

**Investigation of removal of seminal and lateral root systems and how it affects the Gibberellin synthesis in Scarlet runner bean**

Extended Essay: Biology

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Research question: How does removal of seminal or lateral parts of the root system of Scarlet Runner Bean (*Phaseolus coccineus*) affect the synthesis of Gibberellic acid and therefore the growth of the plant?

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## Abstract

In this experiment the research question was how does removal of seminal or lateral parts of the root system of Scarlet Runner Bean (*Phaseolus coccineus*) affect the synthesis of Gibberellic acid and therefore the growth of the plant?

Beans of *Phaseolus coccinus* were planted and grown until seedlings were about 10 cm tall. Then they were put into three different conditions; the control were nothing was modified, seminal group condition were their lateral root system was removed and lateral condition group were their seminal root (tap root) was removed. The plants were labeled, their stems measured in length and then they were replanted and grown outdoors for 6 weeks. Measurement of all samples stem length was done every week.

The conditions were then compared by their mean overall growth and mean growth for every week and also test of significance and other measurements were done.

The results from the measurements and processing of data was that the growth was retarded in both experimental conditions (seminal condition group and lateral condition group) compared to the control condition group, but at a more significant level in the lateral condition group.

It was then concluded that this could indicate that the location for GA<sub>3</sub> synthesis is in the root system of the plant, and that the main site of synthesis is believed to be in the seminal root (the tap root).

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## Introduction

The plant hormone Gibberellic acid ( $GA_3$ ) is often in research mentioned as a hormone responsible for plant growth, it is said to be a growth regulator. (4)(6)(8)(9). Gibberellic acid is one of the over 110 different types of the hormone group Gibberellins (named  $GA_n$ , where  $n$  is the order in which the special type was discovered). (6)

Gibberellins are said to have an effect on stem elongation, bolting and flowering, germination of seeds and stimulation of germination of pollen as well as growth of pollen tubes. (1)(4)(8)

Gibberellic acid, along with other plant hormones, have also been shown to be synthesized in young, growing meristematic cauline tissue, but high concentrations of Gibberellic acid has also been found in root apices. (1)(8) A theory has been developed that Gibberellins actually are produced in the roots of higher plants and then transported out into the plant through the vascular tissue. (1)

Roots are also responsible for plants uptake of water and nutrients from soil. The root systems consist of the seminal root (from the seed, the tap root) and the lateral root system that later grows from the seminal root or adventitious root growing from the stem. (2)

Studies of common garden bean *Phaseolus vulgaris* indicated that mutation of Gibberellic acid ( $GA_3$ ) resulted in plant dwarfism, but that treatment of the mutation with the hormone gave plants that were the same size as non-mutated control group. (8)

Similar results were obtained, showing gibberellic acids' role as a growth regulator, in experiments of other species including Bernay Phinney et al's investigation of rice and Tamura S et al's research on Gibberellic acid and its properties (4).

Scarlet runner bean (*Phaseolus coccineus*) is chosen for this investigation since effective research was shown on the closely related *Phaseolus vulgaris* and it is also a common crop that germinates and grows quickly and can reach a length of about 3 meters (7). It has also been indicated that if root apices of *Phaseolus coccineus* are removed the concentration of gibberellins is affected (5). It is an annual flowering vine that germinates in the spring and flowers in summer (3) (Figure 1, page 4, below), which was optimal at the time when the experiment was conducted.

### The flowering of the first bean plant of *Phaseolus coccineus* (from the control group)



Figure 1

In this investigation the effect of removal of root apices on growth is to be measured. The aim is to investigate the localization of gibberellin synthesis in the root system. Parts of the root system of *Phaseolus coccineus* seedlings are to be removed to see how the growth is affected. If the results obtained shows a difference in growth in the different conditions the location of gibberellin synthesis can be assumed.

Method:

Beans of *Phaseolus coccineus* were planted in soil from the same bag and put outdoors to germinate and start growing. When seedlings had grown to about 10 cm, all were measured in length, modified for their condition (see below), labeled and replanted. Seedlings were put into three different conditions;

Control condition group; all seedlings were measured and labeled. See figure 3 on page 4.

Seminal condition group; all seedlings were measured, labeled and all roots were cut off except for the seminal root. See figure 4 on page 6.

Lateral condition group; all seedlings were measured, labeled and seminal root was cut off leaving only the lateral root system. See figure 5 on page 6.

