

ISSN 1916-9752 (Print)
ISSN 1916-9760 (Online)

JOURNAL OF AGRICULTURAL SCIENCE

**Vol. 1, No. 1
June 2009**



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Utility of Dried Distillers Grain as a Fertilizer Source for Corn

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Abstract

Increased ethanol production may result in excessive dried distillers grains (DDGs) that could be utilized as a fertilizer source for corn (*Zea mays* L.). Research was conducted to evaluate the effects of 1) DDG rates on weed suppression, changes in soil properties, and differences in grain yield and quality and 2) DDGs, polymer-coated urea (PCU), and anhydrous ammonia (AA) fertilizer sources on grain yield and quality. DDGs had a total N-P-K composition of 38.2-6.9-11.5 g kg⁻¹, respectively. There was no corn injury, common cocklebur or jimsonweed control with DDG rates up to 3600 kg ha⁻¹. Corn grain yield increased 1.41 and 1.56 kg ha⁻¹ for every kg ha⁻¹ of DDGs applied in medium and high yield environments, respectively. Grain yield was ranked non-treated control < DDGs ≤ AA = PCU when fertilizers were applied at N equivalent rate of 140 kg ha⁻¹ in medium and high yield environments.

Keywords: Soil amendment, Nitrogen, Organic, Weed control, Anhydrous ammonia, Polymer-coated urea

1. Introduction

Ethanol production in the U.S. has created new jobs in rural communities, increased the amount of land area cropped to corn (*Zea mays* L.), stimulated higher local grain prices, and raised the amount of corn used by the dry-milling industry in the Midwest (Parcell & Westhoff, 2006). Dried distillers grains (DDGs) are a co-product of converting corn into ethanol. Continued growth in ethanol production may create a surplus of DDGs (Erickson, Klopfenstein, Adams, & Rasby, 2005; Rausch & Belyea, 2006) and alternative uses for this material may be needed. DDGs are commonly fed to livestock as a protein source in the wet or dry form as a yellow meal (Erickson et al., 2005; Harris, Cupp, Roberts, & Funston, 2008; Martin, Cupp, Rasby, Hall, & Funston, 2007; Martinez Amezcuela, Parsons, & Noll, 2004). DDGs are an excellent source of phosphorus and other nutrients in the diets of livestock (Belyea, Rausch, & Tumbleson, 2004; Erickson et al., 2005; Harris et al., 2008; Martinez Amezcuela et al., 2004; Rausch & Belyea, 2006).

A portion of plant nutrient elements contained in land-applied DDGs should become available to the plant over the growing season through the decomposition process similar to manure and other compost materials (Gale et al., 2006). Several studies have evaluated the effect of compost, livestock manure (Eghball, 2002; Gale et al., 2006; Lithourgidis, Matsi, Barbayiannis, & Dordas, 2007; Singer et al., 2004), oily feed wastes (Rashid & Voroney, 2004), paper mill solids (Aitken, Evans, & Lewis, 1998; Curnoe, Irving, Dow, Velema, & Unc, 2006; N'Dayegamiye 2006), organic fertilizers, and biofertilizers (Jilani et al., 2007) on crop production. Manure, compost, and organic fertilizer research has reported corn grain yields similar (Lithourgidis et al., 2007) or lower (Jilani, et al., 2007) than comparable

applications of synthetic N-P-K fertilizers. Among the additional benefits of using organic soil amendments, such as manure, are improvements in soil physical biological and chemical properties, including increased soil water-holding capacity, aggregation, cation exchange capacity, decreased soil erosion, and addition of micronutrients which may be deficient when only synthetic N-P-K fertilizers are applied (Edwards, Burt, Raper, & Walker, 1995). Potential problems with land application of organic soil amendments include the possible presence of toxic substances (e.g., excessive micronutrients or other trace elements, industrial byproducts, and agrochemicals) and biological pathogens, odor, and salinity (Stevenson, 1985).

Increased use of corn for ethanol production may also lead to use of corn contaminated with aflatoxin (*Aspergillus flavus*). Aflatoxins are not affected by fermentation, but are concentrated in the DDGs. This may be particularly important to ethanol plants located near drought-prone, claypan soils (Blanco-Canqui, Gantzer, Anderson, Alberts, & Ghidey, 2002). Disposal of aflatoxin-contaminated DDGs as fertilizer grade material may provide ethanol facilities with an alternative use for a product with no animal feed value. No research has evaluated the impact of DDGs on crop and weed response as a fertilizer source or a possible preemergence herbicide in crop production systems. Therefore, the objectives of this research were to: 1) evaluate the effect of DDG rates on weed suppression, changes in soil properties, and relative grain yield and quality; and 2) evaluate grain yield and quality response to DDGs with slow release urea and anhydrous ammonia fertilizer sources at an equivalent N source application rate.

2. Materials and methods

2.1 General methods

Field research was conducted at the University of Missouri Greenley Research Center near Novelty (40° 01' N, 92° 11' W) on a Putnam silt loam (fine, smectitic, mesic Vertic Albaqualfs) from 2003 to 2007 in the DDG rate experiment and from 2004 to 2007 in the N fertilizer source experiment. The experiments were arranged in randomized complete block designs with four replications. Locally produced DDGs (POET Biorefining, Macon, MO) were collected from three of the four replications each year prior to application and analyzed for chemical and mineral properties using recommended manure analysis methods for total N, ammonium-N, P, K, Ca, Zn, Fe, Mn, Cu, pH, and electrical conductivity (Peters et al., 2003). A 1:2 solid:water solution was used for determining pH, electrical conductivity, and ammonium-N. The average analyses of DDGs are presented in Table 1. DDGs were broadcast-applied using a hand spreader following planting. The experiments were conducted on previously untreated areas of the same field following soybean [*Glycine max* (L.) Merr.] each year. Grain yield was determined using a Massey Ferguson 10 small plot combine (Kincaid Equipment Manufacturing, Haven, KS) to harvest the center two rows. Grain moisture was adjusted to 150 g kg⁻¹ prior to analysis. Grain samples were collected and ten subsamples analyzed for oil, protein, starch, and extractable starch with a Foss 1241 (Eden Prairie, MN) near infrared spectrometer using previously established calibrations (Paulsen, Pordesimo, Singh, Mbuvi, & Ye, 2003; Paulsen & Singh, 2004; Singh, Paulsen, Tian, & Yao, 2005).

2.2 DDG rates

This site had been in an organic corn-soybean production system for six years prior to the initiation of this research. Initial soil test characteristics are presented in Table 2. Plots were 3 by 15.2 m. The soil was fall or spring moldboard plowed, spring field cultivated, and cultipacked smooth prior to planting. An 'NC+ 112E1' organic corn hybrid was planted at 59 280 to 64 220 seeds ha⁻¹ on 27 May 2003, 6 May 2004, 10 May 2005, 24 April 2006, and 22 May 2007. DDGs were broadcast-applied prior to crop and weed emergence at 0, 1200, 2400, and 3600 kg ha⁻¹ with an average of 102.0 ± 1.39 g kg⁻¹ moisture. The 1200 kg ha⁻¹ rate was equivalent to an N-P-K application rate of 46-8-14 kg ha⁻¹ based on total nutrient content (Table 1).

All plots were rotary hoed twice and cultivated up to three times following planting for weed control. In-row weed control with DDGs was evaluated approximately 8 weeks after planting. Weed control was based on the effects of DDGs on a combined visual score (0 = no visual injury to 100 = complete plant death) of weed stunting and reduction in population density. The primary weed species present were common cocklebur (*Xanthium strumarium* L.) and jimsonweed (*Datura stramonium* L.). Soil was sampled using a composite of 20 soil cores in the fall or early the following spring using a stainless steel push probe to a depth of 15 cm to evaluate the effects of DDGs on selected soil properties.

2.3 DDGs and conventional N sources

Polymer-coated urea (ESN, Agrium, Calgary, AB) and DDGs were broadcast-applied while anhydrous ammonia was knife-injected below the soil surface prior to planting at an equivalent of 140 kg N ha⁻¹ in plots 3 by 12.2 m. The DDG rate calculation was based on total N (Table 1). Treatments were applied to non-treated portions of the field following soybean each year. 'RX 752RR/Bt' was no-till planted at 74 100 seeds ha⁻¹ on 9 April 2004, 3 May 2005, 28 April 2006, and 23 April 2007. Crop injury due to the DDG treatment was evaluated 7, 14, and 21 d after application. Visual injury was based on combined visual score (0 = no visual crop injury and 100 = complete plant death) of leaf necrosis,

chlorosis, and stunting. Plots were maintained weed-free using a combination of a preemergence application of chloroacetamide and atrazine herbicides, postemergence herbicides, and manual weed removal as needed.

2.4 Statistical analysis

Data were subjected to ANOVA using the general linear model procedure (PROC GLM) (SAS Institute, 2007). Percent data for weed control ratings were transformed to the arcsine prior to the analysis. Data were combined over years with low (2003 and 2005), medium (2007) and high (2004 and 2006) yields since no interactions within these environments were observed. Means were separated using Fisher's Protected LSD at $P = 0.05$. Linear regression analysis was performed using best-fit analysis determined with SigmaPlot (Vers. 8.02, SPSS Inc., Chicago, IL).

