



## Assessing the regression of characters in shoot induction Of potatoes (*Agria* cultivar)

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### Abstract:

To evaluate the effect of six combined hormones and vitamins as treatments on some of traits in potatoes cultivar (*Agria*), an experiment was conducted in 201۴-201۵ in a randomized complete block design with three replications in laboratory condition. Linear and quadratic regression analysis showed that between the two evaluated traits ( $y$  = main shoot length,  $x$  = number of secondary shoot) there was a significant relationship as follows:  $y=75/278-2/087x$  and  $y=76/546-2/699x+0/061x^2$ . The above equation shows that by increasing the number of secondary shoot, main shoot length decreases. Linear and third degree regression analysis showed that between the two evaluated traits ( $y$  = the maximum of secondary shoot length,  $x$  = number of secondary shoot) there was a significant relationship as follows:  $y=11/621-0/571x$  and  $y=4/179+5/816x-1/295x^2-0/074x^3$ . The above equation shows that by increasing the number of secondary shoot, the maximum of secondary shoot length decreases. Quadratic regression analysis showed that between the two evaluated traits ( $y$  = the maximum of secondary shoot length,  $x$  = number of secondary shoot) there is no significant relationship as



follows:  $y=8/307+1/028x-0/159x^2$ . The above results show that by increasing the number of secondary shoot, the maximum of secondary shoot length may be increased or reduced. Linear and quadratic regression analysis showed that there was a significant relationship between the two traits evaluated ( $y$  = the minimum of secondary shoot length,  $x$  = number of secondary shoot) as follows:  $y = 8 / 844-0 / 774x$  and  $y=7/041+0/096x-0/086x^2$ . Above relationship shows that by increasing the number of secondary shoot, the minimum of secondary shoot length decreases. Based on regression analysis between the third grade of two traits showed that ( $y$  = the minimum of secondary shoot length,  $x$  = number of secondary shoot) there was a significant relationship between them ( $y=2/791+5/025x-1/256x^2-0/076x^3$ ). Linear and quadratic regression analysis showed that between the two evaluated traits ( $y$  = the mean of secondary shoot length,  $x$  = number of secondary shoot) there was a significant relationship as follows:  $y=10/781-0/786x$ ,  $y=8/383+0/3714x-0/115x^2$ . The above equation shows that by increasing the number of secondary shoot, the mean of secondary shoot length decreases. Quadratic regression between the two above traits was significant at 1 % ( $y=4/126+5/306x-1/286x^2-0/076x^3$ ). The results showed that among the two traits mean secondary shoot length and number of secondary shoot first, second, and third grade regressions, was significant at 1% level.

**Keywords:** hormonal compounds shoot induction, solidarity, potatoes



## Introduction

Potato is one of the most valuable herbs in the world. This plant cultivated in 140 countries and at an area equivalent to 20 million hectares. The most allocated cultivation areas of potatoes in the world are located in Asia and Europe that China is the largest area under potato cultivation and about 25 percent of the world's potatoes are produced in China (Mikeny, 2006). Potato production in Iran has special value. Iran ranks twelfth in the world in terms of potato production and in Asia, after China and India are in third place (FAO, 200). Potato has the supply of seeds, fertilizers, agricultural operations, pest control and labor in different areas, and is an expensive product. Thus the average cost of this product is high (Hooker, 1990; Seabrook, 2005). Since the potato propagation by non-sexual organs (glands and micro-glands) and propagation by seed to occur, the availability of disease-free plants and tubers are important. So in addition to classical and traditional methods, using modern techniques of genetic engineering and tissue culture, is the good news in the quickest way to potato breeding purposes. Reservation, germplasm conservation, healthy and virus-free plants, saving time and costs, including application of tissue culture of haploid production (Bajaj, 1987).

Due to the sensitivity of potato to viruses, production of virus-free plants through in vitro cultivation and their propagation, leads to reduced costs and increased performance (Fatima et al., 2005). Meristem culture for production of virus-free plants for the first time, were used by Morel and Martin 60 years ago. (Quote from Fatima et al., 2005; Espinoza et al., 1984).

