

ROOTING OF DATE PALM (*PHOENIX DACTYLIFERA* L.) OFFSHOOTS BY ISOPROTHIOLANE (IPT).

Abdullatif. A. Alkhateeb

Agriculture biotechnology department ,College of agriculture and food sciences, King Faisal University, Alhassa, Kingdom of Saudi Arabia

Suliman. A. Alkhateeb

Environment and natural resources department ,College of agriculture and food sciences, King Faisal University, Alhassa, Kingdom of Saudi Arabia

T. Ohmura

Japan Turfgrass II , Tokyo, Japan

R. Okawara

Bio-Environment Engineering Section, Civil Engineering Research Institute, Technology Center, Taisei Corporation , Totsuka-ku, Yokohama, Japan

H. Ali-dinar

Food and Agriculture Organisation (FAO),
Projects and Planning, Saudi Arabia

Abstract

The experiment was conducted at Alhassa Oasis (25° 22' N' latitude; 49°34' E longitude) and altitude is 179 m a.s.l , Kingdom of Saudi Arabia. Treatments included the following Isoprothiolane (IPT) concentrations incorporated at the bottom of offshoot hole: control (without IPT), 25g, 50g, 75g, 100g, 200g and 500g / offshoot hole. The offshoots weight ranged between 25 – 30 kg. The IPT concentrations of 75 and 100 g/ offshoot hole seemed to be quite optimal for improving rooting of date palm offshoots. The best rooting percentage, length of root, root fresh and dry weights were obtained under 75 and 100 g/ offshoot whole IPT concentrations. Offshoots under the control and the lowest and highest IPT treatments reflected poor rooting ability. The chlorophyll content data although looked relatively similar between treatments, a slight edge of 75 and 100 g/ offshoot hole was noticeable. This relative edge might have played a significant role in the photosynthetic ability of offshoots. The efficiency of rooting of offshoots under both concentrations might have resulted from their edged photosynthetic ability.

Keywords: Date plam, ISOPROTHIOLANE

Introduction

Date palm (*Phoenix dactylifera* L.) is the major fruit crops in Kingdom of Saudi Arabia. Its utilization is quite variable ranging from food security to ornamental and landscape purposes. Offshoots are normally used in propagation due to their ability of root regeneration (El-hamady et al. 1992; Zaid & de Wet 2002; Chao & Krueger 2007; Rahnema & Rah khodaei 2013). This ability is highly cultivar dependent with certain high quality ones difficult to root. However, with the increasing demands for date palm offshoots in recent years, farmers tendency to use offshoots of low quality has tremendously increased. Offshoots of certain high quality commercial cultivars have been always difficult to root and their survival ability is low (Al-Mana et al. 1996; Qaddoury & Amssa 2004; Afzal et al. 2011).

Isoprothiolane (diisopropyl-1, 3-dithiolan-2-ylidenemalonate, IPT) is a fungicide (12% hormone, 126 IPT) mainly used for control of rice blast. Recently, IPT has been proven to demonstrate a hormonal action in rice (Ohtsuka & Saka 1988). Improvement of development of seminal root, promotion of root formation in kidney bean (*Phaseolus vulgaris* L.) and azuki bean (*Phaseolus angularis* L.) by using IPT was also reported (Ohtsuka et al. 1990). Okawara et al. 2003 were able to dramatically enhance top and root growth of young date palm plants with application of IPT. Other hormones as well showed their substantial ability to initiate rooting of several plant species (Gasper & Hodinger 1989 ; Ismail & Egaili 1993; Qaddoury & Amssa 2004; Yamada & sawa 2013). Auxins in general, have been intensively used in adventitious root initiation of many hardwood crops (Gupta & Godara 1984; Gasper & Hodinger 1989; Shibli et al. 2000; Bhuiyan et al. 2011). Next to the obligatory presence of endo- or exogenously applied auxin (Haissig 1972), rooting initiation seems to involve the mobilization or redistribution of certain minerals. Several authors revealed that N had great effects on root initiation of cuttings with a low or medium N supply resulting in the highest rooting (Desnos 2008; Peret et al. 2009; Lima 2010). The significant role of N in root growth and development was also noticed and explained by its need for nucleic acid and protein synthesis in addition to carbohydrate metabolism. The high C/N ratio was noticed to always favor rooting of several plant species (Pearse 1943). However, other reports revealed negative correlations between N and root number and weight in cuttings of certain plant species (Kurvits & Kirkby 1980). The negative correlations have led the authors to demonstrate that N composition or source was more important in mineral uptake and carbohydrate and organic acid content of shoots during rooting. It was also found that other rooting co-factors could possibly play a partial role in root