3. Results and discussion

3.1 DDG rates

This research was conducted on drought prone, claypan soils that are primarily a result of low precipitation during the summer months. Precipitation from 2003 to 2007 was presented in Figure 1. Years were grouped into high, medium, and low yield environments. Data were combined within these yield environments since no interactions existed. Initial soil test P was 92 to 149 kg ha⁻¹, and exchangeable K was 515 to 721 kg ha⁻¹, Ca was 4449 to 5994 kg ha⁻¹, and Mg was 527 to 610 kg ha⁻¹ from 2003 to 2007 (Table 2). All these soil test values were in the medium to very high range for Missouri soil test interpretations (Buchholz, 1992). Therefore, crop response from a balanced N-P-K source was not expected and not included in this experiment. Initial soil organic matter levels ranged from 29 to 32 g kg⁻¹ and pH (0.01 M CaCl₂) from 6.5 to 6.8 from 2003 to 2007 (Table 2). DDGs had an average pH of 4.15 ± 0.15 which may lower soil pH when applied at high rates (Table 1). No significant effect of DDGs on soil pH was observed in low, medium and high yielding years (Table 3). When combined over all five years, soil pH was lower when DDGs were applied at 3600 kg ha⁻¹ compared to the non-treated control ($P \leq 0.08$). The effect of manure and organic fertilizer sources on soil pH depended on the source (Curnoe et al., 2006). Long-term manure applications have increased pH (Eghball, 2002) in some instances and lowered soil pH (Meng, Ding, & Cai, 2005) in others.

Soil tests after harvest also indicated no difference in soil organic matter, soil test P, and exchangeable K, Ca, or Mg concentrations among DDG rates for low, medium, and high yield environments (Table 3). Beef cattle and swine manure applications to corn increased organic matter, P, and K concentration (Eghball, 2002; Singer et al., 2004) while dairy cattle manure had no effect on soil organic C, P, and K concentrations (Lithourgidis et al., 2007). Paper mill waste applications caused a temporary increase in soil organic matter (Curnoe et al. 2006). The N/P ratio for DDGs was 5.5 (Table 1) which is close to the N/P corn grain uptake ratio of 5.9 reported by Gilbertson et al. (1979); therefore, no change in soil P concentration should occur using DDGs as a fertilizer source.

The organic production system used for this experiment selected for large-seeded broadleaf weeds over time including common cocklebur (40 plants m⁻²) and jimsonweed (50 plants m⁻²). There was no effect of DDGs on either weed species during the five years of research (data not presented). Other research has indicated that corn gluten meal at 324 g m⁻² suppressed small-seeded broadleaf and grass weeds when applied preemergence or preplant incorporated, but little effect on large seeded broadleaf weed suppression such as velvetleaf (*Abutilon theophrasti* Medik.) was reported (Bingaman & Christians, 1995; McDade & Christians, 2000). For organic production, application of a fertilizer source such as DDGs that could supply nutrients, act as a soil conditioner, and suppress weed growth would be a valuable input. However, DDGs were not effective in weed suppression in this study.

There was no effect of DDG rate on harvested corn population (data not presented) which was similar to research evaluating low rates of incorporated corn gluten meal on sweet corn survival (McDade & Christians, 2000). Grain oil concentration was reduced 2.5 g kg⁻¹ with DDGs at 3600 kg ha⁻¹ in a low yield environment, but DDG rate did not affect oil concentration in medium and high yield environments (Table 3). Protein concentration increased 7.5 to 12.0 g kg⁻¹ as DDG rate increased while starch decreased 6.7 and 11.2 g kg⁻¹ in high and medium yield years, respectively. Extractable starch decreased 11.7 to 20.8 g kg⁻¹ in low to high yield environments as the DDG rate increased from 0 to 3600 kg ha⁻¹. This was similar to site-specific evaluations of preplant and sidedress N rates on corn grain composition (Singh et al., 2005).

Since rainfall was limiting in 2003 and 2005 (Figure 1), no grain yield response to DDG rates was observed (Figure 2). Corn grain yield increased 2500 and 2300 kg ha⁻¹ with DDGs at 3600 kg ha⁻¹ in medium (2007) and high (2004 and 2006) yield environments, respectively (Figure 2). Grain yield increased 1.41 and 1.56 kg ha⁻¹ for every kg ha⁻¹ of DDGs applied in medium and high yield environments, respectively.

3.2 DDGs and conventional N sources

Data were combined over low, medium, and high yield environments similar to the rate experiments since no interactions existed within these environmental classifications. DDGs caused no visual injury to corn plants up to 21 d after emergence, and no differences in harvested population were observed from 2004 to 2007 (data not presented). In

2003 and 2005, rainfall was limited (Figure 1) and there was no significant yield response to the DDG treatment compared to the control (Table 4), which was similar to the rate experiment. However, corn grain yield increased 3080 and 3580 kg ha⁻¹ with N supplied by PCU and anhydrous ammonia treatments, respectively. DDGs increased grain yield over the non-treated control 2820 to 4390 kg ha⁻¹ in medium and high yield environments. The prediction equations from the rate experiment (Figure 2) indicated an expected yield of 7420 and 11340 kg ha⁻¹ which was similar to the observed 8600 and 11300 kg ha⁻¹ yields for medium and high yield environments, respectively, in this experiment. Grain yield with DDGs was similar to anhydrous ammonia in 2007. The use of DDGs as a fertilizer source for corn was more consistent in medium and high yield environments, but grain yields indicated that the availability of nutrients from DDGs was less than a slow-release PCU fertilizer source or anhydrous ammonia in a high yield environment. Grain oil was not affected by the N sources evaluated in this experiment (Table 4). Protein concentration was maximized as yield increased while starch and extractable starch concentrations decreased. Incorporation of DDGs with tillage or changes in DDG particle size (Rausch et al., 2005) may increase nutrient availability to the plant, but changes in management and application rate may also increase toxicity to corn (McDade & Christians, 2000). Combining synthetic N fertilizer with soil organic amendments may improve crop response, especially with organic materials with high C:N ratios, which may result in net N immobilization (Bellamy, Chong, & Cline, 1995). Additional strategies to overcome potential N immobilization from high C:N organic soil amendments include mixture with higher N-containing organic materials (e.g. poultry litter), composting prior to application, and delaying planting after application of the organic soil amendment (Motavalli & Diambra, 1997).

4. Conclusions

The results of these field studies indicate that DDGs may be utilized as a fertilizer source for corn production. However, agronomic performance was dependant on rainfall and growing conditions during the year. Grain yield increased 1.41 and 1.56 kg ha⁻¹ for every kg ha⁻¹ of DDGs that were applied in medium and high yield environments, respectively. Corn raised in low yield environments did not respond to increasing DDG rates while PCU or anhydrous ammonia increased grain yields in these environments when compared to DDGs. Application rates of DDGs may need to be higher to attain grain yields equivalent to other N sources when surface applied. In this research, DDGs did not affect large seeded broadleaf weed or corn development, SOM, P, K, Ca, or Mg concentration. There was a decrease in soil pH when DDGs were applied at 3600 kg ha⁻¹. Additional research is needed to identify consistent systems that provide responses to DDGs as a fertilizer source in low and medium yield environments if the cost of DDGs is low enough to be used as a fertilizer source for corn. This may require management changes, such as supplemental synthetic N fertilizer applications at reduced rates (N'Dayegamiye, 2006; Rashid & Voroney, 2005;) or incorporation (McDade & Christians, 2000) to achieve yields similar to conventional N fertilizer sources.

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Table 1. Average chemical and physical properties of dried distillers grains (DDGs) applied in the rate and N source experiments from 2003 to 2007.

Properties	Total content mean \pm standard deviation
Moisture (g kg ⁻¹)	102.0 \pm 1.4
Nitrogen (g kg ⁻¹)	38.2 \pm 0.10
Phosphorus (g kg ⁻¹)	6.9 \pm 0.02
Potassium (g kg ⁻¹)	11.5 \pm 0.03
Zn (mg kg ⁻¹)	78.30 \pm 7.02
Fe (mg kg ⁻¹)	86.99 \pm 11.82
Mn (mg kg ⁻¹)	18.85 \pm 2.87
Cu (mg kg ⁻¹)	6.47 \pm 1.17
NH ₄ (mg kg ⁻¹)	334.12 \pm 60.89
pH	4.15 \pm 0.15
Electrical conductivity (S m ⁻¹)	0.66 \pm 0.38

Table 2. Selected initial soil properties before dried distillers grains (DDGs) treatments were applied from 2003 to 2007 in the rate experiment.[†]

Year	Soil organic matter - g kg ⁻¹ -	pH		Exchangeable (1 M NH ₄ AOc)			
		(0.01 M CaCl ₂)	Bray 1 P	K	Ca	Mg	
2003	32 ± 1	6.8 ± 0.1	132 ± 2	693 ± 30	5,994 ± 423	527 ± 30	
2004	31 ± 2	6.6 ± 0.2	126 ± 8	721 ± 58	5,442 ± 466	610 ± 54	
2005	31 ± 2	6.7 ± 0.2	149 ± 2	676 ± 76	4,600 ± 496	545 ± 68	
2006	29 ± 3	6.5 ± 0.2	105 ± 10	575 ± 89	4,449 ± 516	543 ± 108	
2007	29 ± 2	6.5 ± 0.2	92 ± 6	515 ± 80	5,013 ± 431	565 ± 87	

[†]The experiment was conducted on a previously untreated area of the same field following soybean each year.

Table 3. The effect of dried distillers grains (DDGs) on soil organic matter (SOM), pH, P, K, Ca, Mg, and grain composition in low (2003 and 2005), medium (2007), and high (2004 and 2006) yield environments. Data were combined over years with a similar yield potential since no interactions were detected within a yield environment.

Years	Rate of DDGs kg ha ⁻¹	Soil properties				Grain composition					
		SOM g kg ⁻¹	pH	Bray P	K	Ca	Mg	Oil	Protein	Starch	Extractable starch
2003 and 2005 (Low)	0	31	6.7	149	676	4600	545	35.6	86.4	746.7	657.6
	1200	31	6.7	150	792	5045	601	36.1	89.3	745.3	654.3
	2400	31	6.6	160	741	4685	575	33.9	92.7	745.3	649.5
	3600	29	6.6	140	653	4496	549	33.1	93.9	751.8	645.9
	LSD (P = 0.05)	NS	NS	NS	NS	NS	NS	2.1	4.8	NS	6.4
2007 (Medium)	0	25	6.6	109	595	4962	581	42.5	61.5	746.5	692.3
	1200	24	6.7	105	556	4988	572	42.9	62.8	744.5	690.0
	2400	25	6.6	108	546	5043	590	42.4	68.8	742.5	678.3
	3600	25	6.7	114	538	4931	574	43.3	73.5	735.3	671.5
	LSD (P = 0.05)	NS	NS	NS	NS	NS	NS	NS	4.6	7.1	7.1
2004 and 2006 (High)	0	27	6.5	102	527	4741	562	40.9	67.4	726.3	703.3
	1200	27	6.5	107	534	4952	588	39.4	69.6	727.6	699.8
	2400	28	6.5	105	523	4775	565	40.2	73.9	721.3	693.4
	3600	29	6.4	114	556	5082	612	39.9	77.1	719.6	688.1
	LSD (P = 0.05)	NS	NS	NS	NS	NS	NS	NS	4.0	5.1	9.3

[†]Abbreviations: LSD, least significant difference.; NS, not significant.