The seedlings were all put together outside again and measured every Thursday evening for six weeks (until the first bean plant flowered, see figure 1 on page 4).

**Pictures showing the scarlet runner bean seedlings, their root systems and their respective removal**



Figure 2

Full root system (control condition group):

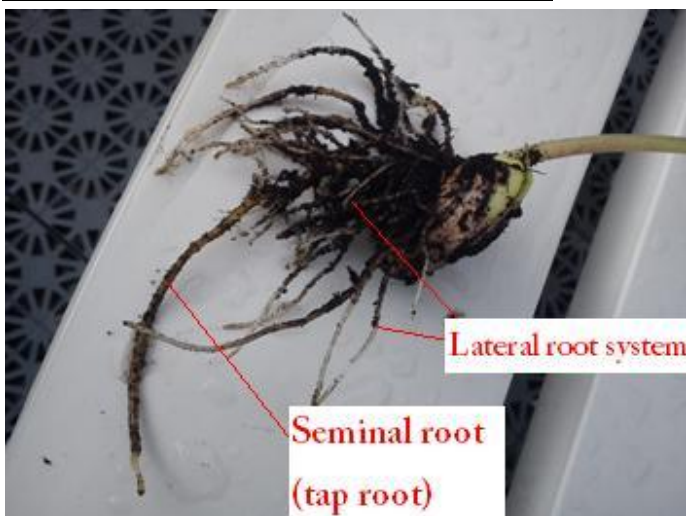


Figure 3

The removed root systems of seminal condition group

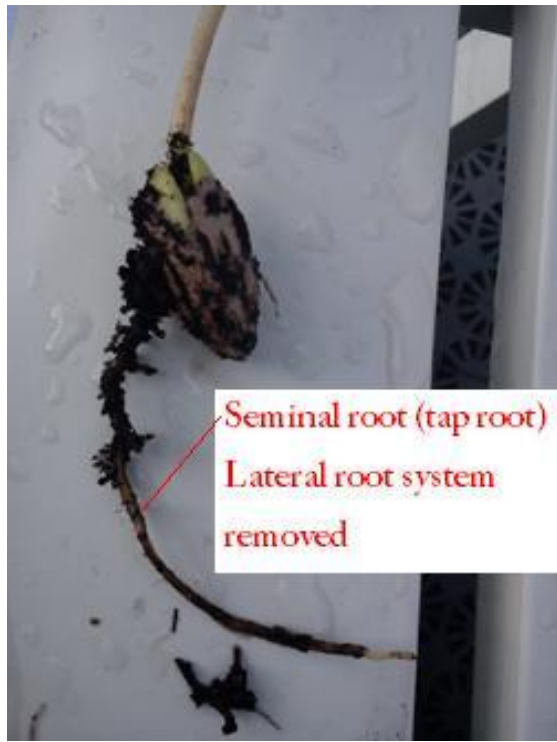


Figure 4

The removed root systems of lateral condition group

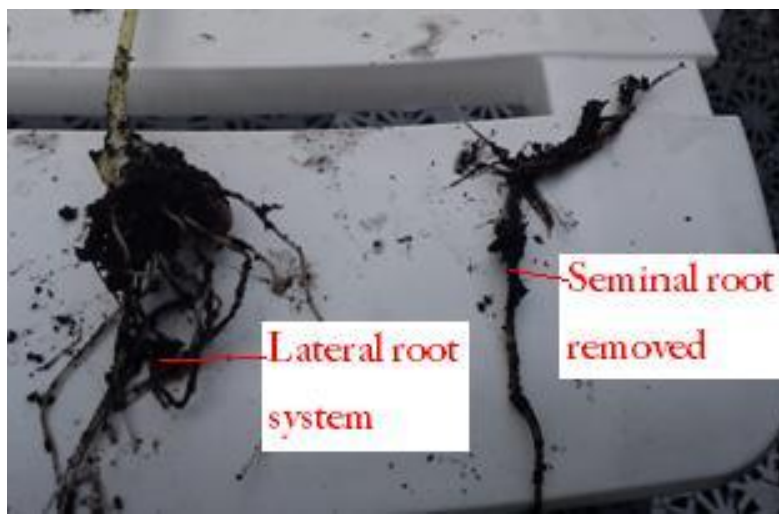


Figure 5

**Raw data collection notation**

The condition groups weekly measurements of length of their respective stems in  $\text{cm} \pm 0,05$  are found in the appendices, table 1,2 and 3 on page 14-17.