initiation (James & Thumbon 1981; Bhuiyan et al. 2011). The cofactors may partially prevent the oxidation of the naturally occurring rooting hormone IAA by the enzyme IAA-oxidase (Bhuiyan et al. 2011). Ismail & Egaili 1993 have used 1000, 2000 and 3000 ppm concentrations of IBA to root different date palm offshoots of different cultivars and sizes. After one year, he found that 'Barnsee' and 'Ahmed Sayed' cultivars were the best to root while 'Elhora' cultivar did not produce any roots under all IBA concentrations. Normally young date palm offshoots have to withstand severe drought conditions until the root system is sufficiently developed for sustainable water absorption. The use of rooting hormones to enhance root formation, considerably shorten the period during which the offshoots may be subjected to adverse drought conditions. The purpose of this study was to stimulate and improve rooting of date palm cv. 'Khalas' offshoots by using hormonal fungicide Isoprothiolane (12% hormone, 126 IPT). 'Khalas' date palm is a major high quality commercial cultivar in Kingdom of Saudi Arabia.

Materials and Methods

Plant Materials

Experiment was conducted at Alhassa farm Al-shemia area (25° 22' N' latitude; 49°34' E longitude) and altitude is 179 m a.s.l Kingdom of Saudi Arabia. Uniform offshoots weighing between 25 – 30 kg were separated from mother trees and used for the experiment.

Ipt application

IPT, in a granule form (12% effective hormonal dose) at rates of 0 (control/ without IPT), 25, 50, 75, 100, 200 and 500 g hole⁻¹ was applied to bottom of holes and thoroughly mixed with soil before planting of ground offshoots. Proper management practices were given during offshoots growth as recommended by Al-Khateeb *et al.* 2006.

Experimental Parameters

After 12 months from planting, offshoots were dug and separated into top (shoot and leaves) and roots (solid rhizome and adventitious roots). Data were obtained for offshoot ability of rooting as indicated in the following scale:

1. *REPRESENTS BELOW 25% ROOTING OF OFFSHOOT BASAL AREA.*
2. *REPRESENTS BETWEEN 25-50% ROOTING OF OFFSHOOT BASAL AREA.*
3. Represents above 50% rooting of offshoot basal area.

Average length and fresh weight of roots in addition to offshoots survival rate were also obtained. Dry weight of roots was determined after

oven drying the samples at 80 °C to a constant weight. Offshoot leaf chlorophyll content was determined at the termination of the experiment using a Minolta Chlorophyll Meter SPAD-502. potassium K⁺ and calcium Ca²⁺ contents in roots were determined from dry powdered root tissue after extraction in HCl, using an atomic absorption spectrophotometer (905AA, GBC, Australia). Total nitrogen N was determined using an automated semi-macro Kjeldahl apparatus (4301322, Buchi, Switzerland).

Statistical Analysis

Data subjected to statistical analysis as a randomized complete block design according to Gomez and Gomez (1984). All statistical analysis was performed using the facility of computer and SAS software package (SAS, 2001). Means were separated by their standard errors and the corresponding degrees of freedom.

Results

Rooting percentage and root length of offshoots are shown in Table 1. Data on rooting ability of offshoots have reflected considerable variation between IPT treatments. Substantial rooting was observed on offshoots treated with 75 and 100 g IPT (Fig. 1). Rooting on untreated offshoots (control) and on 25 and 500 g IPT was significantly ($P < 0.05$) reduced with possibility of toxicity at 500 g IPT (Table 1 and Fig. 1). Regression between index of rooting and IPT gave nonlinear correlations with very good coefficient of determination $r^2 = 0.89$ (Fig. 2). Length of offshoot roots followed almost the same rooting percentage pattern. Both 75 and 100 g IPT applications seemed to be quite optimal and promotive for maximum and efficient rooting of ground offshoots. An offshoot rooting under both applications was approximately 8 to 16 folds better than rooting ability of offshoots under control, 25 and 500 g applications. Root fresh and dry weights were relatively higher in offshoots treated with 75 and 100 g IPT (Table 2). Both root fresh and dry weights were drastically reduced on untreated (control) and 25 and 500 g treated offshoots. This is not surprising, since the rooting ability of offshoots under those treatments was shown to be low (Table 1). Leaf chlorophyll content and root mineral content (N, K⁺, Ca²⁺) were significantly ($P < 0.05$) higher under offshoots treated with 75 and 100 g IPT (Table 3 and Table 4).