Table 4. The effect of pre-emergence applied N source at 140 kg N ha⁻¹ on grain yield, oil, protein, starch, and extractable starch compared for a low (2005), medium (2007), and high (2004 and 2006) yield environments.

Year	N source	Grain yield kg ha ⁻¹	Oil	Protein	Starch	Extractable starch
		----- g kg ⁻¹ -----				
2005 (Low)	Non-treated	4960	33.3	67.5	737.8	694.8
	DDGs	4650	33.5	66.0	739.8	694.0
	Anhydrous ammonia	8540	30.2	92.0	730.3	658.3
	PCU	8040	31.5	88.8	728.8	657.0
	LSD (P=0.05)	1130	NS	6.6	5.8	12.9
2007 (Medium)	Non-treated	5780	37.0	65.5	738.8	702.3
	DDGs	8600	37.8	72.3	738.0	685.5
	Anhydrous ammonia	9360	35.1	97.0	721.3	670.5
	PCU	10 490	32.9	91.0	729.3	678.0
	LSD (P=0.05)	1260	NS	7.8	9.0	15.7
2004 and 2006 (High)	Non-treated	6910	42.0	60.5	744.3	700.5
	DDGs	11 300	41.8	73.8	733.8	691.8
	Anhydrous ammonia	13 000	41.6	83.3	730.0	670.8
	PCU	13 060	40.8	78.8	732.2	682.3
	LSD (P=0.05)	1260	NS	4.1	4.9	6.7

[†] Abbreviations: DDGs, dried distillers grains; LSD, least significant difference.; NS, not significant; PCU, polymer-coated urea.

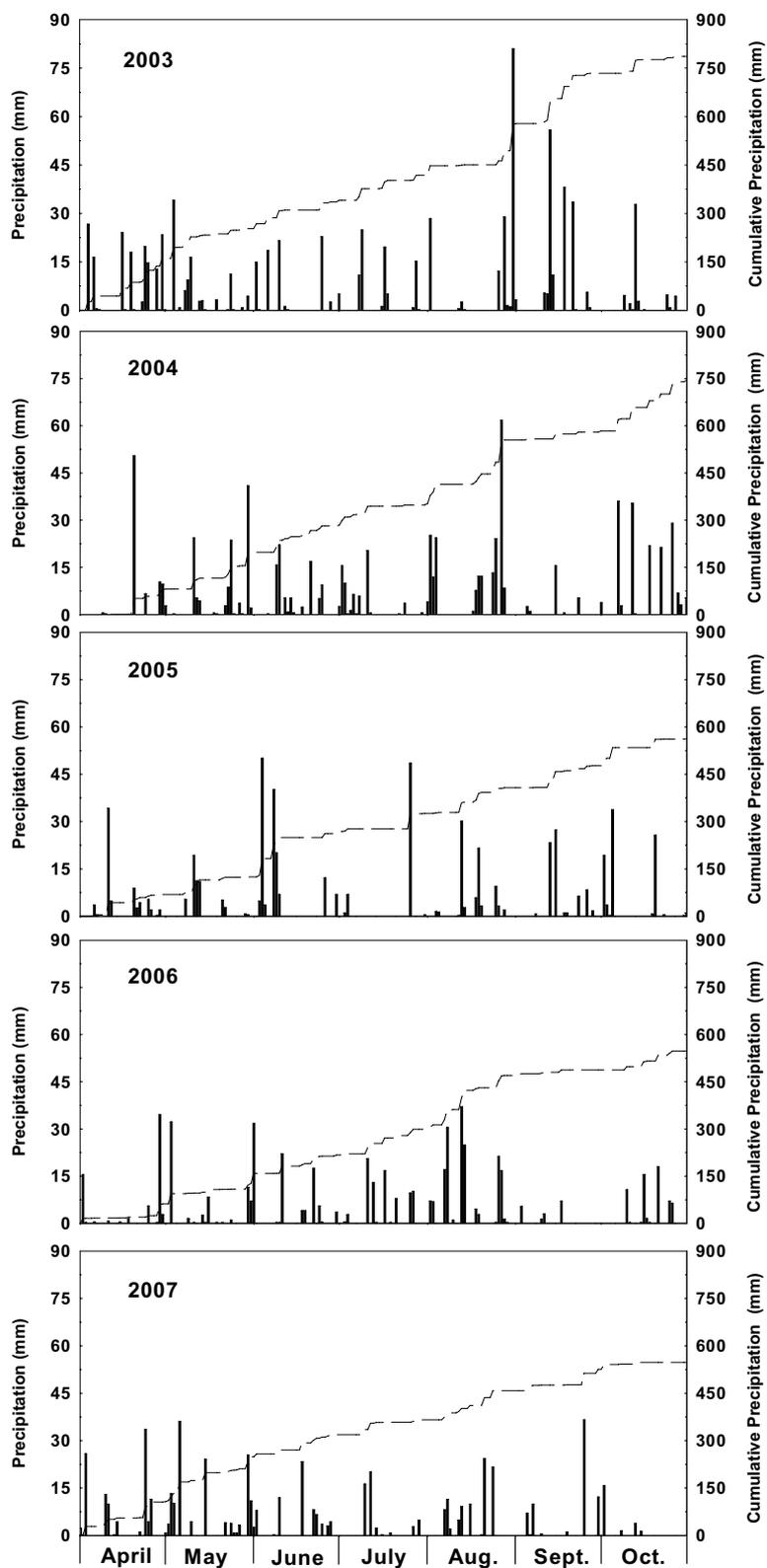


Figure 1. Daily (bars) and cumulative (line) precipitation during the growing season at the Greenley Research Center in Northeastern Missouri from 2003 to 2007

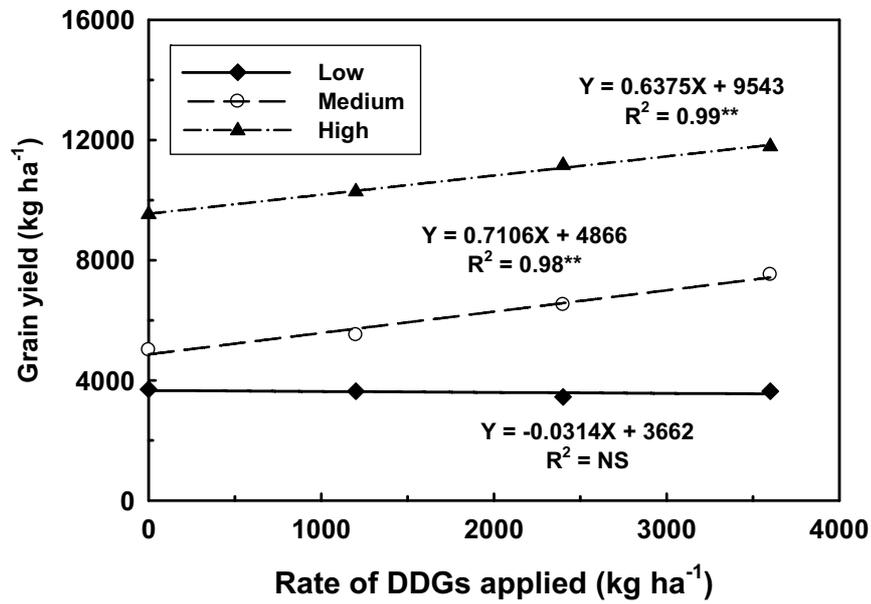


Figure 2. Grain yield response to an application of different rates of dried distiller grains (DDGs) in low (2003 and 2005), medium (2007) and high (2004 and 2006) yield environments. ** indicates statistical significance at $P = 0.01$ while NS = no significant difference among DDG rates



Zero-One Programming Model for Daily Operation Scheduling of Irrigation Canal

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Abstract

Irrigation scheduling is one of the important managerial activities that aim at effective and efficient utilization of water. A number of scheduling techniques are available today. Despite this, irrigation scheduling is only at inception level in most of developing countries. In India also there are many methods of irrigation scheduling to canals are available. The drawback of this method of operation of laterals is highlighted in this paper. Further, operations of the laterals are to be simple so that the system can be managed easily. In addition, the supply to laterals should match with the day supply in the canal and total supply for the period. In this paper a Mixed Linear Integer Programming model is described, which aims at daily scheduling of laterals from the canal considering the constraints of the system. It is proposed to run the laterals, (except a lateral which is proposed to operate at variable discharge) either full/half or closed condition for making the laterals operation simple. This Zero-One Mixed Linear Integer Programming model is applied to a field problem to derive daily operation scheduling of laterals of the system.