In vitro propagation of potato (production of virus-free plants, micro-glands) are reliable methods for seed production, is free from any contamination (Al-Safadi et al., 2000). Due to this, in vitro propagation of virus-free plants is important (Hussain et al., 2006; Taboriet al., 1999). In vitro micro-glant production is used for the first time as a tool for solving problems in potato pathology (Coleman et al., 2001), by culturing individual nodes with lateral buds to produce virus-free were in potato seed tubers (Gopal et al., 2004; 1998). Since the potato plantlets in vitro at a rapid rate during transportation, re-cultivation will be destroyed, therefore, to provide an alternate source to decrease damage during cultivation seems to be necessary.

This paper is optimized for medium branch and plant regeneration in vitro on MS medium with BAP concentration was performed using three vitamins and MS medium, and finally the correlation between traits were assessed.

## Materials and methods

In this survey, firstly the MS medium was prepared and potatoes single-node cuttings were planted. To prepare the medium, ingredients for 6 solution's stock includes macro elements, micro elements, KI, CaCl<sub>2</sub>, Fe and vitamins were prepared and was kept in the fridge. To prepare the medium, the required stock solution is removed and with combinations of growth



regulators were added to the medium. Volume of the solution become up to 1000 ml and depending on the medium 30 gr of sucrose is added. After the addition of 0.1 grams per liter Myo inositol, solution *Ph.* by using a NaOH or HCl was adjusted to 8.5. Finally 8 gr agar was added to the medium. Hormonal treatment used in this study for the shoot induction, cytokinins, Pyridoxine, Thiamine, Biotin, respectively. BAP to dissolve completely in water a few drops to NaOH or KOH was used. Single-node scion taken from the middle of the shoot, in order to have homo genic explants, medium salts containing MS, hormonal treatments and vitamins were transferred.

In this experiment to study shoot induction and roots, under conditions of darkness and light in a medium consisting of salts of MS, vitamins treatments and BAP were studied. Cultures were maintained in 16 h light and 8 hours in the dark at  $2 \pm 25^{\circ}$  C for one month.

In this experiment, effect of hormonal treatments, vitamins on the main shoot length was conducted on number of secondary shoot, the maximum number of secondary shoot, the maximum number of secondary shoot and the mean of secondary shoot length of potato in a completely randomized design with 3 replications.

Table 1. Description of the treatments

Treatment No.	Treatments(Hormone Levels)	Description of the treatments
0	Control	(0ppm)
1	PBA2	2ppm cytokinin
2	PBA3	3ppm cytokinin
3	PBA4	4ppm cytokinin
4	PBA5	5ppm cytokinin
5	P5	5ppm pyridoxine
6	P20	20ppm pyridoxine
7	T5	5ppm thiamin
8	T20	20ppm thiamin
9	B5	5ppm biotin
10	B10	10ppm biotin
11	P5 PBA2	2ppm cytokinin + 5ppm pyridoxine
12	P20 PBA2	2ppm cytokinin + 20ppm pyridoxine
13	T5 PBA2	2ppm cytokinin + 5ppm thiamin
14	T20 PBA2	2ppm cytokinin + 20ppm thiamin
15	B5 PBA2	2ppm cytokinin + 5ppm biotin
16	B10 PBA2	2ppm cytokinin + 10ppm biotin
17	P5 PBA3	3ppm cytokinin + 5ppm pyridoxine
18	P20 PBA3	3ppm cytokinin + 20ppm pyridoxine
19	T5 PBA3	3ppm cytokinin + 5ppm thiamin
20	T20 PBA3	3ppm cytokinin + 20ppm thiamin
21	B5 PBA3	3ppm cytokinin + 5ppm biotin
22	B10 PBA3	3ppm cytokinin + 10ppm biotin





