.1



Figure 2.1.1

The Consort and Soissons plants were cut into three parts. These three parts were to later make up the four extracts. Each plant part was individually liquidised in a domestic blender and mixed with distilled water. The individual extracts were then bottled separately. A mixture of all three plant parts was then made in the same way as before and then mixed with distilled water and bottled. The bottles were then stored in the fridge to reduce any fermentation of the extracts.

The next stage was to prepare thirty petri dishes with filter paper, which is where the chickweed seeds were germinated. All this involved was inserting two pieces of pre cut filter paper into the base of the petri dishes.

The dishes were then separated into three groups of ten these were later to become the three replicas in the experiment the same as in experiment 1. Each one of the dishes

from each of the groups had five millilitres of an individual extract applied to it and labelled accordingly. The last two dishes of the three groups of petri dishes were used as a control this only had five millilitres of distilled water applied.

Thirty chickweed *Stellaria media* seeds were manually counted out and applied to the surface of the filter paper located in each petri dish see figure 2.1.2 and figure 2.1.3.





Figure 2.1.2

Figure 2.1.3

The lids were then places on each of the petri dishes. The three groups of petri dishes were then arranges in a randomised block on each shelf of the incubator and a layer of plastic was placed over the shelf to help to retain the moisture in the petri dishes which may escape from the sides of the petri dishes see figures 2.1.4, 2.1.5 and 2.1.6



Figure 2.1.4



Figure 2.1.5



Figure 2.1.6

Each shelf was located below the three sets of fluorescent lamps in an attempt to achieve even light interception across the shelves see figure 2.1.7. Each of the petri dishes were monitored over two weeks. They were assessed on number of seeds that germinated and the length of the shoots (figure 2.1.8) so that growth rate could be calculated.





Figure 2.1.7

Figure 2.1.8

Note: Chickweed *Stellaria media* seed was bought from Herbiseed of New Farm, Mire Lane, West End, Twyford, England on the 14<sup>th</sup> of November 2003.

### RESULTS AND ANALYSIS

### Experiment 1

The propose of this experiment was to screen for Allelopathy over a range of plants.

### Germination



Table 1.2.1

Table 1.2.2





Table 1.2.4



Table 1.2.5

Table 1.2.6





Table 1.2.9

Table 1.2.10

What Tables 1.2.1 to 1.2.10 show is the number of seeds out of the twenty five seeds that germinated along with the standard error for each point of each graph. In the control (Table 1.2.1) over the experimental period the rate of standard error appears to be a high. The rate at which the seeds germinated appears to be relatively constant.

Tables 1.2.2 to 1.2.4 illustrate the number of seeds that germinated when grown in the pine needle extract. The rate of standard error appears to be lowest in the green pine needles (Table 1.2.4) and momentarily highest in the liquidised pine needles (Table 1.2.2). In both the green pine needle mixtures the rate at which the seeds germinated appears to be constant mean while the senesced pine needles experienced an increased rate of germination during the fourth and sixth day of the experiment.

Tables 1.2.5 and 1.2.6 illustrate the number of seeds that germinated when grown in the rice and liquidised couch rhizomes extract. The errors in Table 1.2.5 (Liquidised Rice) appear to be low throughout the experiment. What this means is that the graph in Table 1.2.5 shows a good representation of what effect rice has on the germination of chickweed *Stellaria media* seeds.

Tables 1.2.7 and 1.2.8 illustrate the number of seeds that germinated when grown in a eucalyptus extract. Again like the rice and couch results the eucalyptus experiments show a low standard error this would suggest that the results gained are a good representation of the effect of these extracts on the germination of chickweed *Stellaria media* seeds. The seeds in Table 1.2.8 appear to germinate more constantly than in Table 1.2.7. The seeds in Table 1.2.7, which were grown in liquidised eucalyptus, leaves experienced an increased rate of germinating seeds between the fourth and sixth day of the experiment.