Keywords: Water allocation, Optimization model, Canal operation, Irrigation water, Scheduling, Zero-One programming

1. Introduction

Irrigation scheduling is the use of water management strategies to prevent over-application of water while minimizing yield loss due to water shortage or drought stress. Irrigation scheduling will ensure that water is applied to the crop when needed and in the amount needed. (Solanki.A.S -2003). Effective irrigation scheduling requires reliable and timely water delivery through coordinated action between the supplier and the user. However, allocation and delivery of water, particularly in large canal systems, is much more complex and difficult than commonly recognized. It involves complicated social, organizational, legal and economic questions in addition to the technical matters. Manual control, Automatic on site-operation of a control structure, fixed method of operation control structures are some of the alternative methods of operation of canal on basis of control of hydraulic structures adopted in scheduling warabandi, osrabandi, shejpali, block and satta system are some of the methods adopted in India in canal system operation. Also in canal operation, significant progress has been made using computer facilities, information techniques, measurements, canal control concepts, etc. in order to improve the canal operation. In real situation, however, the progress is much slower, often much below acceptable levels. The reason may be due to technical gap, financial gap or social gap. It is

therefore essential that operations should be as simple, as possible. The designer should think of ease of operation, frequent resetting of structures and complicated procedures should be avoided. In most of the irrigation projects maintenance activities are deferred because of inadequate recurrent funds. The lack of maintenance results in a rapid deterioration of public investments, including failure of canal lining and malfunctioning of control structures. The other aspects in canal water distribution are inequity in the pattern of water distribution. Head reach farmers often take advantage of their location and take an unfair share of water when the service is not reliable. Therefore the objective of this paper is to provide an improved scheduling programme which takes in to account all the above aspects.

2. Equity planning in allocation

In most of the Irrigation systems, the irrigation department is responsible for management of the system. At the beginning of the each season after ascertaining the water available in the reservoir, it announces the dates of water release to each of the outlets below the distributary. The department in the command area also announces the crop to be grown in each plot in the season before the starting of the season. However studies indicated that there is no control over the farmers by the system managers; hence violation of the cropping pattern is common in most of the canal system. (Venugopal. K, Ramaseshan. S, Ramesh. B.R (2005)). Hence prioritization of the water allocation is needed to attain equity in allocation. In the present methodology discussed in this paper, first priority is given to authorised plots which have adhered to the departmental notification. If the water is available further the water is allocated next to the plots under violation, In this, plots which are under dry crop is given first priority, since they require less water. Next priority is given the garden crop and last priority is given to wet crop. If further water is available it is allocated to unauthorised plots. Hence after knowing the areas of different crops under different category (authorized, violated.etc) water requirement of different crops can be found by Modified Penman's method (FAO Irrigation and Drainage Paper No. 56).since the crop water requirement differ in different stages of the crop, the total number of days the water supplied to the canal in the season is divided in to suitable period for computation of crop water requirement (Ex. If the number of days the canal supplied water in the season is 130 days, it can be divided in to 13 number of 10 day period block for crop water computation).

3. Model Description

Linear and Mixed integer linear programming are effectively used in scheduling irrigation canal (Anwar A. A., Clarke D.,(2001) De varies., Anwar A.A.,(2004)., Suryavanshi, A. R., and Reddy, J. M. (1986). Reddy, J. M., Wilamowski, B., and Sharmasarkar, F. C. (1999).). The present model is aimed at not only catering the demand of the laterals of the branch canal (wherein branch canal is supplied water continuously for whole season and laterals are to be controlled during the period (10 day block)) for the water requirement, but also operation of the laterals should be simple. It is decided to operate the laterals of the canal either full/half/closed condition on any day of the 10 day block. Hence The laterals are to be operated to cater this demand with the constraints (i) The sum of supplies to laterals on any day should be less than or equal to net daily supply in the canal.(ii)The sum of daily supply of the laterals for the complete period should be less than or equal to demand (iii)Other system constraints. Apart the laterals are to be supplied at minimum head of half supply capacity so that the water can reach the tail end. In the present stated operation scenario described above of the system, on any day during the period, all the laterals except one lateral will be running in full supply/half supply or closed condition. Zero-One programming model is developed for this operation policy with some modifications in the demand of the laterals. The demand modification is necessary as the laterals are to be operated either half/full/closed condition. Hence the demands of laterals for ten day period are rounded off to nearest value which is divisible by half supply capacity of the lateral (except of the lateral which is decided to operate at variable supply). Hence the demand of each lateral will be either little less or little more of their demand for the 10 day period. Cumulative excess water or deficit water available due to this adjustment is allocated to the lateral which is decided to run at variable supply. This modification does not deviate much from their original demand, if suitable lateral selected to run at variable discharge. Mixed integer linear programming model (Zero-One programming model) is used for daily operation scheduling model.

In this 0-1 integer-programming model the decision variable will be either "ZERO" or "ONE". The "ZERO" value of the decision variable may be represented as "OFF" condition of the lateral and value "ONE" as "ON" condition of the lateral.

The lateral on any day may be running at full supply or half supply or nil supply (closed). Hence, in this model three operating (full/half/closed) conditions of the lateral are to be integrated. Therefore, each lateral is imagined to comprise of two-sub channels with capacity equal to half the capacity of the lateral. A zero-one decision variable is defined for each of the sub channel of the lateral. The decision variable will take either 'ZERO' or 'ONE' on days of the period to represent 'OFF' or 'ON' condition of the sub channel respectively. The operating condition of the lateral on any day will be the sum of the decisions of the sub channels of the lateral

The sum of the supplies to laterals taking off from the branch canal on any day should be equal to net supply in the branch canal. This condition could be satisfied only when one of the laterals runs at variable supply. The decision

variable of this lateral will be a continuous variable. Hence, the model comprises of one continuous variable for a lateral, which is decided to run at variable supply and 0-1 discrete variable for each sub channel with two-sub channel for each lateral representing all other laterals. Therefore, the model is a 'Mixed Integer Linear Programming' model. The formulation of the model is explained below.

The mixed integer linear programming formulation with following constraints and objective function are used for day wise allocation of the modified releases for the periods of each lateral.

Let i represents the lateral number, j the sub channel number, k the day number in the period. For formulation, the period is assumed as 10 days. Hence k will take value between 1 and 10. Let X_{ijk} represents the 0-1 decision variable for i^{th} lateral, j^{th} sub channel on k^{th} day.

$$X_{ijk} = \begin{cases} 0 & \text{when the sub channel } j \text{ of } i^{\text{th}} \text{ lateral is closed on } k^{\text{th}} \text{ day.} \\ 1 & \text{when the sub channel } j \text{ of } i^{\text{th}} \text{ lateral is open on } k^{\text{th}} \text{ day.} \end{cases}$$

Let v represents the lateral number, which is proposed to run at variable supply. X_{vk} is defined as a continuous variable in this model represents the supply in the v^{th} lateral on k^{th} day. Following constraints are used for the canal system

3.1 Constraints:

3.1.1 Branch canal supply constraint

The daily branch canal supply at the head of the canal is not constant for the complete period. It is gauged every day.

Hence the sum of the supplies to the laterals on any day should be less than or equal to net supply (Supply at the head of branch canal minus losses in branch canal) at the head of the branch canal. Mathematical representation of this constraint is as under.

$$\sum_{\substack{i=1 \\ i \neq v}}^n \sum_{j=1}^2 \frac{c_i}{2} X_{ijk} + X_{vk} \leq q_k \quad \text{for } k=1,2,\dots,10$$

Where,

$$X_{ijk} = \begin{cases} 0 & \text{when the sub channel } j \text{ of } i^{\text{th}} \text{ lateral is closed on } k^{\text{th}} \text{ day.} \\ 1 & \text{otherwise (i.e. when the sub channel } j \text{ of } i^{\text{th}} \text{ lateral is open on } k^{\text{th}} \text{ day)} \end{cases}$$

i = Lateral number = 1,2,..... n ; v = Lateral number proposed to run at variable supply ; n = Number of laterals in the system ; c_i = capacity of the i^{th} lateral (cumec)

Since each lateral is imagined to comprise of two sub channel of capacity equal to half the lateral capacity, the term

$\frac{c_i}{2} X_{ijk}$ represents the supply in the j^{th} sub channel of i^{th} lateral on k^{th} day in cumec-day.

X_{vk} = supply in the v^{th} lateral on k^{th} day (cumec). This is a Continuous variable (i.e. the lateral v runs at variable supply); k = Day number in the period. It is assumed ten day period is followed in this system hence $k= 1, 2, \dots, 10$; q_k = Net supply in the branch canal on k^{th} day (cumec) = $Q - L_s$; Q = Supply in the branch canal at the head (cumec) ; L_s = Branch canal seepage loss (cumec)

3.1.2 Lateral demand constraint

The supply to the lateral during any period should not exceed its modified releases for the period. This constraint is represented by

$$\sum_{k=1}^{10} \sum_{j=1}^2 \frac{c_i}{2} X_{ijk} \leq D_i \quad \text{for all } i=1,2,\dots,n \text{ and } i \neq v$$

and for lateral v running at variable supply

$$\sum_{k=1}^{10} X_{vk} \leq D_v$$

where D_i = modified release of i^{th} lateral for the period (cumec-days); D_v = modified release of the v^{th} (variable supply lateral) lateral for the period (cumec-days)

3.1.3 Lateral capacity constraint

On any day in the period, the supplies to lateral cannot exceed the capacity of the lateral. As the laterals are proposed to be operated at full/half/closed condition the total supply to lateral for the period will be in multiple of half capacity.

Each lateral is imagined to comprise of two sub channel of capacity equal to half the lateral capacity. The term $\frac{c_i}{2} X_{ijk}$ represents the supply in the j^{th} sub channel of i^{th} lateral on k^{th} day in cumecs. X_{ijk} is a discrete decision variable takes value 0-1 and c_i represents the capacity of lateral i . The sum of supplies of two sub channels ($j=1$ and 2) of i^{th} lateral on k^{th} day gives the supply on k^{th} day of the period to i^{th} lateral. Hence declaring X_{ijk} as a 0-1 variable in the model can satisfy lateral capacity constraint.

One of the laterals in the system is proposed to be run at variable supply to adjust the daily supply in the branch canal. This lateral supply on any day of the period should be maintained at least or above half supply in order that lateral runs with sufficient depth of flow. This constraint is mathematically represented as below.

$$X_{vk} \leq c_v$$

And
$$X_{vk} \geq \frac{c_v}{2}$$

for all $k=1,2,\dots,10$ days.