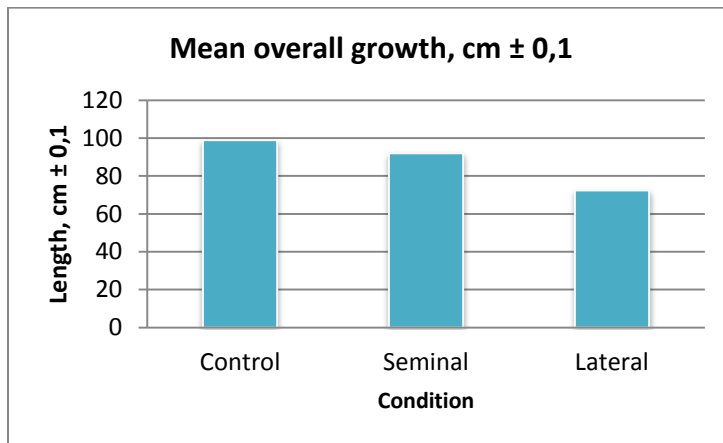
## Data processing and presentation

For each condition the mean overall growth, i.e the mean length plants grown from when the root systems were altered and plants replanted to final measurements (sixth week).

### Mean overall growth for each condition

Condition	Mean overall growth, cm $\pm$ 0,1
Control	98,9
Seminal	92,0
Lateral	72,4

Table 4



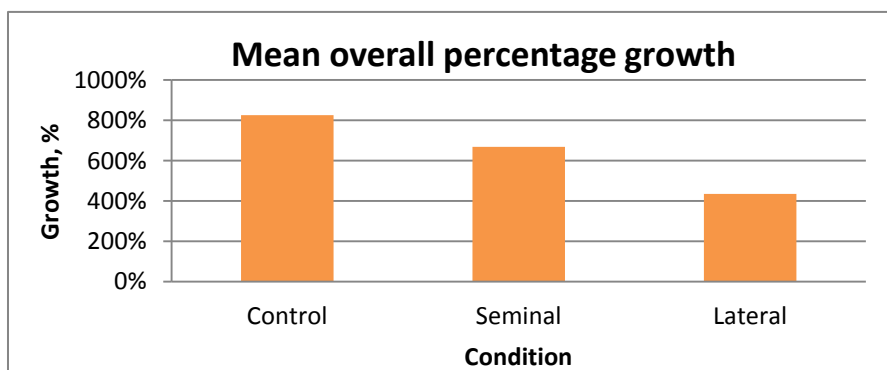
Graph 4

The percentage overall growth was calculated by dividing the mean overall growth by the mean initial length (first measurement when seedlings root systems were cut and replanted) and multiplying by 100.

### Mean percentage overall growth for each condition

Condition	Mean overall percentage growth, %
Control	826%
Seminal	668%
Lateral	435%

Table 5



Graph 5

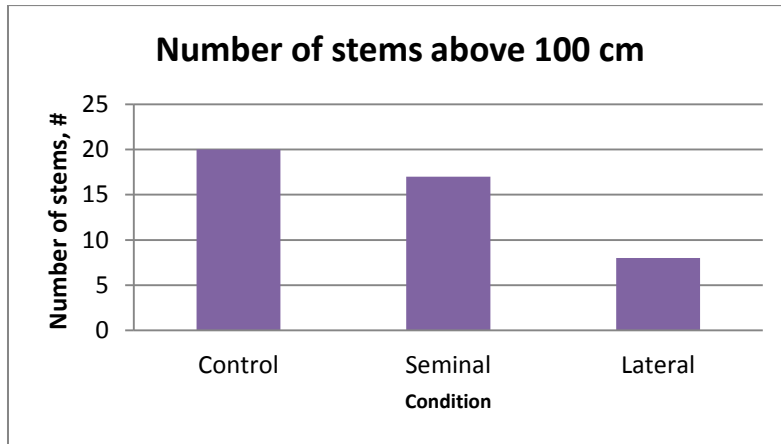


The number of plants stems that had grown to a length over 100 cm was counted.