Tables 1.2.9 and 1.2.10 illustrate the number of seeds that germinated when grown in wheat extracts. The results shown in Table 1.2.9 have a relatively low rate of standard error compared to Table 1.2.10. Table 1.2.10 shows a good level of chickweed *Stellaria media* control along side eucalyptus, couch and rice. The Consort extract (Table 1.2.10) after the forth day of the experiment showed a steady control of the germination of chickweed *Stellaria media* seeds, mean while the Soissons allowed a steady germination rate of the chickweed *Stellaria media* seeds throughout the experiment







Table 1.2.12





Table 1.2.11 shows the number of seeds that germinated in the pine needle extracts compared with the control. All of the pine needle extracts show a suppressed number of seeds that germinated. The liquidised pine needles had the greatest effect followed by the green pine needles and then finally the senesced pine needles. The senesced pine needles showed the least amount of seed germination suppression due to the fact that the cell contents have become dehydrated making the cell contents less mobile than that of the green pine needles.

Table 1.2.12 shows the number of seeds that germinated in the rice and wheat extracts compared with the control. The rice extract shows a high level of suppressed germination of chickweed *Stellaria media* seeds in the early stages of the experiment this effect declined slightly over time. Soissons has shown an equal level of suppression of the germination of chickweed *Stellaria media* as rice by the end of the experiment although Consort appears to have stimulated growth. The reason for this was later found to be due to the breaking down of the wheat cells in a fermenting process, which occurred during the storage of the extracts resulting in the releasing nitrogen, which acted as a fertiliser.

Table 1.2.13 shows the number of seeds that germinated in the couch and eucalyptus extracts compared with the control. Couch appears to have had the least suppressed effect on the germination of chickweed *Stellaria media* out of the three in this graph. Latterly all three extracts showed the same amount of germinating seeds. Both eucalyptus leaf extracts showed a random rate of germination when compared to the linier germination encountered by that of the couch extract.

To summarise there is a degree of germination suppression when chickweed *Stellaria media* is grown in the presence of the following: rice, green pine needles, eucalyptus leaves, couch rhizomes, senesced pine needles and wheat (Soissons). This clearly shows the effects of Allelopathy. As a rule of thumb the liquidised crop species showed a stronger effect of suppression of the growth this maybe because the chemicals from within plant cells are released making them more mobile. Although the results for wheat (Consort) in this case is a little disappointing. As it does not show any form of suppression at all if anything it has encouraged the germination of the chickweed *Stellaria media* seeds. Further analysis of the wheat (Consort) extract showed that it

was high in nitrogen this is because during the storage of the extract it had started to ferment causing the plant sells to produce nitrogen which acted as a fertiliser.

Currently there is some work being done at the Scottish Agricultural College by a Dr Ken Davies on the levels of weed suppression within wheat species. Dr Ken Davies experiments along with H. Wu, J. Pratley, D. Lemerle and T. Haig of Farrer Centre for Conservation Farming and NSW Agriculture, Agricultural Research Institute in Australia have shown that there is an Allelopathic effect from wheat, which would suggest that an error has occurred in this experiment, which has affected the results. Something to consider is the experiments carried out by H. Wu, J. Pratley, D. Lemerle and T. Haig used Annual ryegrass *Lolium rigidum Gaud* which may suggest that chickweed *Stellaria Media* is not effected in the same way as Annual ryegrass *Lolium rigidum Gaud*.







Table 1.2.15





Table 1.2.17





Table 1.2.19



Table 1.2.20

Table 1.2.21



Table 1.2.22

Table 1.2.23

What Tables 1.2.14 to 1.2.23 show is the growth rates of the germinated seeds along with the standard error for each point of each graph. Throughout the experiment there appears to be a low rate of standard error suggesting that this is close to being a true representation of the growth pattern of chickweed *Stellaria media*. The control (Table 1.2.14) shows a extremely steady growth rate thought the duration of the experiment when compared with the other Tables.

Tables 1.2.15 to 1.2.17 illustrate the growth rates of the germinating seeds when grown in the pine needle extracts. The growth rate of the germinating seeds grown in the senesced pine needle (Table 1.2.16) extract shows a reduced rate of growth for the first four days of the experiment this is because no seeds have germinated at this point. Both green pine needle extracts exhibit more linear growth rate although the seedlings in the green pine needles of Table 1.2.17 grow slightly faster than that of the seedlings in the liquidised pine needles.