23	B5 PBA4	4ppm cytokinin + 5ppm biotin
24	P20 PBA5	5ppm cytokinin + 20ppm pyridoxine
25	T5 PBA5	5ppm cytokinin + 5ppm thiamin
26	T20 PBA5	5ppm cytokinin + 20ppm thiamin
27	B5 PBA5	5ppm cytokinin + 5ppm biotin
28	B10 PBA5	5ppm cytokinin + 10ppm biotin
29	P5 PBA4	4ppm cytokinin + 5ppm pyridoxine
30	P20 PBA4	4ppm cytokinin + 20ppm pyridoxine
31	T5 PBA4	4ppm cytokinin + 5ppm thiamin
32	T20 PBA4	4ppm cytokinin + 20ppm thiamin
33	B5 PBA6	6 ppm cytokinin + 5ppm biotin
34	B10 PBA4	4ppm cytokinin + 10ppm biotin
35	P5 T5 B5 PBA2	2ppm cytokinin + 5ppm biotin + 5ppm thiamin + 5ppm pyridoxine
36	P20 T20 B10 PBA2	2ppm cytokinin + 10ppm biotin + 20ppm thiamin + 20ppm pyridoxine
37	P20 T5 B10 PBA2	2ppm cytokinin + 10ppm biotin + 5ppm thiamin + 20ppm pyridoxine
38	P5 T5 B5 PBA3	3ppm cytokinin + 5ppm biotin + 5ppm thiamin + 5ppm pyridoxine
39	P20 T20 B10 PBA3	3ppm cytokinin + 10ppm biotin + 20ppm thiamin + 20ppm pyridoxine
40	P20 T5 B10 PBA3	3ppm cytokinin + 10ppm biotin + 5ppm thiamin + 20ppm pyridoxine
41	P5 T5 B5 PBA4	4ppm cytokinin + 5ppm biotin + 5ppm thiamin + 5ppm pyridoxine
42	P20 T20 B10 PBA4	4ppm cytokinin + 10ppm biotin + 20ppm thiamin + 20ppm pyridoxine
43	P20 T5 B10 PBA4	4ppm cytokinin + 10ppm biotin + 5ppm thiamin + 20ppm pyridoxine
44	P5 T5 B5 PBA5	5ppm cytokinin + 5ppm biotin + 5ppm thiamin + 5ppm pyridoxine
45	P20 T20 B10 PBA5	5ppm cytokinin + 10ppm biotin + 20ppm thiamin + 20ppm pyridoxine
46	P20 T5 B10 PBA5	5ppm cytokinin + 10ppm biotin + 20ppm thiamin + 20ppm pyridoxine

T (Thiamin) = Vitamin B1

P (Pyridoxine) = Vitamin B6

PBA (Cytokine) = Benzyl -9- (2-Tetrahydropyran-1-yl) 9- H purine

B (Biotin) = 5-[(3aS, 4S, 6aR)-2-oxohexahydro-1H-thieno [3, 4-d] imidazol-4-yl] pentanoic acid

After sufficient growth of shoots (4 weeks after planting) notes were recorded traits including: main shoot length, number of secondary shoot, the maximum number of secondary shoot, the minimum number of secondary shoot and the mean of secondary shoot length. Length was measured with a ruler. Data in completely randomized design was analyzed with SPSS software. Regression equations between variables was performed by using SPSS software and multiple linear STEPWISE regression method for input and output variables and the probability of F, respectively, 5% and 1% were considered.



## Results and discussion

### Polynomial regression

**y = main shoot length with x = number of secondary shoot**

Linear regression analysis (Tables 1 and 2) showed significant correlation between the two evaluated traits as follows:

$$y=75/278-2/087x$$

y = main shoot length

x = number of secondary shoot

The above equation shows that with the increasing number of secondary shoot, main shoot length decreases. Quadratic regression (Tables 3 and 4) showed insignificant correlation between the two evaluated traits as follows:

$$y=76/546-2/699x+0/061x^2$$

y = main shoot length

x = number of secondary shoot

Tables 5 and 6 show the quadratic regression that indicates insignificant correlation between the two evaluated traits follows:

$$y=75/460-1/440x-0/238x^2-0/019x^3$$

The above results indicate that the main shoot length and number of secondary shoot between the two variables are linearly related and by increasing the main shoot length, number of secondary shoots decrease.