**Number of plants stem with a length above 100,0 cm for each condition**

Condition	Number of stems above 100 cm
Control	20
Seminal	17
Lateral	8

Table 6



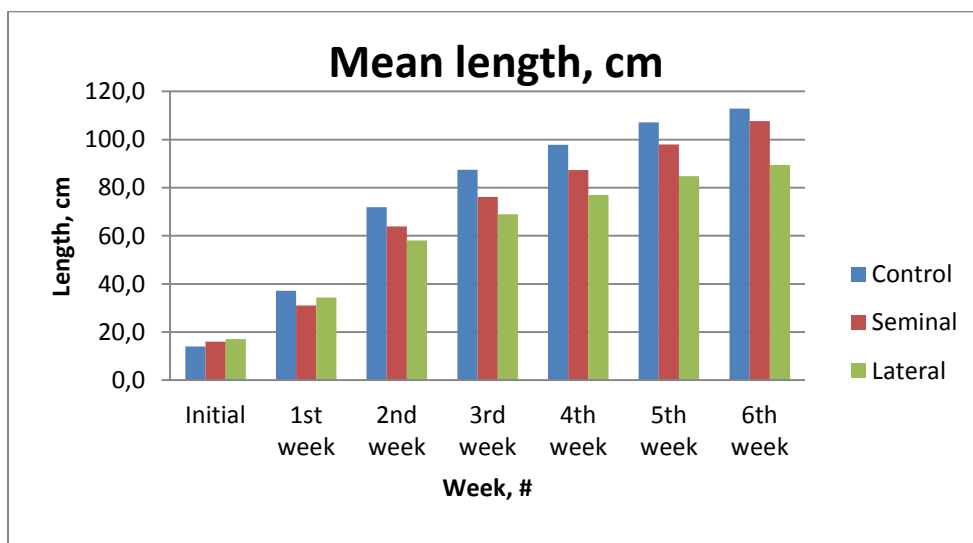
Graph 6

The mean length of the plants of each condition was calculated summing all the plants length up and dividing by the number of plants in each condition.

**Mean length of plants in each condition on every weekly measurement**

Mean length/ cm ± 0,5	Initial	1st week	2nd week	3rd week	4th week	5th week	6th week
Control	14,0	37,1	71,9	87,4	97,9	107,2	112,9
Seminal	16,0	31,0	63,9	76,2	87,3	98,0	107,7
Lateral	17,1	34,4	58,00	69,0	76,9	84,8	89,5

Table 7



Graph 7



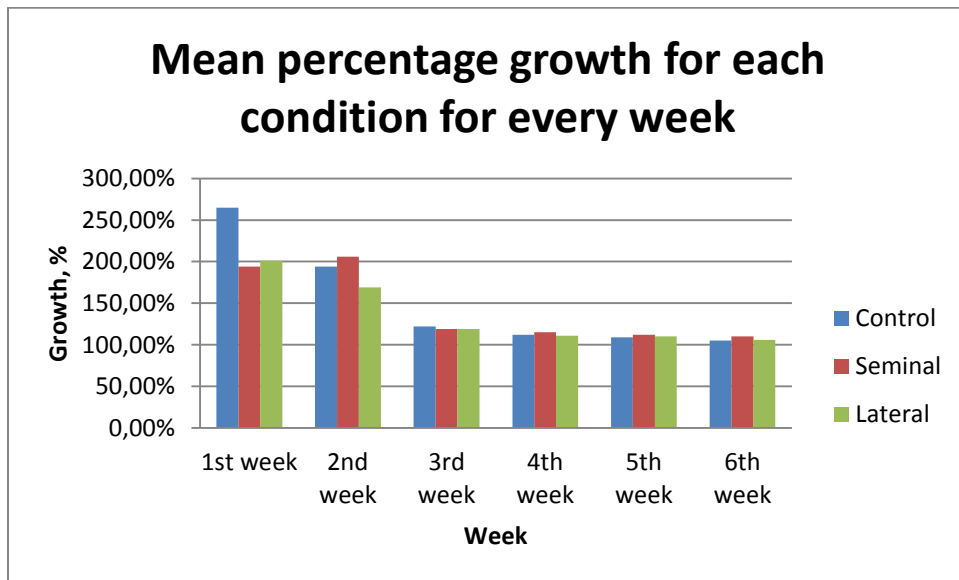
Candidate number: 000511050

The mean percentage growth for each week was calculated by dividing each conditions mean length every week by the previous weeks mean length for that condition and multiplying by 100.