Tables 1.2.18 and 1.2.19 illustrate the growth rates of the germinating seeds when grown in the rice and liquidised couch rhizomes extracts. The liquidised couch rhizomes in Table 1.2.19 has a much more linear growth rate than the rice extract in Table 1.2.18. It can also be said of all the extracts the couch rhizomes have the most linear growth rate.

Tables 1.2.20 and 1.2.21 illustrate the growth rates of the germinating seeds when grown in the eucalyptus extracts. The liquidised eucalyptus (Table 1.2.20) leaves have a good suppression of the growth rate of the seedlings for the six days before the growth of the chickweed seedlings experiences an accelerated stage of growth before

stabilising again. The whole eucalyptus leaves of Table 1.2.21 have the least suppressing effect on the growth rate of the seedlings.

Tables 1.2.22 and 1.2.23 illustrate the growth rates of the germinating seeds when grown in the wheat extracts. Both Table 1.2.22 and 1.2.23 have a linear rate of growth. Consort in Table 1.2.23 does show a slightly higher level of growth rate suppression compared to Soissons in Table 1.2.22.







Table 1.2.25



Table 1.2.26

Table 1.2.24 shows the growth rate of the germinated seedlings in pine needle extracts compared with the control. You can see by the end of the experiment there is no beneficial effect from any of the pine needle extracts. Both whole and liquidised green pine needles show no suppression of the growth rate throughout the experiment. The senesced pine needles show a slight amount of growth rate suppression over the first four days but this quickly deteriorates over the following two days

Table 1.2.25 shows the growth rate of the seedling in rice and wheat extracts compared with the control. What can be seen from Table 1.2.25 is that rice shows the best suppression of the growth rate compared to both wheat varieties and the other extracts. When the results both wheat varieties are traced alongside the results of the control it can be seen that again the Soissons has in fact stimulated the rate of growth. This is likely to be down to the same reasons as that encountered with the seed germinations. Consort has also shown an increased growth rate. The reason for this is currently unknown.

Table 1.2.26 shows the growth rate of the seedling in couch and eucalyptus extracts compared with the control. Couch has had a suppressing effect on growth rate of the chickweed *Stellaria media* seedlings. Couch rhizomes on average shows a suppression rate of around quarter when compared with the control. Both of the eucalyptus extracts have shown a suppression effect on the growth rate of chickweed *Stellaria media* seedlings. Of the three in this Table the liquidised eucalyptus has shown the highest level of suppression of the growth rate of the seedlings.

To summarise the second set of data gathered during this experiment shows that while the growth rate was not affected by the wheat species the number of seeds that germinated was. In practice this would still give the crop an advantage over chickweed *Stellaria media*, as it would mean that there would still be an increased area for the crop to draw nutrients from. Also shading would be reduced, as there would be a lower population of chickweed *Stellaria media*. Rice again has the best suppression effect allowing a conclusion to be made at this point that rice has the best suppressive effect on chickweed *Stellaria media* from the samples in this experiment. The whole eucalyptus leaves had a very poor effect this maybe down to the surface area in which is available for the translocation of the chemicals from the leaf to the seed. Another observation that can be made from the results is that the chickweed *Stellaria media* only has a sixteen percent germination rate. This can be interoperated in two ways either the seed has a dormancy period which is common in weed seeds or the seed has been of a poor quality. The only way this could be determined is by carrying out an experiment over a longer period to see if any new growth occurs. The only problem with expending the time duration of the experiment is that the extract could start to ferment in the humid environment created in the petri dishes. This humid environment also favours the growth of moulds that attack the seeds resulting in reduced germination.

#### **RESULTS AND ANALYSIS**

#### Experiment 2

The propose of this experiment was to look at the difference in allopathic effects between two winter wheat varieties (Consort and Soissons) on Chickweed *Stellaria media*. And to find which part of the plant had the greater affect.





Date



Date





Table 2.2.8



Table 2.2.9

What Tables 2.2.1 to 2.2.9 show is the number of seeds out of the twenty five seeds that germinated along with the standard error for each point of each graph. In the control (Table 2.2.1) over the experimental period the rate of standard error appears to be a low. The rate at which the seeds germinated appears to be relatively constant after the first four days where no seeds germinated.