Figure 1 graph shows linear, quadratic and cubic regressions of the two evaluated traits.

**y = the maximum of secondary shoot length with x = number of secondary shoot**

Linear regression analysis (Table 1 and Table 3) showed significant correlation between the two evaluated traits as follows:

$$y=11/621-0/571x$$

y = the maximum of secondary shoot length

x = number of secondary shoot

The above equation shows that by increasing the number of secondary shoot, the maximum of secondary shoot length decreases.

Quadratic regression (Tables 3 and 4) showed significant correlation between the two evaluated traits as follows:



$$y=8/307+1/028x-0/159x^2$$

y = the maximum of secondary shoot length

x = number of secondary shoot

The above equation shows that among the two evaluated traits there was a significant quadratic regression and by increasing the number of secondary shoot, the maximum of secondary shoot length may be increased or reduced. Tables 5 and 6 show the results of regression analysis of the third degree that there is significant correlation between the two evaluated traits as follows:

$$y=4/179+5/816x-1/295x^2-0/074x^3$$

y = the maximum of secondary shoot length

x = number of secondary shoot

This relationship indicates that there is third degree regression equation among two evaluated traits and to increase a character trait may increase or decrease. Figure 2 shows the graph of first, second and third regressions among the two evaluated traits. The results indicate that between the number of secondary shoot and the maximum of secondary shoot length variables, there exist third grade regression and depending on the value of the attribute x (number of secondary shoot), the maximum of secondary shoot length may be increased or reduced.

**y = the minimum of secondary shoot length with x = number of secondary shoot**

Linear regression analysis (Table 1 and Table 2) showed that between the two evaluated traits there is significant correlation as follows:

$$y=8/844-0/774x$$

y = the minimum of secondary shoot length

x = number of secondary shoot

This relationship shows that by increasing the number of secondary shoot, the minimum of secondary shoot length decreases. Quadratic regression analysis (Tables 3 and 4) showed that between the two evaluated there is significant correlation as follows:

$$y=7/041+0/096x-0/086x^2$$

y = the minimum of secondary shoot length

x = number of secondary shoot

These relationship shows that the relationship between the two traits which is examined, have a significant quadratic and by increasing one trait, the other trait may increase or decrease. Tables 5 and 6 show the results of third regression that there is significant correlation between the two evaluated traits as follows:

$$y=2/791+5/025x-1/256x^2-0/076x^3$$

y = the minimum of secondary shoot length

x = number of secondary shoot



Figure 3 Figure showed first second and third grade regression between the two evaluated traits. The results may indicate that regression relations between the first second and third grade in two evaluated traits, is significant at 1%.

**y = the mean of secondary shoot length with x = number of secondary shoot**

Linear regression analysis (Tables 1 and 2) showed the significant correlation between the two evaluated traits as follows:  $y=10/781-0/786x$

**(y = the mean of secondary shoot length, x = number of secondary shoot)**

The above equation shows the increasing number of secondary shoot, the mean of secondary shoot length decreases. The result of quadratic regression showed (Tables 3 and 4) significant correlation between the two evaluated traits as follows:

$$y=8/383+0/3714x-0/115x^2$$

**(y = the mean of secondary shoot length, x = number of secondary shoot)**

Tables 5 and 6 show the result of quadratic regression analysis shows a significant correlation between the evaluated traits at 1% as follows:

$$y=4/126+5/306x-1/286x^2-0/076x^3$$

**(y = the mean of secondary shoot length, x = number of secondary shoot)**

Figure 4 show the first, second and third regressions, among two evaluated traits.

The results indicate that the two evaluated traits have first, second, and third regressions and it was significant at 1% level.