**Mean percentage growth for each condition for every week**

Mean percentage growth	1st week	2nd week	3rd week	4th week	5th week	6th week
Control	265,00%	194,00%	122,00%	112,00%	109,00%	105,00%
Seminal	194,00%	206,00%	119,00%	115,00%	112,00%	110,00%
Lateral	201,00%	169,00%	119,00%	111,00%	110,00%	106,00%

Table 8



Graph 8

**Standard deviation calculations:**

Calculated on GDC (Texas Instrument, TI-84 Plus) using the overall growth for each sample.

Control: 23,6

Seminal: 24,9

Lateral: 18,5

Analysis of values in Analysis of data, page 11.

### **T-test**

Calculated on GDC (Texas Instrument, TI-84 Plus) using overall growth for each sample.

Comparing difference between control and seminal group:

$t$ : 1,117

Degrees of freedom:  $28+27-2=53$

Probability ( $p$ ) that chance alone could produce the difference, calculated on GDC:

0,134= 13,4%

Comparing difference between control and lateral group:

$t$ : 4,653

Degrees of freedom:  $28+27-2=53$

$p$ : 0,0000119=0,001%

Comparing difference between seminal and lateral group:

$t$ : 3,229

Degrees of freedom:  $27+27-2=52$

$p$ : 0,0112=1,12%

### **Analysis of data**

From the calculations and measurements made, it can be concluded that the greatest growth was seen in the control condition group (826% overall growth as seen in table 5, page 7), followed by the seminal condition group (668% overall growth) and the smallest growth was seen in the lateral condition group (435% overall growth). These results indicate that the removal of the root systems decreases the growth rate, especially if the seminal root system is removed, as it was for the lateral condition group. This could also give evidence of that gibberelin synthesis occurs in the root system and therefore the removal of the location of synthesis results in a retarded growth rate since there is a lack of growth hormone.

Table 7 and graph 7 on page 8, showing the mean length (growth) for each condition every week shows that the lateral condition group was actually the 'tallest' group to begin with, but after their seminal root had been removed the mean growth was significantly lower than that of the control and seminal condition group throughout the whole experiment.

The seminal condition group started off having a greater initial mean length than control condition group (table 7 and graph 7, page 8), but after removal of the lateral root system, had the lowest growth rate (194 % growth, seen in table 8 and graph 8, page 9) compared to control and lateral condition group on the measurements after 1 week. After the first weeks' measurement the seminal condition group had a mean percentage growth of 206% (table 8 and graph 8, page 9) to the second week, which were 12 percent units more than the control condition groups' and 37 percent units more than the lateral condition groups'. From then on the mean growth rate for the seminal condition group seems to have been stabilized but mean growth was always lower than that of the control condition group.

The control condition group started off as the group with the shortest initial length but had a rapid growth rate of 265% mean percentage growth (table 8 and graph 8, page 9) to the first weeks' measurement and had then the highest mean length (table 7 and graph 7, page 8) throughout the

whole experiment. To the second week's measurement the growth continued to increase with a 194% mean growth with a mean length of  $71,9 \text{ cm} \pm 0,05$  which at that point made the control condition group 12% ( $8,0 \text{ cm} \pm 0,1$ ) taller than seminal condition group and 24% ( $13,9 \text{ cm} \pm 0,1$ ) taller than the lateral condition group. From then the mean growth continued to increase at a stable rate, the control condition group being the group with the highest mean length at each week's measurement, ending up with a mean length of  $5,2 \text{ cm} \pm 0,1$  (5,0%) greater than seminal condition group and  $23,4 \text{ cm} \pm 0,1$  (26%) greater than lateral condition group.

Also the control condition group had over twice as many (150%) stems above 100 cm of length, as seen in table 6 and graph 6 on page 8, than what the lateral condition group, and 18% more than seminal condition group.

In the lateral condition group the overall growth for each sample is clustered more closely to the mean (with a standard deviation of 18,5) than that of the control condition group (s.d=23,6) and seminal condition group (with a standard deviation of 24,9), as seen from the Standard Deviation calculations on page 9. This could indicate that the overall growth is more or less the same for each sample if the main location for gibberellin synthesis is removed, which gives more reason to believe that the growth hormone synthesis is located in the seminal root.