Tables 2.2.2 and 2.2.3 illustrate the number of seeds that germinated when grown in the root extract. Both Consort (Table 2.2.2) and Soissons (Table 2.2.3) exhibit a good level of germination suppression over the first four days before experiencing a reduced level of control. There is a low rate of standard error in Table 2.2.2 suggesting that the results gained are close to being a true representation of the effect of Consort roots on chickweed *Stellaria media*.

Tables 2.2.4 and 2.2.5 illustrate the number of seeds that germinated when grown in the stem extract. Both Consort (Table 2.2.4) and Soissons (Table 2.2.5) show a good level of germination suppression over the first four days before experiencing a reduced level of control. There is again low rate of standard error in both Table 2.4 and 2.2.5 suggesting that the results gained are close to being a true representation of the effect of Consort and Soissons stems on chickweed *Stellaria media*.

Tables 2.2.6 and 2.2.7 illustrate the number of seeds that germinated when grown in the leaf extract. Tables 2.2.6 and 2.2.7 show a good level of germination suppression over the first four days before experiencing a reduced level of control. By simply looking at these two graphs you can see that Soissons leaves have a longer period in which the chickweed seeds are suppressed from germinating although after this period the control breaks down faster than that of the Consort leaves mixture.

Tables 2.2.8 and 2.2.9 illustrate the number of seeds that germinated when grown in the all three plant parts mixed together. The length of the initial germination delay that has been quite apparent in the previous graphs has been reduced. This is probably down to the fact that all three plant parts have been diluted into each other. The standard error has remained low throughout the experiment.







Table 2.2.11



Table 2.2.12



Table 2.2.13

Table 2.2.10 shows the number of seeds that germinated in the root extract compared with the control. The Soissons roots show a reduction in the germination of the chickweed *Stellaria media* seeds. While the consort shows a stimulated germination after the fifth day of the experiment. Form this we can say that Soissons roots express a higher level of allopathic effect on chickweed than Consort roots.

Table 2.2.11 shows the number of seeds that germinated in the stem extract compared with the control. Table 2.2.11 shows that Soissons is the only extract that shows suppression of the number of germination of chickweed *Stellaria media* seeds after the fourth day of the experiment. The chickweed *Stellaria media* seeds in the Consort extract appear to be experiencing stimulated growth but closer observation of the results show it is a steady one extra shoot from the control this would suggest the presence of a several dormant seeds one of the control dishes is effecting the true values of the control values shown.

Table 2.2.12 shows the number of seeds that germinated in the leaf extract compared with the control. Table 2.2.12 again like Tables 2.2.11 and 2.2.10 Soissons is showing the best level of suppression of the germinating seeds. Again like Table 2.2.10 and 2.2.11 Consort appears to be stimulating growth.

To summarise there are variations between the different parts of the plant in controlling the germination of chickweed *Stellaria media* seeds. Soissons has also shown a better

suppression rate than Consort, which in many cases has shown a stimulated growth. Muller. C.H (1965) claims that the effects of allelopathy can suppress or stimulate growth. Another factor that could be causing this is the fact that the chickweed *Stellaria media* is a weed species and so the seed has not come from a plant line that has been selected to have even germination. The stems on the whole show the highest amount of germination suppression. This is interesting as initial expectations were that the roots would have the highest effect as part of the allelochemicals have to be translocated through the water in the soil surrounding the roots plants.

#### Growth Rate





Table 2.2.15





Table 2.2.17





Table 2.2.19



Table 2.2.20

Table 2.2.21



Table 2.2.22

What Tables 2.2.14 to 2.2.22 show the growth rate of the seeds that germinated along with the standard error for each point of each graph. In the control (Table 2.2.14) over the experimental period the rate of standard error appears to be a low. The rate at which the seeds germinated appears to be relatively constant throughout the experiment.

Tables 2.2.15 and 2.2.16 illustrate the rate at which the seedlings grew in the root extract. Soissons roots (Table 2.2.16) appear to have a better suppressing effect than the Consort roots (Table 2.2.15). There is a higher rate of standard error in the Soissons roots (Table 2.2.16).