**Table1. Result of linear regression of evaluated traits**

Regression equation				
$y=75.278-2.087x$ y = main shoot length x = number of secondary shoot	$y=11.621-0.571x$ y = the maximum of secondary shoot length x = number of secondary shoot	$y=8.844-0.774x$ y = the minimum of secondary shoot length x = number of secondary shoot	$y=10.781-0.786x$ y = the mean of secondary shoot length x = number of secondary shoot	
Adjusted R <sup>2</sup> =0.157	Adjusted R <sup>2</sup> =0.083	Adjusted R <sup>2</sup> =0.190	Adjusted R <sup>2</sup> =0.185	





		$R^2=0.163$	$R^2=0.089$	$R^2=0.196$	$R^2=0.191$
Sources of variation	df	Mean square	Mean square	Mean square	Mean square
Regression	1	2496.734**	186.781**	343.303**	354.488**
Error	139	92.416	13.711	10.138	10.790

\*\* and \* significant at 1% and 5% respectively

Table2. Results of t-test for linear regression coefficients

Regression equation	y=75.278- 2.087 x y = main shoot length x = number of secondary shoot		y=11.621- 0.571x y = the maximum of secondary shoot length x = number of secondary shoot		y=8.844- 0.774 x y = the minimum of secondary shoot length x = number of secondary shoot		y=10.781- .786x y = the mean of secondary shoot length x = number of secondary shoot	
	Adjusted $R^2=0.157$		Adjusted $R^2=0.083$		Adjusted $R^2=0.190$		Adjusted $R^2=0.185$	
	$R^2=0.163$		$R^2=0.089$		$R^2=0.196$		$R^2=0.191$	
	Coefficients B	T-test	Coefficients B	T-test	Coefficients B	T-test	Coefficients B	T-test
X	-2.087	-5.198**	-0.571	-3.691**	-.774	-5.819**	-.786	-5.732**
Constant	75.278	30.961**	11.621	12.409**	8.844	10.983**	10.781	12.976**

\*\* and \* significant at 1% and 5% respectively

Table3. Regression analysis of second- order evaluated traits



Regression equation		Adjusted R <sup>2</sup> =0.152 R <sup>2</sup> =0.164		Adjusted R <sup>2</sup> =0.144 R <sup>2</sup> =0.156		Adjusted R <sup>2</sup> =0.208 R <sup>2</sup> =0.220		Adjusted R <sup>2</sup> =0.220 R <sup>2</sup> =0.231	
		Mean square		Mean square		Mean square		Mean square	
$y=76.546-2.699x+0.061x^2$ y = main shoot length x = number of secondary shoot		1258.614**		163.375**		192.375**		213.882**	
$y=8.307+1.028x-0.159x^2$ y = the maximum of secondary shoot length x = number of secondary shoot		92.937		12.796		9.911		10.338	
$y=7.041+.096x-.086x^2$ y = the minimum of secondary shoot length x = number of secondary shoot									
$y=8.383+.371x-.115x^2$ y = the mean of secondary shoot length x = number of secondary shoot									
Sources of variation	df								
Regression	2								
Error	138								

\*\* and \* significant at 1% and 5% respectively

Table4. Results of t-test for quadratic regression coefficients

Regression equation	Adjusted R <sup>2</sup> =0.152 R <sup>2</sup> =0.164		Adjusted R <sup>2</sup> =0.144 R <sup>2</sup> =0.156		Adjusted R <sup>2</sup> =0.208 R <sup>2</sup> =0.220		Adjusted R <sup>2</sup> =0.220 R <sup>2</sup> =0.231	
	Coefficients B	T-test	Coefficients B	T-test	Coefficients B	T-test	Coefficients B	T-test
$y=76.546-2.699x+0.061x^2$ y = main shoot length x = number of secondary shoot	-2.699	-1.979*	1.028	2.032**	.096	.216	.371	.815
$y=8.307+1.028x-0.159x^2$ y = the maximum of secondary shoot length x = number of secondary shoot	0.061	.470	-.159	3.307***	-.086	-2.045**	-.115	2.662***
$y=7.041+.096x-.086x^2$ y = the minimum of secondary shoot length x = number of secondary shoot	76.546	21.040***	8.307	6.154***	7.041	5.926***	8.383	6.909***
$y=8.383+.371x-.115x^2$ y = the mean of secondary shoot length x = number of secondary shoot								