Since the samples in the seminal condition group had their individual overall growth more spread around the mean for the group, it could show that the lateral root system (which was removed in the seminal condition group) indeed is a location for gibberellins synthesis, but not the main one, and therefore the growth might not be affected to the same extent as it did in the lateral condition group where the seminal root was removed. Then the removal of these 'smaller' synthesis sites would not retard the growth as significantly and hence the consistency in growth rate is not even in the group, but more individual. Though the control condition group had a pretty high standard deviation (23,6) as well, it may just be the fact that not all plants grow equally, which is true for various plant species. Since the standard deviation for the lateral condition group was 12% smaller than that of the control condition group and 16% smaller than the seminal condition group it may be concluded that there is a more even spread of overall growth when growth is retarded, by the removal of the seminal root system, that thus possible is the main location of gibberellic acid ( $\text{GA}_3$ ) synthesis.

From the t-test values on page 10, it can be seen that there is a very low probability (0,001%) that chance alone would cause the difference between the overall growth of the samples in control condition group and lateral condition group. This shows that the removal of the seminal root did show a significant change in the growth of the plants.

For the seminal condition group compared to the control condition group there is a much higher (13,4%) probability that the difference in growth is due to chance alone, but it is still rather low and hence there is still belief that there is a difference in the overall growth if the lateral root system is removed from the plants, and thus it is still believed to have some locations of  $\text{GA}_3$  synthesis, but not being the main site.

Between the lateral and seminal condition group there is a 1,12% probability that chance alone caused the difference in growth. From this very low value it can be deduced that the difference in overall growth between the two experimental conditions is significant. This furthermore supports the outcome that what appears to be the main location for gibberellins synthesis is indeed the seminal root system (the tap root) and that the removal of it causes significant growth retardation.

### **Conclusion and evaluation:**

From this investigation growth retardation can be seen in both experimental conditions, especially in the lateral condition group where the seminal (tap) root was removed. This could indicate that the synthesis of the growth hormone gibberellin ( $GA_3$ ) is located in the roots, especially in the seminal system.

In the other experimental condition, seminal condition group, the lateral root system was removed. For this condition growth retardation could also be seen but not to the same extent and significance as for the lateral condition. Hence it is to be believed that some gibberellin synthesis is located in the lateral root system, without being main site.

The values from the t-test (page 10) indicate that the differences are significant (to a 95% degree for lateral condition compared to both seminal and control condition group and to a 85% degree for seminal compared to control condition group), but the growth of a plant can perhaps be determined by more factors than just the availability of  $GA_3$ . Other growth hormones might be responsible for different stages in the growth cycle of a plant. From the table and graph showing mean growth rate of the conditions for each week (table 8 and graph 8, page 9), it can be seen that the biggest difference in growth rate occurred already in the first two weeks after removal of the different root systems. Hence the gibberellins may only be responsible for initial growth rate but later it is affected by different factors. Perhaps growth hormones synthesized in the leaves and stem contribute to the growth rate in later phases.

This could also indicate that maybe the main initial synthesis of gibberellins is located in the roots but then as time goes by other parts of the plant continues synthesizing gibberellins and then in greater amounts if there is a lack of  $GA_3$ . When then the main locations where removed the other locations began synthesizing more gibberellins, and that is why the main differences in growth rate and length are in the first couple of weeks. After that the synthesis of gibberellins may have reached the right amount necessary for the plant growth and as can be seen from the results, the percentage growth rate for all conditions remain at a very similar level (table 8 and graph 8, page 9).

Amount of sunlight, nutrients and pH of the soil, water distribution, temperature and wind was equal for all plants since they were grown at the same place, at the same time, planted in the same soil and given the same amount of water. These factors may be tried to be controlled, wind and sunlight might have been greater for the plants at the 'edges' of the pack, i.e. the plants that were in the middle might have obtained less sunshine or less wind and this might have affected their growth for them. But since there is big differences in overall growth trend between the condition groups and a lot of samples being used this external factor is not considered to cause a significant difference, causing systematic errors. The plants were evenly spread out when planted so no group would be located in the middle and the other two on the edges, then all conditions would have an equal spread of locations in the pack of plants.

The random errors that can be caused by measuring the length of each plants stem is put into the uncertainties of the measurement, which was at a very low level and did not affect the results at a significant level. Eyesight and misreading done by the experimenter might have occurred, but since the sample size was large and there were no real extremes for the conditions and the Standard Deviation values were at fairly similar degrees this seems to not have caused any significant difference for the outcome. To be sure of that no random errors caused an effect, experiment can be repeated.