Tables 2.2.17 and 2.2.18 illustrate the rate at which the seedlings grew in the stem extract. Table 2.2.18 showing the Soissons stems results there is a lower rate of standard error than in Table 2.2.16 which shows the Consort results. The rate of control break down in Table 2.2.18 is slightly slower than the Consort roots (Table 2.2.15).

Tables 2.2.19 and 2.2.20 illustrate the rate at which the seedlings grew in the leaf extract. Table 2.2.20 showing the Soissons leaves experiment results has a higher rate of standard error than the Consort experiment. There is also a faster rate of control break down with Consort leaves than with Soissons resulting in higher shoot lengths by the end of the experiment. Of all three plant parts the leaves have shown the highest rate of suppressing the growth rate of chickweed *Stellaria media* seedlings.

Tables 2.2.21 and 2.2.22 illustrate the rate at which the seedlings grew in the all three plant parts mixed together. Both Table 2.2.21 and 2.2.22 have a low standard error, which is a good indication that the plots on both Tables are close to being a true representation of the shoot growth over the duration of the experiment.







Table 2.2.24



Table 2.2.25



Table 2.2.26

Table 2.2.23 shows the growth rate of the seedlings that germinated in the root extract compared with the control. The Soissons roots show the highest a reduction in the growth rate of the chickweed *Stellaria media* seedlings. Form this we can say that Soissons roots express a higher level of allelopathic effect on chickweed than Consort roots. Previously Consort has shown a stimulated growth rate in this case there would appear to be a slight reduction of the rate of germination this in the field would give a crop an advantage over the weed population as it would allow the crop to develop a canopy before the weed in this case chickweed *Stellaria media* has had a chance to develop a canopy. What this means is that the crop would be able to effectively restrict the rate in which the weed species can photosynthesise reducing the weeds growth/development rate this could eventually result in the weed dieing or growth rate being reduced.

Table 2.2.24 shows the growth rate of the seedlings that germinated in the stem extract compared with the control. The Soissons stems show again the highest reduction in the growth rate of the chickweed *Stellaria media* seedlings. Table 2.2.24 shows that the stems are part of both wheat varieties that has the highest suppressing effect on the growth rate of chickweed *Stellaria media* seedlings.

Table 2.2.25 shows the growth rate of the seedlings that germinated in a mixture of all three plant parts compared with the control. Again in Table 2.2.25 Consort is exhibiting

a stimulated rate of growth of chickweed *Stellaria media*. This is down to the dilution, which occurs when, mixing all three plant parts.

To summarise Consort roots and stems have some control over the growth rate but not the actual germination of the chickweed *Stellaria media* seeds. Soissons on the other hand has shown control in both suppression of germination of chickweed *Stellaria media* seeds and in growth rate of the seedlings. It would appear that the greatest suppression occurs from stem extract and the least from the leaves of both Soissons and Consort.

#### **CONCLUSION**

This dissertation set out to look at whether the effects of allelopathy could be used as an effective control of chickweed *Stellaria media* in winter wheat. The way that this was carried out was by carrying out two experiments along with extensive research of other projects on allelopathy.

The first experiment was just set up to screen for allelopathic effects over several different plant extracts these included plants with known allelopathic abilities such as eucalyptus leaves and pine needles. The second experiment was more focused on winter wheat. The second experiment looked at whether there were varietal differences within species. Experiment one looked at the differences between Consort and Soissons it also looked at whether the three main parts of a plant had different levels of allelopathic effects.

Experiment one showed that all of the plant extracts apart form Consort (wheat) showed a suppression effect on the germination of chickweed *Stellaria media*. Less of the plant extracts showed a suppression effect on the growth rate. Only eucalyptus leaves, couch rhizomes, rice and liquidised pine needles showed control over both germination and growth rate. Experiment one showed that it was possible to control chickweed *Stellaria media* by using the allelochemicals present in plant extracts. It also became apparent that the different plants effected the chickweed *Stellaria media* in different ways, some extracts had a good all round control while some such as the senesced pine needles only controlled the germination and not the growth rate. This maybe down to the type of allelochemical that is present in the plant extract, as there are several different modes of action as previously covered in the literature review.