\*\*\*, \*\* and \* significant at 1%, 5% and 10% respectively



Table5. Regression analysis of third-order evaluated traits

Regression equation		Adjusted R <sup>2</sup> =0.147 R <sup>2</sup> =0.165	Adjusted R <sup>2</sup> =0.273 R <sup>2</sup> =0.289	Adjusted R <sup>2</sup> =0.37 R <sup>2</sup> =0.387	Adjusted R <sup>2</sup> =0.376 R <sup>2</sup> =0.389
$y=75.460-1.440x-0.238x^2+0.019x^3$ y = main shoot length x = number of secondary shoot					
$y=4.179+5.816x-1.295x^2+0.074x^3$ y = the maximum of secondary shoot length x = number of secondary shoot					
$y=2.791+5.025x-1.256x^2-0.076x^3$ y = the minimum of secondary shoot length x = number of secondary shoot					
$y=4.126+5.306x-1.286x^2-0.076x^3$ y = the mean of secondary shoot length x = number of secondary shoot					
Sources of variation	df	Mean square	Mean square	Mean square	Mean square
Regression	3	845.463**	201.241**	226.099**	240.719**
Error	137	93.476	10.867	7.841	8.264

\*\* and \* significant at 1% and 5% respectively

Table6. Results of t-tests for regression coefficients of the Cubic degree

Regression equation	Adjusted R <sup>2</sup> =0.147 R <sup>2</sup> =0.165		Adjusted R <sup>2</sup> =0.273 R <sup>2</sup> =0.289		Adjusted R <sup>2</sup> =0.374 R <sup>2</sup> =0.387		Adjusted R <sup>2</sup> =0.376 R <sup>2</sup> =0.389	
	Coefficients B	T-test	Coefficients B	T-test	Coefficients B	T-test	Coefficients B	T-test
$y=75.460-1.440x-0.238x^2+0.019x^3$ y = main shoot length x = number of secondary shoot	-1.440	-.465	5.816	5.503**	5.025	5.598**	5.306	5.758**
$y=4.179+5.816x-1.295x^2+0.074x^3$ y = the maximum of secondary shoot length x = number of secondary shoot								
$y=2.791+5.025x-1.256x^2-.076x^3$ y = the minimum of secondary shoot length x = number of secondary shoot								
$y=4.126+5.306x-1.286x^2-.076x^3$ y = the mean of secondary shoot length x = number of secondary shoot								



$X^2$	-.238	-.354	-1.295	-5.646**	-1.256	-6.447**	-1.286	-6.431**
$X^3$	0.019	.453	.074	5.048**	.076	6.119**	.076	5.969**
Constant	75.460	17.282**	4.179	2.807**	2.791	2.207*	4.126	3.178**