The method carried out was pretty simple and straightforward, the conditions were easy to create and overall very effective. The believed locations of GA<sub>3</sub> synthesis were removed and hence deductive reasoning can be used to rule out where it was.

There could have been more controlled variables in the method, such as the plant could have been grown in a greenhouse (no wind affecting outcome) and all plants could be separated into more neatly rows (no sunlight distribution affecting outcome). The amounts of water that the plants were given could have been measured more carefully and also each plant could have gotten its own pot to grow in so that possible competition for nutrients from soil and water could not have affected the results.

This experiment cannot for sure deduce the location of synthesis of gibberelic acid or that it specifically affects the growth but there are clear indications in the growth rate differences between the experimental condition groups and the control condition group that supports the theory of gibberellins being synthesized in roots, also from this experiment shown, especially in the seminal root, and then distributed to the rest of the plant.

Considering how plant cells grow by absorbing water by osmosis through their cell wall that is created after cell division. The stem cells of a plant then get their water supply from root cells. Hence when the roots were cut off this may have reduced the amount of water that the stem can obtain from the soil, limiting the elongation and growth of the stem. To be certain that this was not a systematic error affecting the growth of the stems and therefore the results, the experiment can be repeated and the stem cells could be sectioned and measured and compared by mean lengths for the different conditions.

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**Appendices****Raw data collection:**

Measurements of seedlings length (cm  $\pm$  0,05) every Thursday over six weeks from June 23<sup>rd</sup> to August 4<sup>th</sup>.

Table 1, control condition group:

Length/cm  $\pm$  0,05

#	23-jun	30-jun	07-jul	14-jul	21-jul	28-jul	04-aug
1	13,2	22,9	49,1	76,2	90,5	92,5	107,2
2	20,5	40,1	78,7	100,3	110,0	125,0	132,5
3	17,8	36,3	84,6	93,6	124,3	129,4	138,5
4	11,3	30,1	55,5	90,6	123,0	165,2	173,5
5	8,0	27,8	45,4	60,7	90,5	126,8	139,7
6	15,1	30,2	48,8	60,3	61,9	71,4	95,6
7	15,4	40,4	-	-	-	-	-
8	12,5	27,3	61,2	67,8	68,1	76,4	79,0
9	17,3	39,0	75,0	97,6	104,5	112,3	115,0
10	12,2	31,8	74,3	92,0	99,7	103,6	119,7
11	20,8	50,0	111,7	132,2	144,4	152,4	157,0
12	9,9	30,5	72,0	82,3	90,0	92,8	99,9
13	7,5	32,4	75,4	68,0	72,4	73,7	74,3
14	9,6	-	-	-	-	-	-
15	14,0	31,8	57,9	76,4	85,0	87,5	88,2
16	16,9	39,5	75,3	92,3	127,3	129,4	130,5
17	12,8	42,3	109,5	116,7	129,8	134,1	139,7
18	18,3	50,6	68,1	91,1	106,6	110,5	112,3
19	20,1	43,7	79,2	95,0	96,9	107,7	111,4
20	7,3	30,1	66,3	75,0	75,9	76,7	77,3
21	16,6	-	-	-	-	-	-
22	3,2	28,3	82,0	105,9	107,0	107,2	107,8
23	13,4	39,0	81,9	93,9	95,6	103,5	105,7
24	13,8	46,7	73,1	103,1	119,5	127,8	132,2
25	18,3	54,3	95,1	96,4	99,7	114,8	126,5
26	20,1	46,7	72,6	88,3	90,9	96,2	104,1
27	16,9	56,1	88,5	116,0	120,0	122,8	124,1
28	10,7	39,9	70,2	84,3	89,6	97,7	102,2
29	13,0	30,5	53,5	54,6	62,5	74,6	76,2
30	14,7	38,3	69,4	84,5	86,0	88,7	90,0
31	12,7	22,9	37,7	52,6	70,0	99,7	101,6