Discussion

This study has quantified in details the rooting behavior of ground offshoots of date palm cv. 'Khalas' in response to IPT. Rooting ability of offshoots have reflected considerable variation between IPT treatments with substantial rooting observed on offshoots treated with 75 and 100 g IPT. The

relationship between IPT and index of rooting indicate good coefficient of determination r^2 0.89 (Fig 2) . This relationship showed that 100 g IPT/ offshoot hole as the ideal concentration for optimum rooting in date palm. The efficiency of rooting under all treatments could probably be explained by the data on offshoot leaf chlorophyll content and root mineral content. It is quite reasonable to postulate that leaves with higher chlorophyll content had much better photosynthetic ability to support root growth and development. Furthermore, the provision of optimum photosynthates to offshoots might have stimulated the roots of the same offshoots to accumulate higher N, K^+ and Ca^{2+} contents which is true in the present study. Regression between N and IPT gave nonlinear invers correlations with good coefficient of determination r^2 0.75 showing lower N content with increasing IPT concentration (Fig. 3). This could be due to the fact that IPT had no significant effect on root N absorption at high concentrations. On the other hand regression between K^+ and Ca^{+2} and IPT gave nonlinear correlations but with relatively very good coefficient of determination r^2 0.83 and 0.86, respectively (Fig. 3). These relationships indicate that 100 g IPT/ offshoot hole as the ideal concentration for K^+ and Ca^{+2} absorption. This indicates that IPT dose not only promote root formation but it may also promote root functioning effectively. Higher K^+ concentration under 75 and 100 g IPT may effectively enhance gas exchange capacity by controlling stomatal conductance (Tais & Zeiger 2010). On the other hand higher Ca^{+2} concentration under 75 and 100 g IPT may effectively contribute well in membrane permeability (Tais & Zeiger 2010). The stimulation and development of roots in plants have been related to hormonal (Qaddoury & Amssa 2004; Afzal *et al.* 2011, Yamada & sawa, 2013), nutritional (Desnos 2008; Lima 2010) and cofactors (Fadl *et al.* 1979; Peret *et al.* 2009) mechanisms. Auxins, in particular, have been reported to exert the primary control over root formation (Gupta & Godara 1984). However, AlKhateeb 2008 have demonstrated that meristematic differentiation either to root primordium or to callus and leaf primordium is dependent on the proportion of auxin to cytokinin or to other substances that may stimulate cell division. IPT is a fungicide that has recently been known for its growth- regulatory action in rice (Ohtsuka & Saka 1988). Its application to soil has also been shown to promote initial root growth in date palm offshoots (Okawara, *et al.* 2003). In the current study, optimal levels of IPT have distinctly promoted rooting of ground offshoots. Since IPT is not known as an auxin related hormone, it can reasonably assume that its promotive action to root formation possibly reflects interactive indirect effects. Those effects might have been stimulated and favored by a positive proportion of auxins to other hormones, particularly cytokinin, or/ and by stimulating an active translocation of cofactors with photosynthates from leaves to root zone to

promote rooting (AlKhateeb 2007). In conclusion, the IPT concentrations of 75 and 100 g/ offshoot hole seem to be quite optimal for improving rooting of date palm offshoots. The chlorophyll content data supported the edge effect of 75 and 100 g IPT/ offshoot hole. This edge effect might have played a significant role in the photosynthetic ability of offshoots under those treatments. The photosynthetic ability of the offshoots under the mentioned treatments might have resulted in a relatively balanced nutrient content within the plants to support the photosynthetic ability and hence improve rooting of offshoots.

Acknowledgement

This work was financed with grant from Deanship of research at King Faisal University and *Japan Turf grass*.