Experiment two showed that there are varietal differences within wheat species along with variations in control over the three main parts of a plant. In both germination and growth rate Soissons out preformed Consort. From this an assumption can be made that Soissons has higher levels of allelochemicals than Consort. In both Soissons and Consort the stem achieved the highest level of control followed by the roots. What this means it that the highest level of allelopathic potential of a wheat crop is reliant on

rainfall to wash the allelochemicals from the stems into the soil. The second highest levels were found to be located in the root systems this is a far more efficient location as the allelochemicals can be released into the ground water directly from the roots.

Both experiments one and two showed that it was possible to have a low level of control on chickweed *Stellaria media* using a plants natural production of allelochemicals. This in a way answers the question set in the introduction, which was whether allelopathy could be used to control weeds in winter wheat crop. At this point it can be said that it is possible to obtain some control but not enough to be able to solely rely on.

This does not mean that allelopathy cannot be used practically as a weed control in winter wheat. A way in which allelopathy could be perhaps be used is if the allelochemical compounds were produced and mixed with a reduced dose of herbicide. By mixing there are many benefits such as increasing the modes of action which reduces the chance of resistance developing and more environmentally friendly as the allelochemicals are more biodegradable than artificially produced chemicals.

The experiments that were carried out in this dissertation could be improved by using a weed seed that has been selected for even germination. Unlike that of crop seed weed seeds have not gone threw a selection process that ensures that an even germination is achieved. Naturally seeds do not germinate all at the same time this is a survival technique ensuring that some plants are allowed to survive a season. The most common way that a weed seed does this is by having a dormancy period this can be from a week to several years depending on the weed species. Uneven germination has effected the results gained by the experiments, By using perhaps a crop seed such as oil seed rape which is both fast growing and completive like that of the chickweed *Stellaria media* might give more precise results.

Perhaps further research could be carried out to increases the levels of allelochemicals produced by a plant and to where the highest levels of allelochemicals are produced. The most effective way that this could be carried out is by means of genetic manipulation. Sadly due to public pressures this method is unlikely to be adopted in the UK resulting in slower methods such as cross breeding and selecting to be carried out. If it were possible to solely select a variety that was able to control a weed problem like that of selecting disease resistance in crops many benefits would follow. The benefits would not only benefit British farmers and farmers in developed countries but also farmers in developing countries where weeds can pose real problems to crops and in many cases the farmer cannot afford to apply large amounts of herbicide to control weed problems. This potential cost saving would improve arable crop production worldwide and is one of the major improvements that today's agriculture needs.

If it were to be found to be impossible or financially unfeasible to achieve high enough levels of allelochemicals to be produced naturally in order to obtain full weed control. Research could be done into looking at producing allelochemicals artificially and increasing the concentration producing a new range of synthetic herbicides (Pers. Com. 2004). By applying the allelochemicals artificially this would give a more controlled application and more even results across a field. Applying the allelochemicals by means of a sprayer this would over come the reliance of the weather to transport the allelochemicals from the leaves and stems to the soil. The result of this would be a more reliable weed control but with low environmental hazard implications.

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### **APPENDICES**

# Experiment 1 reading's (Germination)

No of Shoots	20-Nov	24-Nov	26-Nov	28-Nov	02-Dec	04-Dec
Water (Control) 1	0	2	2	4	4	4
Water (Control) 2	0	1	2	3	6	6
Water (Control) 3	0	2	2	2	2	2
Rice 1	0	0	0	0	1	1
Rice 2	0	0	0	1	2	2
Rice 3	0	0	0	0	2	3
Green Pine Needles (L) 1	0	1	1	1	2	2
Green Pine Needles (L) 2	0	0	1	3	3	3
Green Pine Needles (L) 3	0	0	1	1	2	2
Eucalyptus Leaves (L) 1	0	0	2	2	2	2
Eucalyptus Leaves (L) 2	0	0	2	2	3	3
Eucalyptus Leaves (L) 3	0	0	1	1	1	1
Wheat (Soissons) (L) 1	0	3	3	5	5	5
Wheat (Soissons) (L) 2	0	1	1	3	3	3
Wheat (Soissons) (L) 3	0	2	3	4	6	6
Wheat (Consort) (L) 1	0	2	2	3	3	3
Wheat (Consort) (L) 2	0	0	0	0	0	0
Wheat (Consort) (L) 3	0	2	2	2	2	3