Candidate number: 000511050

Table 2, seminal condition group

Length/cm  $\pm$  0,05

#	23- jun	30- jun	07- jul	14- jul	21- jul	28- jul	04- aug
1	14,2	33,5	82,7	105,9	119,0	126,4	138,7
2	18,1	33,9	42,0	41,2	47,2	50,0	80,5
3	15,6	26,7	36,2	42,3	46,0	60,2	82,7
4	17,0	39,3	66,9	83,6	90,7	102,3	112,2
5	8,7	24,7	72,9	93,2	95,0	-	-
6	9,3	16,6	33,2	-	-	-	-
7	14,7	25,8	34,0	44,9	80,5	90,7	106,4
8	22,9	37,9	80,1	81,4	97,5	104,3	105,2
9	19,6	27,4	48,2	46,1	54,6	60,2	82,2
10	16,2	30,4	45,1	57,1	78,0	84,0	89,9
11	12,1	27,0	55,5	60,9	73,8	90,0	115,2
12	20,1	39,3	97,1	114,3	120,6	129,0	133,3
13	11,0	31,2	52,9	59,0	66,6	70,8	72,5
14	15,6	29,1	59,3	57,6	63,9	79,6	89,3
15	16,3	25,7	84,0	104,2	118,8	132,2	141,9
16	17,7	30,3	95,2	56,5	67,0	70,0	79,4
17	20,9	36,0	82,2	82,1	87,0	102,6	108,3
18	14,0	33,1	80,6	132,7	142,5	158,3	164,2
19	14,6	33,5	55,7	62,1	67,5	74,5	76,3
20	7,3	33,3	73,9	113,5	121,0	123,7	129,9
21	5,5	18,2	64,0	89,5	93,5	97,9	98,2
22	19,1	39,7	48,3	60,9	74,2	88,9	103,1
23	23,8	43,5	76,2	104,2	105,8	107,2	112,9
24	11,4	26,1	79,0	111,1	121,0	133,5	141,0
25	10,9	17,0	31,1	40,6	62,5	122,4	128,9
26	12,7	29,2	52,8	82,6	102,0	108,0	112,9
27	16,6	32,1	74,2	96,1	102,0	103,1	106,0
28	15,3	24,3	41,9	44,0	53,3	67,9	72,4
29	20,6	32,4	85,1	82,8	99,7	107,5	123,5

Table 3, lateral condition group  
length/cm  $\pm$  0,05

#	23-jun	30-jun	07-jul	14-jul	21-jul	28-jul	04-aug
1	18,9	35,6	62,4	75,9	93,5	100,3	100,3
2	17,0	37,1	76,3	100,5	101,0	103,6	112,3
3	20,8	41,0	62,3	74,1	79,0	81,8	88,0
4	12,1	28,0	42,7	46,2	45,5	46,7	57,7
5	16,2	35,4	69,0	73,4	82,3	94,3	98,6
6	21,2	50,2	85,5	95,4	98,1	99,9	101,0
7	18,9	36,2	68,1	77,4	83,4	90,2	98,8
8	18,6	28,6	32,5	42,0	66,8	75,0	82,2
9	18,9	32,3	43,2	42,9	60,6	77,2	82,9
10	19,2	38,0	66,6	89,2	104,5	131,6	139,6
11	25,4	49,7	72,2	90,7	93,0	96,2	98,5
12	24,3	42,3	80,1	83,3	-	-	-
13	15,4	36,1	61,1	67,4	75,5	83,2	89,9
14	15,1	29,1	45,9	51,9	59,0	63,7	68,5
15	18,8	27,2	38,4	41,6	49,0	64,5	70,2
16	13,1	39,2	65,0	80,5	91,2	109,7	117,5
17	19,7	38,5	66,6	87,7	91,8	98,6	101,2
18	16,3	33,4	69,1	95,0	98,0	103,1	105,7
19	15,5	37,2	60,9	61,2	69,3	85,9	79,9
20	15,4	35,7	39,2	50,8	52,7	63,8	65,5
21	14,6	31,0	74,5	66,2	75,0	80,7	93,0
22	11,0	27,7	60,9	63,2	64,3	65,9	67,5
23	15,1	26,3	37,3	41,6	50,8	62,5	74,1
24	19,9	40,9	69,7	92,6	93,3	96,0	96,3
25	10,9	23,5	43,7	-	-	-	-
26	13,7	25,7	35,2	40,5	53,8	56,4	59,5
27	21,1	29,1	36,2	52,3	76,0	85,1	87,0
28	17,6	30,0	62,9	64,5	68,5	70,0	72,2
29	12,4	30,0	61,2	98,4	101,5	105,0	107,8