3.1.4 Branch canal sectional capacity constraint

The cross section of branch canal is not constant in complete reach of the canal. Hence the flow in the branch canal at any section can't exceed the design capacity of the canal at that section. This constraint is formulated as follows.

The sum of the supplies to the laterals downstream of the section 's' (say) and the branch canal conveyance loss below this section on any day in the period, should not exceed the branch canal capacity at the section s.

If the section s is upstream of lateral v the constraint is written as

$$\sum_{\substack{i=m_s \\ i \neq v}}^n \sum_{j=1}^2 \frac{c_i}{2} X_{ijk} + X_{vk} \leq C_s - l_s$$

for all $k=1,2,\dots,10$

If the section s is down stream of lateral v the constraint is written as

$$\sum_{i=m_s}^n \sum_{j=1}^2 \frac{c_i}{2} X_{ijk} \leq C_s - l_s$$

for all $k=1,2,\dots,10$

where,

m = lateral number just downstream of the section s.

C_s = Capacity just below the section s in the branch canal (cumec)

l_s = seepage loss below the section (cumec)

s= section in branch canal

3.1.5 Objective Function

The main objective of this MILP modeling is daily allocation of the net available water at the head of the branch canal to the laterals of the system in accordance with the modified releases for the period of the lateral, operation criteria of the lateral and the constraints cited above. Therefore, the objective function is defined as maximization of the total supply to the laterals for the period. If the constraints are all satisfied for the period the MILP objective function results will be equal to net supply at the head of the branch canal for the period. The mathematical representation of the objective function is,

$$\text{Maximise } \sum_{k=1}^{10} \sum_{\substack{i=1 \\ i \neq v}}^n \sum_{j=1}^2 \frac{c_i}{2} X_{ijk} + \sum_{k=1}^{10} X_{vk}$$

where X_{ijk} = discrete variable for j^{th} sub channel of i^{th} lateral on k^{th} day

$$X_{ijk} = \begin{cases} 0 & \text{when the sub channel } j \text{ of } i^{\text{th}} \text{ lateral is closed on } k^{\text{th}} \text{ day.} \\ 1 & \text{otherwise} \end{cases}$$

c_i = capacity of i^{th} lateral

X_{vk} = supply in v^{th} lateral on k^{th} day. (v^{th} lateral runs at variable supply)

For illustration the model is applied to Harihar Branch canal of Bhadra canal system in Karnataka state.

4. Illustration of model

The Harihar branch canal is a sub system of Bhadra irrigation system in Karnataka state of India. It is designed for discharge capacity of $10.3 \text{ m}^3/\text{s}$. runs for length of 22 kms and irrigates 14 996 Ha. The canal irrigates both in Rabi (general period between January and June) and Kharif (general period between July and December) season. The branch canal comprise of 18 outlets/ distributaries. The details of the distributaries are shown in fig 1. For the analysis the cropping pattern of Kharif 2002 is considered. During this season, the branch canal was operated continuously between 12th July and 20th November for 132 days. There were 13 periods. First and thirteenth periods were of 11 days and remaining periods are of 10 day periods. The crop water requirement of each lateral after prioritization is found for the period. The demand of each lateral of the period is rounded off to nearest half supply capacity, except of the lateral which is decided to run at variable supply (Third lateral is decided to operate at variable supply). The model of daily scheduling of lateral is illustrated for third period of 10 days. Table 1 indicates the allocation procedure for the laterals of the distributary. The total supply for the period at the head of the Harihar branch canal on different days of the period and for ten days are indicated in column 2 is available for allocation to different laterals. The crop water requirement for authorised and violated crops (Dry crops were absent in this season) of different laterals are computed and are given in column 6 and column 7. After prioritization (i.e. water demand of laterals of authorised crops are met as first priority-further water available are proportionately allocated violated crops) demand of lateral for the period is shown in column 9. Modified demand of laterals after rounding off to nearest half supply capacity (except third lateral which is decided to operate at variable supply) is given in column 10. Hence the demands in the tenth column of different laterals are taken in the Mixed Integer Programming model. The programming model is run for the third period and the daily allocation to different model is given in Table 2. The model shows how the lateral is to be operated during the period to cater the demand. For example, the demand the lateral 1 is catered by keeping the lateral open on all days of the period. The demand of lateral 12 for the period is catered by operating the lateral at half on 1, 2, 7 and 9th day, full on 3,4,5,6 and 8th day and closed on 10th day. Hence This methodology is quite simple and efficient operation schedule which can be adopted to any canal system.

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Table 1. Allocation to laterals of Harihar Branch canal for Third Period

1	2	3	4	5	6	7	8	9	10
Day	Net Supply at head of canal (cumec)	Lateral No	Capacity of the lateral (cumec)	Capacity of lateral for the period (cumec-days)	water demand for authorised crops for the period (cumec-days)	water demand for violated crops for the period (cumec-days)	Water release for violated crop for the period (cumec-days)	Net release for the period on equitable basis (=col(6)+col(8)) (cumec-days)	Modified allocation for the period (cumec-days)
1	8.7	1	1.19	11.89	1.40	16.21	10.49	11.89	11.89
2	8.7	2	0.06	0.58	0.00	0.83	0.58	0.58	0.58
3	8.7	3	1.84	18.41	0.00	17.29	15.58	15.58	15.94
4	8.51	4	0.06	0.65	0.00	1.15	0.65	0.65	0.65
5	8.7	5	0.25	2.47	0.00	2.50	2.26	2.26	2.22
6	8.7	6	0.16	1.57	0.00	1.60	1.45	1.45	1.41
7	8.7	7	0.11	1.11	0.00	1.78	1.11	1.11	1.11
8	8.51	8	0.09	0.88	0.11	2.24	0.77	0.88	0.88
9	8.7	9	0.08	0.80	0.00	0.51	0.46	0.46	0.48
10	8.51	10	1.76	17.65	1.10	31.43	16.55	17.65	17.65
		11	0.08	0.82	0.00	2.03	0.82	0.82	0.82
		12	0.25	2.50	0.14	1.83	1.65	1.79	1.75
		13	0.08	0.85	0.04	0.23	0.21	0.24	0.25
		14	0.05	0.49	0.07	0.99	0.42	0.49	0.49
		15	1.45	14.46	1.19	27.38	13.27	14.46	14.46
		16	1.24	12.38	0.75	0.86	0.77	1.52	1.24
		17	0.05	0.49	0.04	1.18	0.45	0.49	0.49
		18	1.41	14.10	0.00	25.96	14.10	14.10	14.10
	86.42		10.21	102.10	4.83	135.99	81.59	86.42	86.42

Table 2. MIP Day wise allocation of demands of laterals for Third Period -Kharif season-2002

Lat No	Capacity of lateral (cumec)																		Total allocation (cumec)
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	
Day Number	1.19	0.06	1.84	0.06	0.25	0.16	0.11	0.09	0.08	1.76	0.08	0.25	0.08	0.05	1.45	1.24	0.05	1.41	8.70
1	1.189	0.058	1.773	0.065	0.247	0.157	0.111	0.088	0.040	1.765	0.082	0.125	0.042	0.049	1.446	0.000	0.049	1.410	8.70
2	1.189	0.058	1.773	0.065	0.247	0.157	0.111	0.088	0.040	1.765	0.082	0.125	0.042	0.049	1.446	0.000	0.049	1.410	8.70
3	1.189	0.058	1.691	0.065	0.247	0.157	0.111	0.088	0.040	1.765	0.082	0.250	0.000	0.049	1.446	0.000	0.049	1.410	8.70
4	1.189	0.058	1.425	0.065	0.247	0.157	0.111	0.088	0.080	1.765	0.082	0.250	0.042	0.049	1.446	0.000	0.049	1.410	8.51
5	1.189	0.058	1.769	0.065	0.247	0.078	0.111	0.088	0.040	1.765	0.082	0.250	0.000	0.049	1.446	0.000	0.049	1.410	8.70
6	1.189	0.058	1.608	0.065	0.247	0.157	0.111	0.088	0.080	1.765	0.082	0.250	0.042	0.049	1.446	0.000	0.049	1.410	8.70
7	1.189	0.058	1.816	0.065	0.247	0.157	0.111	0.088	0.040	1.765	0.082	0.125	0.000	0.049	1.446	0.000	0.049	1.410	8.70
8	1.189	0.058	1.548	0.065	0.123	0.157	0.111	0.088	0.080	1.765	0.082	0.250	0.042	0.049	1.446	0.000	0.049	1.410	8.51
9	1.189	0.058	1.197	0.065	0.247	0.157	0.111	0.088	0.040	1.765	0.082	0.125	0.000	0.049	1.446	0.619	0.049	1.410	8.70
10	1.189	0.058	1.338	0.065	0.123	0.078	0.111	0.088	0.000	1.765	0.082	0.000	0.042	0.049	1.446	0.619	0.049	1.410	8.51
Total allocation (cumec-days)	11.89	0.58	15.94	0.65	2.22	1.412	1.11	0.88	0.48	17.65	0.82	1.75	0.25	0.49	14.46	1.24	0.49	14.102	86.42
Demand (cumec-days)	11.89	0.58	15.94	0.65	2.22	1.41	1.11	0.88	0.48	17.65	0.82	1.75	0.25	0.49	14.46	1.24	0.49	14.102	