Couch Rhizomes (L) 1	0	2	2	2	2	2
Couch Rhizomes (L) 2	0	0	0	1	2	2
Couch Rhizomes (L) 3	0	0	1	1	1	2
Eucalyptus Leaves 1	0	1	3	3	3	3
Eucalyptus Leaves 2	0	1	2	3	3	3
Eucalyptus Leaves 3	0	1	1	2	3	4
Senesced Pine Needles 1	0	0	3	3	3	3
Senesced Pine Needles 2	0	0	2	2	3	4
Senesced Pine Needles 3	0	0	1	1	1	2
Green Pine Needles 1	0	1	5	5	6	6
Green Pine Needles 2	0	1	4	6	7	7
Green Pine Needles 3	0	2	4	7	7	8

# <u>APPENDICES</u>

# Experiment 1 reading's (Growth Rate)

22-Nov	24-Nov	26-Nov	28-Nov	02-Dec	04-Dec
0	10	15	20	22	23
0	5	10	13	15	22
0	4	10	15	20	24
0	0	0	0	5	6
0	0	0	1	4	4
0	0	0	2	3	4
	6	9	10	20	21
0	4	10	15	21	23
0					
	7	8	12	18	21
0	0	3	3	10	12
0					
0	1	1	3	12	14
0	0	0	4	9	10
0					
	7	17	25	26	29
0	4	14	23	26	28
0					
	6	13	24	30	33
0					
	8	15	20	25	28
0					
	5	10	18	23	28
0	2	1 5		27	20
0	3	15	22	27	32
	22-Nov 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0	22-Nov       24-Nov         10         0         0       5         0       4         0       0         0       0         0       0         0       0         0       0         0       0         0       0         0       0         0       7         0       0         0       0         0       0         0       7         0       0         0       7	22-Nov       24-Nov       26-Nov         10       15         0       5       10         0       4       10         0       4       10         0       0       0         0       0       0         0       0       0         0       0       0         0       0       0         0       0       0         0       7       8         0       0       3         0       0       0         0       1       1         0       0       0         0       0       0         0       0       0         0       1       1         0       0       0         0       0       0         0       1       1         0       1       1         0       1       1         0       1       1         0       1       1         0       1       1         0       1       1         0       1       1	22-Nov       24-Nov       26-Nov       28-Nov         10       15       20         0       5       10       13         0       4       10       15         0       0       0       0         0       0       0       0         0       0       0       0         0       0       0       15         0       0       0       10         0       0       0       2         0       0       0       10         0       0       0       10         0       1       10       15         0       1       10       15         0       1       10       15         0       1       1       3         0       1       1       3         0       1       1       3         0       1       1       3         0       1       14       23         0       1       14       23         0       1       15       20         0       1       15       20      <	22-Nov         24-Nov         26-Nov         28-Nov         02-Dec           0         10         15         20         22           0         5         10         13         15           0         4         10         15         20           0         0         0         5         0           0         0         0         5         0           0         0         0         14         4           0         0         0         23         3           0         0         0         23         3           0         0         10         20         3           0         -         -         10         20           0         -         -         -         10         10           0         -         -         -         -         10           0         -         -         -         -         -           0         -         -         -         -         -           0         -         -         -         -         -           0         -         -         -<

Couch Rhizomes (L) 1	0	2	10	10	15	18
Couch Rhizomes (L) 2	0	0	2	5	10	16
Couch Rhizomes (L) 3	0	3	4	10	15	20
Eucalyptus Leaves 1	0	6	10	15	20	20
Eucalyptus Leaves 2	0	8	14	. 17	21	21
Eucalyptus Leaves 3	0	4	12	. 14	20	21
Senesced Pine Needles 1	0	0	15	15	20	21
Senesced Pine Needles 2	0	0	14	20	24	24
Senesced Pine Needles 3	0	0	17	23	25	25
Green Pine Needles 1	0	10	10	20	22	23
Green Pine Needles 2	0	8	10	15	18	23
Green Pine Needles 3	0	14	16	24	27	25