\*\* and \* significant at 1% and 5% respectively

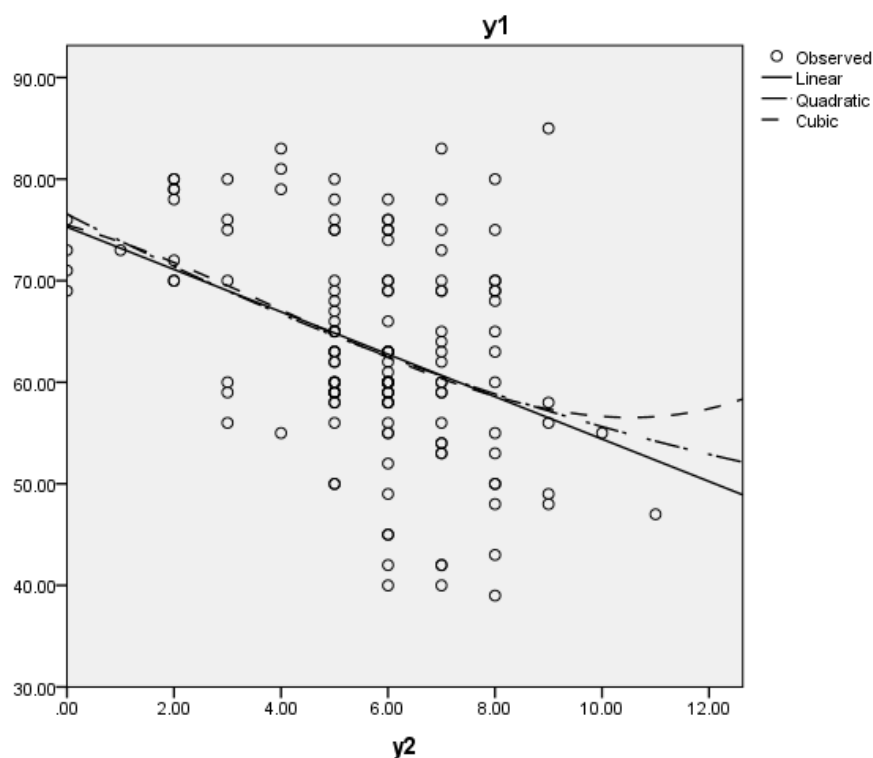
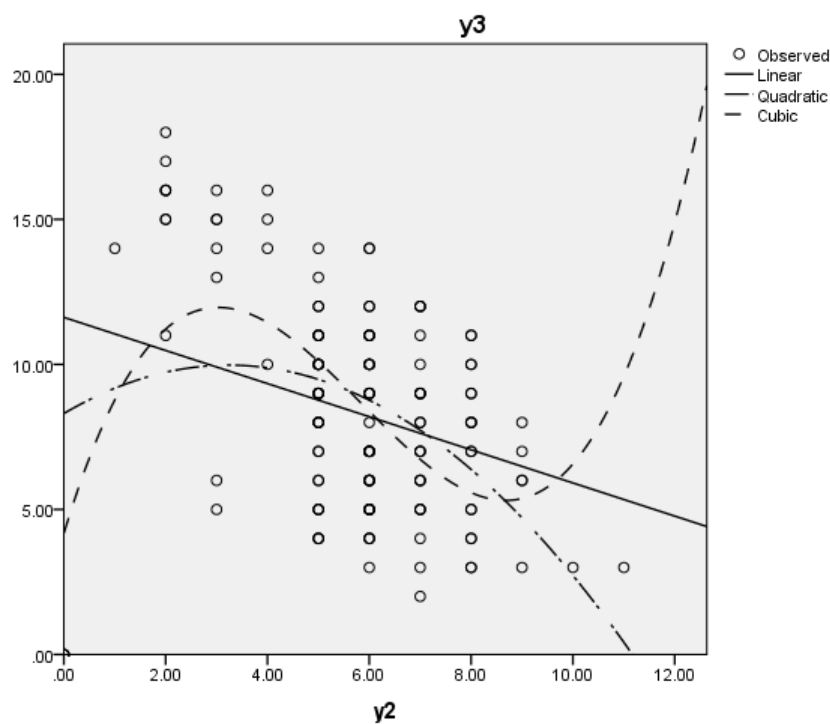