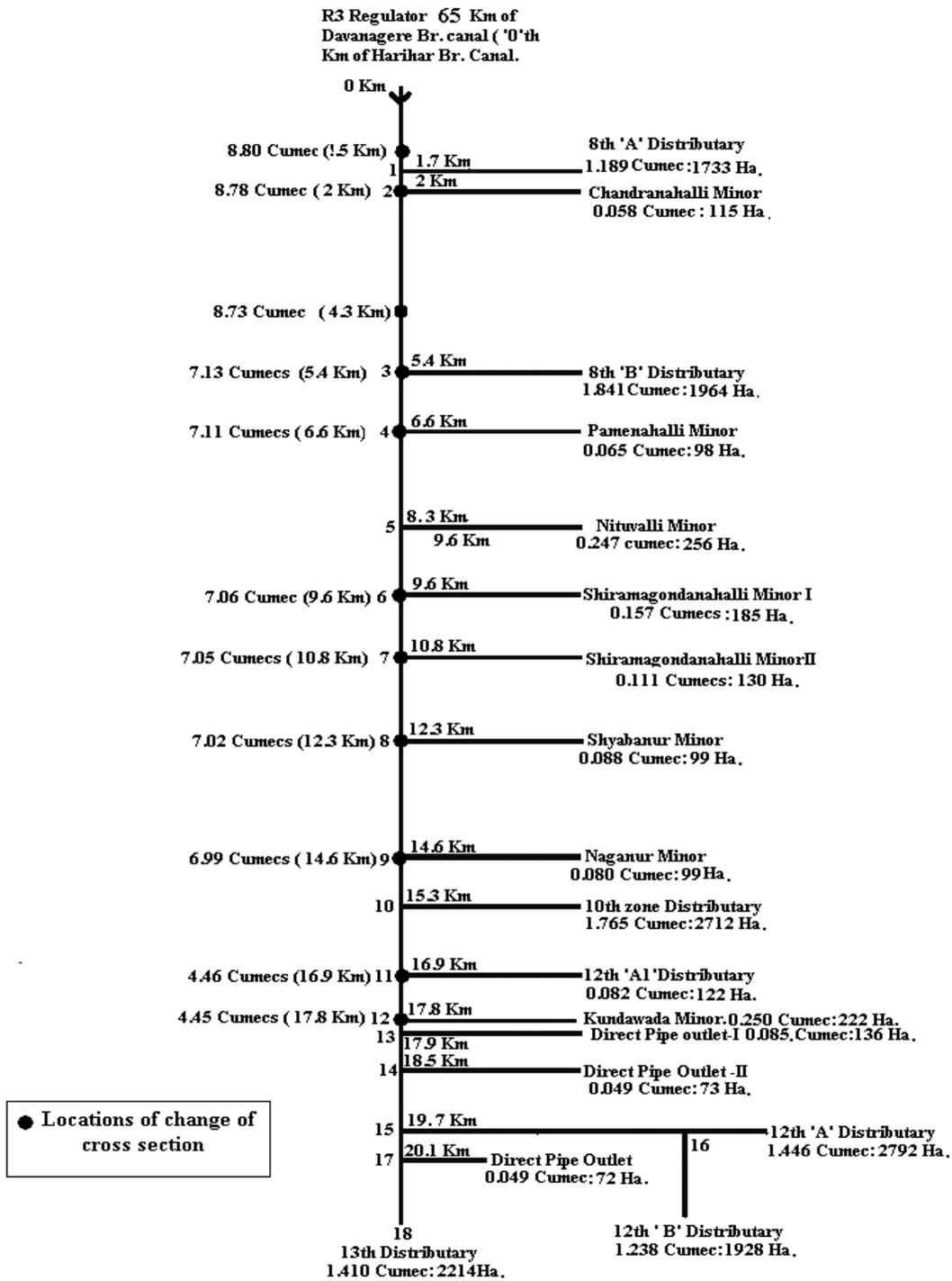


Figure 1. Tree Plan of with details of Harihar Branch Canal of Bhadra Command Area



The Grey Situation Decision Analysis on Regional
Distribution of Grain and Oil Crops
-----A Case Study of Sichuan Province in China:
In Respective of Food Supply

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This paper is funded by the project "Research on Stable Production and Supply of Hog in China" (N1501) sponsored by the Agricultural Census Office of the State Council, and the project "Research on Existing Circular Agricultural Industry Chain Development in the Hilly Region of Middle Sichuan Province and Countermeasures" (SC07B059) sponsored by the Philosophy and Social Science Funds of Sichuan Province.

Abstract

Regional crops distribution is an import subject for developing modern agriculture now. Based on existing distribution of 7 grain and oil crops in Sichuan province, this paper chose 5 economic evaluation indexes to discuss the regional distribution of grain and oil corps in Sichuan with the method of grey situation decision. According to the row decision and column decision, this paper shows that the sorting result of the advantage grain and oil crops in 21 cities and prefectures of Sichuan province and the adaptability sorting result of 7 grain and oil crops in each region in Sichuan province, which provides scientific basis for the distribution of grain and oil industry in Sichuan province.

Keywords: Regional distribution, Grey situation decision, Grain and oil crops

Agricultural distribution is, in certain spatial scope, to study and program the adjustments and changes of agricultural production structure from a spatial angle, based on existing agricultural production distribution, and identify the internal contradictories in former agricultural distribution, offering services for regions providing macro sorting directions and micro implementations for agricultural production (Regional Economics Study Institute of Renmin University of China, 1996). Crops distribution is an important part of agricultural production distribution, which helps to realize the optimal production effect in the whole region by distributing crops in space scientifically. Today, developing modern agriculture becomes the theme of new countryside construction. How to improve the crops distribution is one of important subjects for all regions developing modern agriculture.

Most studies on crops distribution focus on qualitative analysis, drawing conclusions according to subjective senses or experiences, and seldom on quantitative analysis. Theoretical studies on the distribution methods are few (Dehua Jiang, 1983, p188-195; Dianting Wu & Chuanzhou Wang, 1998, p554-558). The mathematical models for industrial distribution includes linear programming model, economic metrology model, multi-objective programming model, non-linear programming model, dynamic programming model, fuzzy programming model, etc. The linear programming model is widely used in researches on agricultural crops distribution (Hongyan Zhai, 2006, p82-83; Feihong Gong & Tianfu Liu, 1986, p29-36). But because of the changeable nature of objective function along with the time sequence, the application of linear mathematical method faces some problems (Tianshun Li, 1987, p31-34). The gray situation decision model is also widely used for agricultural crops distribution (Xinyu Tong, Yuhui Liu & Jianfeng Zhou, 1993, p121-123; Daobo Wang, Xiaoguo Zhou & Guanglu Zhang, 2004, p16-18).

Along with the unification of urban area and countryside, and the urbanization, food safety has already become an urgent issue. Sichuan province, as an agricultural giant in middle China, has excellent natural and geographical conditions for developing agriculture. The development of grain and oil industry is meaningful for realizing regional food safety. Sichuan province must raise 6.8% of Chinese population by 4.2% of national cultivated lands. More people with less cultivated lands gives more pressure on the cultivated lands. To stabilize the areas of cultivated lands for crops and implement a scientific regional distribution of grain and oil crops, by improving the unit yield, is meaningful for driving the development of grain and oil industry, releasing the contradiction between population and lands, realizing optimal agricultural structure, and transferring rural surplus labor powers. This paper adopts the grain situation decision model from the grain system theory, takes 7 grain and oil crops in Sichuan province as objects, based on existing grain and oil production distribution in 21 cities (states) in Sichuan province, select 5 economic evaluation indexes, analyze the comparative advantages of different regions in grain and oil production, and discusses the regional distribution of grain and oil crops in Sichuan province, with the hope of offering scientific basis for the structure adjustment of grain and oil industry in Sichuan province.

1. The gray situation decision analysis method

The gray situation decision (Julong Deng, 2005) is a decision analysis method that uses a mathematic language and makes decisions match up with incidents by taking all decision factors into consideration. The gray situation decision is maybe single-objective decision or multi-objective decision. This paper adopts the multi-objective decision. Its mathematical principle and process are as follow.

1.1 Construct the decision matrix

Suppose there are m incidents in the gray situation decision, namely a_i ($i = 1, 2, \dots, m$); In correspondence to m incidents, there are n decisions, namely b_j ($j = 1, 2, \dots, n$). Name the combination of incident a_i and decision b_j as situation $s_{ij} = (a_i, b_j)$. It means the j decision (b_j) is for the i incident (a_i). The effect measurement value of situation s_{ij} is r_{ij} . Every element in the decision matrix is a decision unit, namely:

$$\frac{r_{ij}}{s_{ij}} = \frac{r_{ij}}{(a_i, b_j)}$$

The decision matrix is:

$$M = \begin{pmatrix} \frac{r_{11}}{s_{11}} & \frac{r_{12}}{s_{12}} & \dots & \frac{r_{1m}}{s_{1m}} \\ \frac{r_{21}}{s_{21}} & \frac{r_{22}}{s_{22}} & \dots & \frac{r_{2m}}{s_{2m}} \\ \vdots & \vdots & \ddots & \vdots \\ \frac{r_{n1}}{s_{n1}} & \frac{r_{n2}}{s_{n2}} & \dots & \frac{r_{nm}}{s_{nm}} \end{pmatrix} \quad (1)$$

1.2 The effect measurement

The effect measurement is to quantitatively measure the effect generated from the situation. It is a specific measurement of practical effect of the situation by comparative analysis. The effect measurement includes three types, namely:

$$(1) \text{ The top effect measurement, } r_{ij} = \frac{u_{ij}}{u_{\max}}$$

$$(2) \text{ The bottom effect measurement, } r_{ij} = \frac{u_{\min}}{u_{ij}}$$

$$(3) \text{ The medium effect measurement, } r_{ij} = \frac{\min(u_{ij}, u_0)}{\max(u_{ij}, u_0)}$$

Here, u_{ij} is the practical effect value of situation s_{ij} . u_{\max} is the maximum practical effect value of all

situations S_{ij} . u_{\min} is the minimum practical effect value of all situations S_{ij} . u_0 is an appointed medium value.

In practice, it is the nature of decision objective that determines the type of measurement. If we pursue for the largest objective value, we can select the top effect measurement. If we want the smallest objective value, we can adopt the bottom effect measurement. If we prefer to a proper value, we can take the medium effect measurement.