### **APPENDICES**

# Experiment 2 reading's (Germination)

Number of shoots	19-Jan	21-Jan	23-Jan	25-Jan	27-Jan	29-Jan	31-Jan
Consort roots (S1)	0	0	0	4	5	7	7
Consort roots (S2)	0	0	2	6	8	11	11
Consort roots (S3)	0	0	1	4	9	10	10
Consort stems (S1)	0	0	0	6	6	9	10
Consort stems (S2)	0	0	0	4	7	9	9
Consort stems (S3)	0	0	4	6	9	9	9
Consort leaves (S1)	0	0	0	4	5	9	9
Consort leaves (S2)	0	0	6	8	14	14	14
Consort leaves (S3)	0	0	0	2	5	6	6
Consort All (S1)	0	0	0	4	8	8	8
Consort All (S2)	0	0	1	6	9	11	11
Consort All (S3	0	0	2	8	9	12	12
Soissons Roots (S1)	0	0	0	3	4	6	7
Soissons Roots (S2)	0	0	0	1	3	7	7
Soissons Roots (S3)	0	0	1	6	10	10	10
Soissons Stems (S1)	0	0	0	1	3	5	5
Soissons Stems (S2)	0	0	0	2	4	6	7

Soissons Stems (S3)	0	0	1	3	5	6	6
Soissons Leaves (S1)	0	0	0	3	3	4	4
Soissons Leaves (S2)	0	0	0	2	5	5	5
Soissons Leaves (S3)	0	0	0	2	8	9	9
Soissons All (S1)	0	0	0	2	4	5	5
Soissons All (S2)	0	0	3	4	4	7	7
Soissons All (S3)	0	0	4	8	8	9	9
Control 1 (S1)	0	0	0	1	2	5	5
Control 2 (S1)	0	0	0	2	4	10	10
Control 1 (S2)	0	0	0	6	8	11	11
Control 2 (S2)	0	0	0	8	10	10	10
Control 1 (S3)	0	0	3	4	7	10	10
Control 2 (S3)	0	0	1	6	8	9	9

# <u>APPENDICES</u>

# Experiment 2 reading's (Growth Rate)

Shoot length							
(mm)	19-Jan	21-Jan	23-Jan	25-Jan	27-Jan	29-Jan	31-Jan
Consort roots (S1)	0	0	0	8	15	26	27
Consort roots (S2)	0	0	5	10	20	20	22
Consort roots (S3)	0	0	4	8	20	20	21
Consort stems (S1)	0	0	0	4	10	15	17
Consort stems (S2)	0	0	0	6	15	20	20
Consort stems (S3)	0	0	10	12	20	22	24
Consort leaves (S1)	0	0	0	10	20	22	26
Consort leaves (S2)	0	0	0	15	27	32	34
Consort leaves (S3)	0	0	6	19	30	30	32
Consort All (S1)	0	0	0	8	15	22	23
Consort All (S2)	0	0	13	15	20	24	24
Consort All (S3)	0	0	5	15	20	30	32
Soissons Roots (S1)	0	0	0	5	6	16	20
Soissons Roots (S2)	0	0	0	7	10	10	12
Soissons Roots (S3)	0	0	3	10	20	25	26
Soissons Stems (S1)	0	0	0	2	8	10	12
Soissons Stems (S2)	0	0	0	10	15	15	17

Soissons Stems (S3)	0	0	8	10	15	20	21
Soissons Leaves (S1)	0	0	0	9	10	15	17
Soissons Leaves (S <u>2)</u>	0	0	0	4	8	9	10
Soissons Leaves (S3)	0	0	4	10	18	25	26
Soissons All (S1)	0	0	0	8	17	20	21
Soissons All (S2)	0	0	3	8	15	20	20
Soissons All (S3)	0	0	9	17	20	20	22
Control 1 (S1)	0	0	0	4	10	11	14
Control 2 (S1)	0	0	0	3	11	12	12
Control 1 (S2)	0	0	0	10	20	30	30
Control 2 (S2)	0	0	0	9	18	20	21
Control 1 (S3)	0	0	15	20	25	30	32
Control 2 (S3)	0	0	10	20	30	32	35