References:

- Afzal M., Khan M.A., Pervez M.A and Ahmed R. (2011). Root induction in the aerial offshoots of date palm (*Phoenix dactylifera* L.) cultivar hillawi. *Pakistan Journal of Agricultural Science* 48, 11-17.
- Alkhateeb A.A. (2007). The effect of different carbohydrate sources and concentrations on in vitro bud and shoot formation of strawberry (*Fragaria x ananassa*). *Journal of Agricultural Science, Mansoura University* 32,591-599.
- Alkhateeb A.A. (2008). Regulation of in vitro bud formation of date palm (*Phoenix dactylifera* L.) cv. Khanezi by different carbon sources. *Bioresource Technology* 99,6550-6555.
- Alkhateeb A.A., Aljabr A.M.S., Aljabr A.M.H. (2006). Date palm in Kingdom of Saudi Arabia. *The National Date Palm Research Center, Ministry of Agriculture, Kingdom of Saudi Arabia*, pp. 136.
- Al-mana F. A., El-hamady M. A., Bach M. A. and Abdelrahman A. O. (1996). Improving root development on ground and aerial date palm offshoots. *Research Bulletin No: 60, King Saud University, College of Agriculture, Kingdom of Saudi Arabia*, pp. 19.
- Bhuiyan M. K. R., Hossain M. J., Rahman M. S., Rahman S. M. L. and Sattar M. A. (2011). Root initiation in Mukhikachu (*Colocasia esculenta*) as influenced by IAA and NAA. *Bangladesh Journal of Agriculture Research* 36, 487-494.
- Chao C. T. and Krueger R. R. (2007). The date palm (*Phoenix dactylifera* L.): Overview of biology, uses and cultivation. *Horticulture Science* 42,5.
- Desnos T. (2008) Root branching responses to phosphate and nitrate. *Current Opinion in Plant Biology* 11, 82–87.
- El-hamady M., Al-mana F. A. and Bacha. A. (1992). Greenhouse rooting of date palm offshoots using an inverted mist system. *Annals of Agriculture Science Ain Shams University* 37, 523-529.

- Fadl M. S., El-deem A. S. and El-mahdy M. A. (1979). Physiological and chemical factors controlling adventitious root initiation in carob (*Ceratonia siliqua*) stem cuttings. *Egyptian Journal of Horticulture* 6, 55-68.
- Gaspar T., and Hodinger M. (1989). Auxin metabolism during rooting. In adventitious root formation in cuttings. T.A. DAVIS, B.E. HAISSING and N. SANKHLA, eds. Portland, Oreg., Dioscorides Press.
- Gomez K. A. and Gomez A. A. (1984). *Statistic procedures for agricultural research*. 2nd ed., 680 pp., John Wiley and Sons Inc., New York.
- Gupta O.P. and Godara N.R. (1984). Rooting in aerial suckers of date palm. *Haryana Agriculture University Journal Research* 14,82- 84.
- Haissig B. E. (1972). Meristematic activity during adventitious root primordium development. Influences of endogenous auxin and applied gibberellic acid. *Plant Physiology* 49, 886-892.
- Ismail M. M., Alegali F. R. (1993). Effects of IBA growth hormone on root formation of young date palm offshoots. *Proceedings of the third symposium on date palm*. King Faisal University, Alhassa, Kingdom of Saudi Arabia.
- James, D. J. and Thurbon I. J. (1981). Shoot and root initiation in vitro in the apple rootstocks M 9 and the promotive effects of phloroglucinal. *Journal of Horticulture Science* 56,15- 20.
- Kurvits A. and Kirkby E. A. (1980). The growth and mineral composition of sunflower plants, *Helianthus annuus*, utilizing nitrate or ammonium-N when grown in a continuous flowing culture system. *Acta Horticulture* 98, 139-148.
- Limaa J. E., Kojimaa, S., Takahashib H. and Von Wirena N. (2010). Ammonium triggers lateral root branching in arabidopsis in an ammonium transporter 1;3- dependent manner. *The Plant Cell* 22, 3621-3633.
- Ohtsuka T., Hikawa M. and Saka H. (1990). Growth regulation action of isoprthiolane in plants. V. Interaction between RNA, protein synthesis inhibitors and isoprthiolane on adventitious root formation in stem segments of plant crops. *Japanese Journal of Crop Science* 59, 566-571.
- Ohtsuka T. and Saka H. (1988). Undestructive measurement of acid phosphates activity in the root of rice seedlings. *Japanese Journal of Crop Science* 57, 722-727.
- Okawara R., Macawi R. M., Alkhateeb S. and Ohmura T. (2003). Improvement of the initial growth of young date palm (*Phoenix dactylifera* L.) plants by the application of Isoprothiolane to soil. *Soil Science and Plant Nutrition* 49, 281-283.
- Pearse H. L. (1943). The effect of nutrition and phytohormones on the rooting of vine cuttings. *Annals of Botany*7, 123-132.
- Peret B., Larrieu A. and Bennett M.J. (2009). Lateral root emergence: A difficult birth. *Journal of experimental Botany* 60, 3637–3643.