1.3 The matrix for calculating the integrated effect measurement

Suppose the situation has p objectives. The effect value of objective k is $r_{ij}^{(k)}$ and its decision unit is $\frac{r_{ij}^{(k)}}{s_{ij}}$. By the decision matrix $M^{(k)}$, we can construct the multi-objective integrated decision matrix, namely:

$$M^{(\Sigma)} = \begin{pmatrix} \frac{r_{11}^{(\Sigma)}}{s_{11}} & \frac{r_{12}^{(\Sigma)}}{s_{12}} & \dots & \frac{r_{1m}^{(\Sigma)}}{s_{1m}} \\ \frac{r_{21}^{(\Sigma)}}{s_{21}} & \frac{r_{22}^{(\Sigma)}}{s_{22}} & \dots & \frac{r_{2m}^{(\Sigma)}}{s_{2m}} \\ \vdots & \vdots & \ddots & \vdots \\ \frac{r_{n1}^{(\Sigma)}}{s_{n1}} & \frac{r_{n2}^{(\Sigma)}}{s_{n2}} & \dots & \frac{r_{nm}^{(\Sigma)}}{s_{nm}} \end{pmatrix} \tag{2}$$

Here in this matrix, $r_{ij}^{(\Sigma)}$ is the integrated effect measurement value of situation S_{ij} . It is the final effect value by integrating multiple objectives. If we endow with each objective with average weight, the calculation is:

$$r_{ij}^{(\Sigma)} = \frac{1}{p} \sum_{k=1}^p r_{ij}^{(k)}$$

1.4 Make decision

According to the multi-objective integrated decision matrix, we can use two ways to select the situation with the most excellent effect, or select the maximum integrated effect measurement value $r_{ij}^{(\Sigma)}$ among the matrix $M^{(\Sigma)}$.

(1) Row decision. Select the largest integrated effect measurement value $r_{ij}^{(\Sigma)}$ from the each row of the multi-objective integrated decision matrix $M^{(\Sigma)}$. The situation S_{ij} is the best, what means the decision b_j is the best choice for the incident a_i .

(2) Column decision. Select the largest integrated effect measurement value $r_{ij}^{(\Sigma)}$ from each column of the multi-objective integrated decision matrix $M^{(\Sigma)}$. The situation S_{ij} is the best, what means the incident a_i is most appropriate incident for the decision b_j .

2. Demonstration on the gray situation decision analysis method

2.1 Select evaluation indexes

This paper aims at studying the regional distribution of grain and oil crops in Sichuan province. The crops include rice, wheat, corn, legume, potato, rapeseed, and peanut. Select five evaluation indexes displayed in table 1. According to data in Sichuan Statistical Yearbook in recent four years (Statistics Bureau of Sichuan Province, 2004-2007), we calculate the decision matrix and finally get the integrated effect measurement matrix.

2.2 Results of model calculation

According to the similarities of time sequence, get the average of each index based on data of 2003 to 2006. Firstly, calculate the decision matrix of the five evaluation indexes based on model (1). Then, calculate the integrated effect measurement matrix based on model (2). In calculation, we endow the five indexes with average weights. Finally, we get the table 2 (display only integrated effect measurement values).

2.3 Make decision

Make decision according to the results of gray situation decision and identify the crops with comparative advantages in different regions. Results are in table 3 (three crops for each region).

Made decision according to the results of gray situation decision and get the sequence of 7 grain and oil crops according

to their comparative advantages in the 21 cities (states) in Sichuan province. Table 4 displays the results (list six regions for each crop).

3. Conclusion

This paper analyzes the regional distribution of grain and oil crops in Sichuan province by the gray situation decision analysis method. Based on data calculation, this paper orders the sequence of grain and oil crops according to their comparative advantages in the 21 cities (states) in Sichuan province. Results show that structures of grain and oil crops with comparative advantages in different regions are similar. Most regions possess comparative advantages in rice, wheat, and corn. The reason may lie in the relatively sound basis of land crops. By ordering the sequence of 7 grain and oil crops' regional integrated measurements in Sichuan province, establish the most appropriate regions for planting 7 grain and oil crops respectively in Sichuan province. By comparing the research result and existing regional distribution of grain and oil crops, we find that existing distribution strays away from this research result to certain degree. Present regional distribution of grain and oil crops is unscientific.

Considering the large population but less cultivated lands, to improve the regional distribution of grain and oil crops in Sichuan province is urgent and also an important issue for establishing regional development programs. In the overall programming distribution in Sichuan province, establish the regional adaptability of each grain and oil crop and increase the planting areas in the region with high adaptability. In the regional programming distribution, establish the comparative advantage of each grain and oil crop in one region and increase the planting area of the crop with high comparative advantage. This paper rightly offers scientific basis for the regional distribution of grain and oil crops in Sichuan province. We should coordinate different relationships, and implement the regional distribution scientifically and reasonably, hoping to realize the goal of ensuring food safety, releasing pressures from limited lands, transferring rural surplus labor powers, optimizing agricultural structure, and driving rural economic development.

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Table 1. The evaluation indexes for the regional comparison of grain and oil crops in Sichuan province.

No.	Index	Unit	Type of effect measurement
1	Unit yield of grain and oil crops in one region	kg/hm ²	Top effect measurement
2	Output per capita in agricultural industry	kg/per capita	Top effect measurement
3	Proportion of cultivated land for one crop to the cultivated land for all regional grain and oil crops	%	Top effect measurement
4	Proportion of regional cultivated land for one crop to the provincial cultivated land for this crop	%	Top effect measurement
5	Proportion of regional output of one crop to provincial output of this crop	%	Top effect measurement

Table 2. Integrated effect measurement.

Region	Rice	Wheat	Corn	Legume	Potato	Rapeseed	Peanut
Chengdu	0.75566	0.39553	0.22287	0.16630	0.25669	0.35385	0.14430
Zigong	0.65274	0.27367	0.28315	0.16350	0.29275	0.09269	0.12683
Panzhihua	0.47824	0.20859	0.30151	0.14358	0.14672	0.03463	0.04132
Luzhou	0.67157	0.22010	0.28126	0.19022	0.34556	0.07395	0.06956
Deyang	0.67333	0.44979	0.29917	0.15476	0.23488	0.31932	0.17443
Mianyang	0.54308	0.44817	0.39131	0.14113	0.25601	0.38667	0.28876
Guangyuan	0.45489	0.38982	0.39925	0.12118	0.23172	0.24439	0.26753
Suining	0.43597	0.40120	0.40747	0.21930	0.39474	0.22957	0.19234
Neijiang	0.48744	0.29345	0.35356	0.15454	0.35857	0.14207	0.16718
Leshan	0.53325	0.12535	0.27637	0.15000	0.27488	0.13834	0.08246
Nanchong	0.52065	0.48587	0.41404	0.40756	0.38447	0.31544	0.50396
Meishan	0.63249	0.36058	0.25608	0.12814	0.27028	0.20006	0.10783
Yibin	0.63307	0.28893	0.34327	0.16584	0.34198	0.09363	0.17658
Guang'an	0.61025	0.26385	0.30953	0.22869	0.32548	0.18432	0.15502
Dazhou	0.60909	0.26568	0.45274	0.31997	0.48069	0.38202	0.19175
Yan'an	0.41590	0.19581	0.34091	0.15603	0.23385	0.13291	0.06415
Bazhong	0.41841	0.34423	0.44607	0.12698	0.36725	0.23135	0.10993
Ziyang	0.45687	0.39643	0.46583	0.25391	0.43005	0.26707	0.25138
A'ba	0.11177	0.15159	0.34525	0.22278	0.21784	0.03890	0.00000
Ganzi	0.16637	0.20951	0.28568	0.16290	0.17776	0.04751	0.02261
Liangshan	0.36480	0.27108	0.46818	0.27553	0.37924	0.06190	0.04761

Table 3. The grain and oil crops with comparative advantages in different regions in Sichuan province.

Region	1	2	3	Region	1	2	3
Chengdu	Rice	Wheat	Rapeseed	Meishan	Rice	Wheat	Potato
Sigong	Rice	Potato	Corn	Yibin	Rice	Corn	Potato
Panzhihua	Rice	Corn	Wheat	Guang'an	Rice	Potato	Corn
Luzhou	Rice	Potato	Corn	Dazhou	Rice	Potato	Corn
Deyang	Rice	Wheat	Corn	Ya'an	Rice	Corn	Potato
Mianyang	Rice	Wheat	Rapeseed	Bazhong	Corn	Rice	Potato
Guangyuan	Rice	Corn	Wheat	Ziyang	Corn	Rice	Potato
Suining	Rice	Corn	Wheat	A'ba	Corn	Legume	Potato
Neijiang	Rice	Potato	Corn	Ganzi	Corn	Wheat	Rice
Leshan	Rice	Potato	Corn	Liangshan	Corn	Potato	Rice
Nanchong	Rice	Peanut	Wheat				

Table 4. The regional adaptability of grain and oil crops in Sichuan province.

Crop	1	2	3	4	5	6
Rice	Chengdu	Deyang	Luzhou	Zigong	Yibin	Meishan
Wheat	Nanchong	Deyang	Mianyang	Suining	Ziyang	Chengdu
Corn	Liangshan	Ziyang	Dazhou	Bazhong	Nanchong	Suining
Legume	Nanchong	Dazhou	Liangshan	Ziyang	Guang'an	A'ba
Potato	Dazhou	Ziyang	Suining	Nanchong	Liangshan	Bazhong
Rapeseed	Mianyang	Dazhou	Chengdu	Deyang	Nanchong	Ziyang
Peanut	Nanchong	Mianyang	Guangyuan	Ziyang	Suining	Dazhou



Population Ecology of Whitefly, *Bemisia tabaci*, (Homoptera: Aleyrodidae) on Brinjal

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This project is financed by Ministry of Higher Education, Malaysia (PASCA)

Abstract

Whitefly, *Bemisia tabaci* (Gennadius), a common insect feeding on plants, belongs to the family Aleyrodidae of the order Homoptera. The quantity of food source especially brinjal, is one of the major factors that has attracted whitefly in the area. In fact, the flight ability of whitefly enables them to search for food quickly. Thus could encourage whitefly to reproduce in great numbers and subsequently cause severe infestation in the fields. Many farmers are not interested to grow brinjal after they have gone through some bad experience due to some whitefly infestations, which have resulted in a total crop loss of brinjal fruits. At present, information on the population dynamics of whiteflies locally on brinjal is still lacking. Henceforth, these studies are indeed appropriate to generate a comprehensive understanding on the insect population, which could support an effective pest management programme and crop improvement strategy. The study was conducted at the Field Laboratory of the Faculty of Applied Science, Universiti Teknologi MARA, Shah Alam. The study on the population of whitefly larvae on brinjal plants covered all the plant strata except for the upper stratum. The populations of whitefly were aggregated (Taylor's Power Law Calculate) in first and second cropping of brinjal plants. It may be concluded that the total number of whitefly larvae were found to be most abundant in the middle stratum of the brinjal plants.