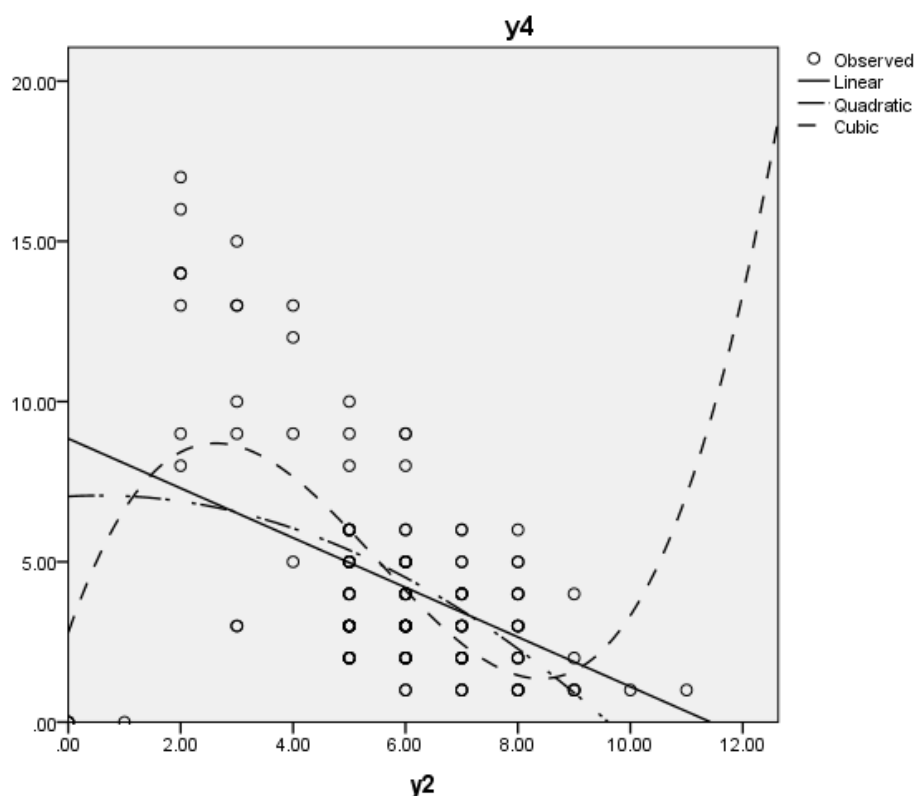
Figure1. Graphs of the relationship between 2 evaluated variables regression

(y = main shoot length with x = number of secondary shoot)

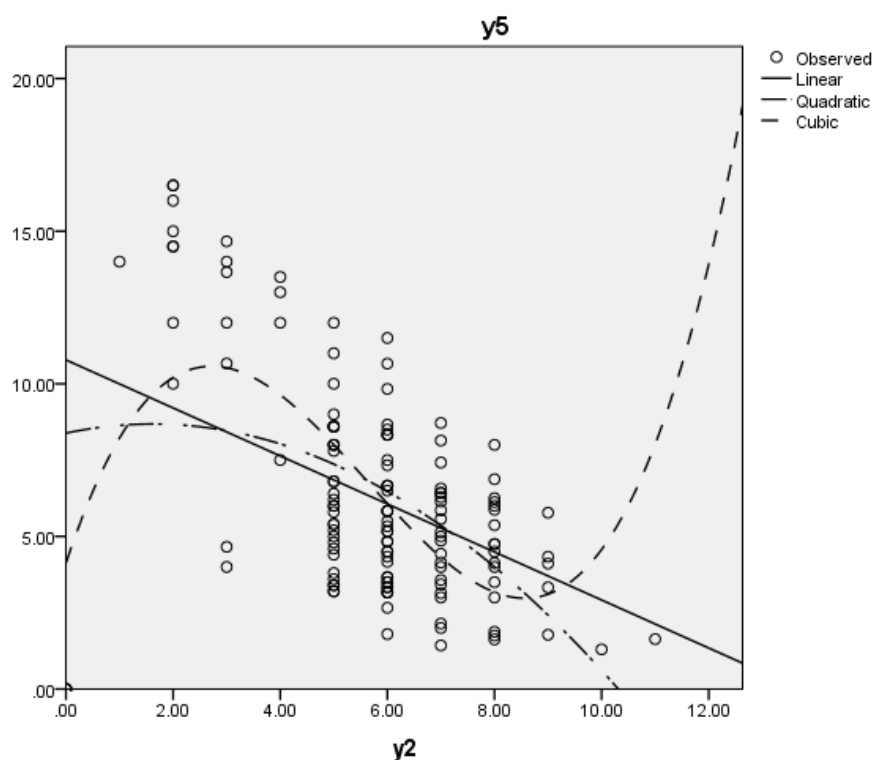




**Figure2. Graphs of the relationship between 2 evaluated variables regression**  
 (y = the maximum of secondary shoot length with x = number of secondary shoot)



**Figure2. Graphs of the relationship between 2 evaluated variables regression**  
 (y = the minimum of secondary shoot length with x = number of secondary shoot)



**Figure4. Graphs of the relationship between 2 evaluated variables regression**  
 (y = the mean of secondary shoot length with x = number of secondary shoot)



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