Qaddoury A. and Amssa M. (2004). Effect of exogenous indole butyric acid on root formation activities and phenolic contents in date palm offshoots. Botanical Bulletin- Academia Sinica 45,127-131.

Rahmana A. and Rahkhodaei E. (2013). Offshoots weight and disinfection on Medjool Date palm survival rate. International Journal of Agriculture and Crop Sciences 5, 1784-1788.

SAS INSTITUTE (2001). SAS for windows, SAS Users Guide: statistics Version 8.0 e. Cary, North Carolina.

Shibli R. A. (2000). Cryopreservation of Black Iris (Irisnigricans) Somatic Embryos by Encapsulationdehydration. Cryo-Letters 21, 39-46.

Taiz L. , Zeiger E. (2010). Plant Physiology. Sinauer Associations, USA.

Yamada M. and Sawa S. (2013). The roles of peptide hormones during plant root development. Current Opinion Plant Biology 16,56-61.

Zaid A. and De Wet P.F. (2002). Date palm propagation. P 73-105. In: A.Zaid (ed). Date palm cultivation. Food and agriculture Organization Plant Production and Protection Paper No.156. Rome Italy.

Table(1): Effect of IPT on rooting percentage and root length of date palm ‘Khalas’

IPT concentration (g/ offshoot hole)	Index of rooting %	Average length of offshoot root (mm)
Control (0)	0.25± .028	267±31
25	0.35± .033	461±39
50	1.42± .19	463±55
75	2.24± .26	561±62
100	2.89± .25	633±71
200	1.67± .21	398±45
500	0.18± .016	232±33

Means ± S.E, D.F=6

Table (2): Effect of IPT on fresh and dry weight of date palm ‘Khalas’ cv. offshoots

IPT concentration (g/ offshoot hole)	Fresh weight (g)	Dry weight (g)
Control (0)	193.6± 25	36.8± 4.0
25	193.5± 24	39.7± 4.1
50	391.6± 45	48.2± 5.1
75	356.4± 31	44.8± 3.9
100	460.8±45	54.9± 5.8
200	237.4± 26	39.5± 4.4
500	145.9±19	34.5± 3.2

Means ± S.E, D.F=6

Table (3): Effect of IPT on chlorophyll content and offshoot survival of date palm ‘Khalas’ cv. offshoots

IPT concentration (g/ offshoot hole)	Chlorophyll content (mg/ 100cm) (SPAD value)	Survival percentage of offshoots plants/100plants
Control (0)	18.2±1.9	60±5.9
25	18.3±2.2	60±6.5
50	19.2±2.5	100±0
75	21.3±1.8	100±0
100	21.1±2.8	100±0
200	18.6±1.9	40±8
500	17.5±1.6	20±3

Means ± S.E, D.F=6

Table (4): Effect of IPT on root N, K⁺ and Ca²⁺ content of date palm ‘Khalas’ cv. offshoots

IPT concentration (g/ offshoot hole)	N mg/g	K ⁺ mg/g	Ca ²⁺ mg/g
Control (0)	13.7±1.21	4.6±.51	15.1±1.9
25	13.5± 1.51	4.8±.49	15.1±1.6
50	14.3±1.81	5.6±.59	16.3±1.5
75	15.9±1.98	7.6±.81	17.4±1.82
100	15.8±1.56	7.4±.79	16.9±1.9
200	14.1±1.62	6.1±.73	14.8±1.6
500	13.1±1.38	4.5±.51	14.0±1.8

Means ± S.E, D.F=6








	
Control (0 IPT)	25 g (IPT) / offshoot hole
	
50 g (IPT) / offshoot hole	75 g (IPT) / offshoot hole
	
100 g (IPT) / offshoot hole	200 g (IPT) / offshoot hole
	
500 g (IPT) / offshoot hole	

Figure1 : Effect of IPT on rooting of date palm cv. ‘Khalas’ offshoots.

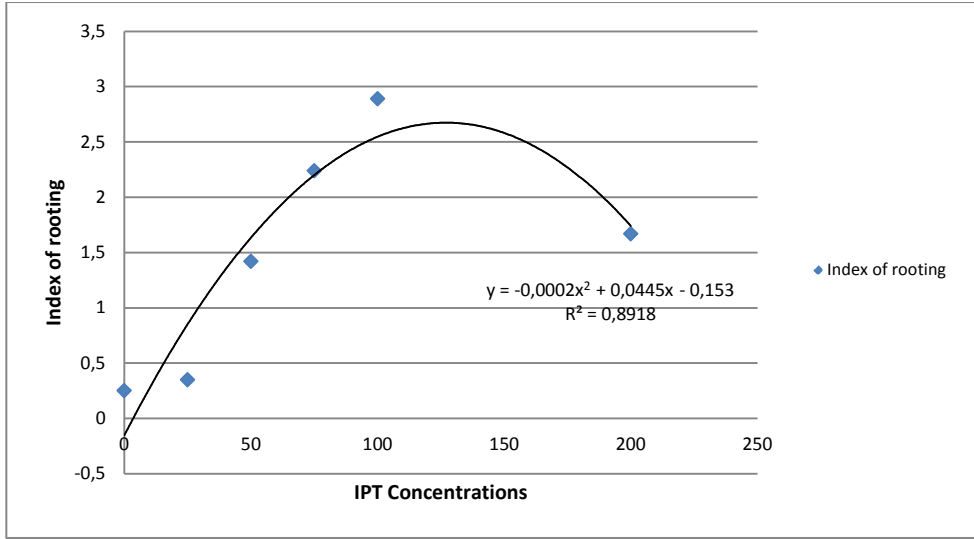


Figure 2: Relationship between Index of rooting and IPT (g/ offshoot hole).

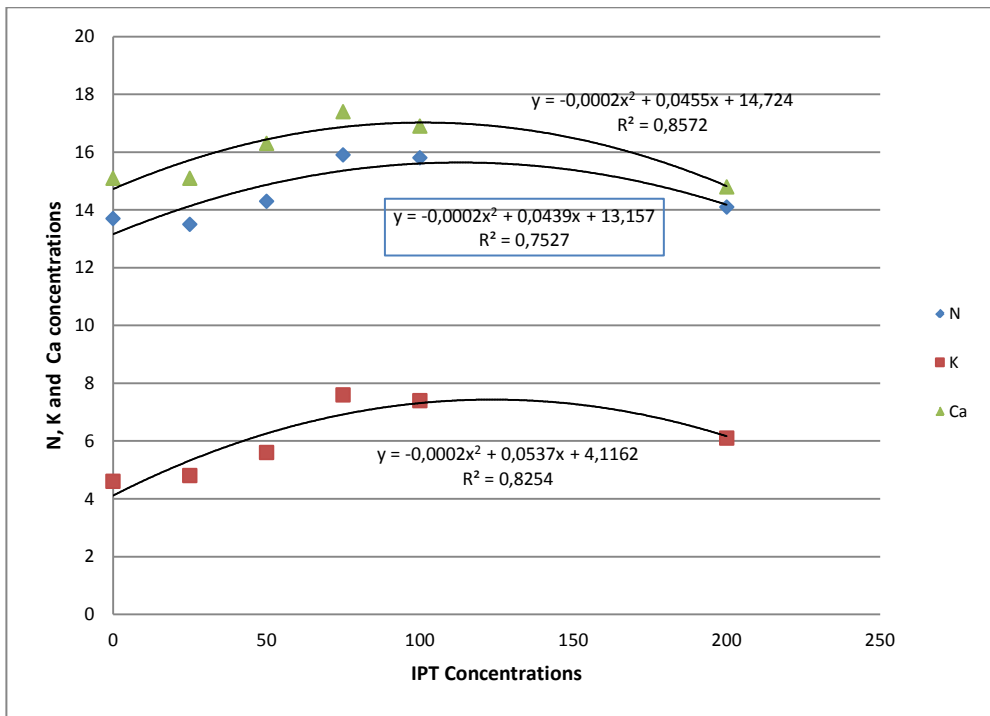


Figure 3: Relationship between root N , K and Ca concentration (mg g⁻¹) and IPT (g/ offshoot hole).