Keywords: Whitefly, *Bemisia tabaci*, Population ecology, Population dynamics

1. Introduction

Initial studies showed that the whitefly *Bemisia tabaci* Gennadius (sweetpotato whitefly) were found in abundance in the lowland areas. This study was carried out to establish the extent which its population and migration would result in colonization of a new habitat by this pest. Furthermore, the economic importance of whitefly is becoming to be a major insect pest especially on brinjal. Brinjal is one of the most popular fruit vegetables grown in Malaysia. Brown and Nelson (1986) reported that *Bemisia tabaci* has caused problems to the crop as a result of direct feeding damage and indirect

damage by acting as a vector for several viral plant pathogens such as gemini viruses and clostero viruses (Duffus, 1996). Primarily, this study focused on the population and distribution of whitefly which influenced the development of whitefly in the field. The distribution patterns of whitefly, within and between plants were observed for over two cropping periods. Most estimated populations of whitefly are immature stages which are easier to count. Leaf sampling was carried out to determine the population density and distribution of immature whitefly on brinjal.

2. Materials and methods

The field experiment was conducted at the experimental plot of the Plantation Technology and Management Programme, Faculty of Applied Sciences, Universiti Teknologi MARA, Shah Alam, Selangor. The plot consisted of four rows of planting beds, each measuring 10 m (length) x 1 m (width) and the distance between beds was 1 m. Each bed was planted with 12 plants of 60 cm apart between plants. A total of 48 plants were planted in four beds. Two cropping periods of brinjal were observed in the experiment. The first crop began in September 2002 until March 2003 and the second crop commenced from January to May 2003. An overlapped cropping period was planned in order to ensure availability of host plants for continuity of whitefly population. The larvae and pupae of whitefly were monitored through leaf random sampling. Sampling of larvae and pupae commenced two weeks after transplanting as brinjal plants started flushing new leaves. Six leaves were randomly sampled per planting bed. From six leaves, two were taken from each of the following plant strata namely the upper, middle, and lower, representing various stages and distribution of leaves on the plants (Kohji et al., 1993). Leaf samples were kept in plastic bags labelled with bed number and leaf stratum. The leaves were then brought to the laboratory where the larvae and pupae of whitefly were counted under a dissecting microscope. Sampling was carried out weekly until the end of the fruiting stage (13 weeks after transplanting) in both crops.

3. Data analysis

Means abundance of weekly whitefly and other insects infesting brinjal were determined and compared between two cropping periods. The data were subjected to one way ANOVA (SAS Programme) (Sandra and Ramon, 1987), and significantly different means ($p < 0.05$) were separated using Duncan Multiple Range Test (DMRT) at 5% probability. The data were found normally distributed in both cropping periods (Kolmogorov-Smirnov test (SPSS)). Hence, data of means and variances of immature whitefly were $\log_{10} x$ transformed to normalize the data for analysing spatial distribution. The degree of whitefly aggregation was evaluated based on a simple empirical relationship as $\log s^2 = \log a + b \log x$ of Taylor's Power Law Linear Regression (Taylor, 1961 and Taylor, 1984). The slope, b of this regression is a measure of aggregation, random and regular, and the y axis intercept, a , is a sampling factor. The value of b , which is significantly larger than 1 ($b > 1$) would indicate aggregation, which is less than one ($b < 1$). It means that the dispersion is uniform while values that are not significantly different from 1 ($b = 1$) shows a random distribution. Differences in mean abundance of *B. tabaci* during the study period were analysed using ANOVA (one way).

4. Results and discussion

Figure 1 shows that total mean larvae (per 6 leaves) of all strata in first cropping was lower than second cropping except at four weeks after transplanting (WAT). In the first cropping the larval population increased to a peak of 39.5 larvae per plant at 4 WAT after which the population decreased drastically at 6 WAT and gradually to very low levels until the end of the study.

<<Figure 1: Distribution and abundance of whitefly larvae on each brinjal plant (total mean larvae in three leaf strata) in two cropping periods.>>

In the second cropping, the population was much higher than the first cropping starting just after transplanting, reaching a high peak of 50 larvae per plant at seven WAT. The population decreased only slightly until the end of cropping. In the first cropping, the distribution of total *B. tabaci* larvae varied among the three strata of the host plants with the means of 2.13, 5.90, and 9.88 for the upper, middle and lower strata, respectively (Figure 2). In the second cropping, the distribution of mean larvae varied among the three strata of the host crop within the means of 3.52, 24.82, and 11.18 for the upper, middle and lower strata, respectively (Figure 2). The distribution of population means was significantly different in both stratum and cropping period at $p < 0.01$ ($F = 6.79$; $df = 2, 5$; $p < 0.05$). The distribution of the whitefly larvae was strongly dependent on strata during different cropping periods as the interaction between strata and WAT was different and highly significant. At the upper stratum, the mean number of whitefly larvae was 0.75 at 2 WAT and increased to the highest peak of 4.5 larvae at 3 WAT (Figures 2a). The larval population decreased to 4.25 larvae and drastically to its lowest level (0.5) in 8 WAT. Then, it slightly fluctuated until 13 WAT with 2 larvae on 2 leaves. Initially, the mean number at 2 WAT was 7.0 and started to increase to 10.5 larvae at 3 WAT. The highest mean number of larvae was observed to be 22 larvae at 4 WAT and drastically decreased at 5 WAT. Subsequently, the insect population fluctuated but it did not exceed 5.25 larvae for the following WAT's until the end of first cropping. Figure 2(a) showed that at the lower stratum, initially the number of larvae was slightly increased from 4.75 to 13.25 larvae 2 WAT until 4 WAT, respectively. It reached the highest peak at 5 WAT with 24.25 larvae. Subsequently, the number decreased dramatically with small fluctuation between 6 and 7 WAT, and 10 and 11 WAT. When the larval distributions were investigated at different leaf strata, it was

found that the pattern of abundance was similar at all three strata for the first cropping. The population drastically increased to the highest peaks at 3 WAT, 4 WAT and 5 WAT for the upper, middle and lower strata, respectively. The population then decreased gradually and fluctuated slightly in the upper stratum. In the middle and lower strata of brinjal plant, the population decreased drastically to very low level towards the end of the cropping. Generally, more larvae were found in the lower leaf stratum compared with the middle and upper strata. In the second cropping, the distribution of larval population started from the higher levels for the middle and lower strata. Interestingly, more larvae preferred to infest the middle rather than the lower leaves in the first cropping. In the second cropping, the larval population of the middle stratum peaked at 2 WAT at a higher level of 38 larvae per two leaves as compared to the first cropping. However, the larval population was peaked at 3 WAT at the lower leaves with 18 larvae per two leaves.

There was a significant difference ($F = 3.284$; $df = 2,5$; $P < 0.05$) in the number of whitefly larvae among all the strata in the second cropping period. Whitefly larvae were not observed at the upper stratum of the brinjal plants. However, the number of larvae started to increase until 4 WAT and decreased before it reached the highest peak at 7 WAT. The population decreased until 11 WAT and increased again at the end of cropping period. In the middle stratum [Figures 2(b)], the population of whitefly started to increase in the early cropping period until 6 WAT and then fluctuated between 8 and 10 WAT. At the end of second cropping period, the number of whitefly larvae increased again. In the lower stratum, the number of whitefly larvae was found to increase in the early stage of second cropping period [Figure 2(b)]. Then, the population decreased slightly until 7 WAT and increased again at 9 WAT. There was a little fluctuation at the end of the cropping period. The number of whitefly larvae varied on the different strata of brinjal plants, ranging from 0.5 to 4.5 larvae in the upper stratum, 1.75 to 22 larvae in the middle stratum, and 2.75 to 24.25 larvae in the lower stratum. The number of whitefly larvae were higher ($p < 0.05$) in the lower and middle strata as compared with upper stratum.

The spatial distribution of *B. tabaci* larvae on brinjal plants in the first cropping period was found to be aggregated in all the strata except for the upper stratum (Taylor's Power Law, Figure 3). Results showed that, *B. tabaci* was uniformly distributed in the upper stratum ($b = 0.51969$) of the first cropping period. However, the dispersion pattern of *B. tabaci* in the middle and lower strata were found to be significantly different to the upper stratum, where both means showed aggregative pattern of distribution. The middle stratum showed a more obvious aggregated pattern due to the coefficient, b value being higher than lower stratum. The coefficient values in the first cropping were 1.39825 and 1.04518 for middle and bottom stratum, respectively (Figure 3). The aggregation of *B. tabaci* larvae in middle and lower strata were due to their large larval number on the under surface of leaves and their immobility. As compared with the upper stratum, possibly the number of larvae emerged on the young leaves was very low. In the second cropping, *B. tabaci* showed aggregated distribution with the b values being 1.311, 1.760, and 1.326 for upper, middle and lower strata (Figure 3), respectively. Generally, the populations of whitefly were aggregated for both seasons in all the strata except for the upper stratum in the first cropping.

<<Figure 2: Mean number of whitefly larvae on upper, middle and lower strata of brinjal plant per 2 leaves (a) First cropping (b) Second cropping>>

<<Figure 3: Spatial distribution of whitefly larvae on brinjal with Taylor's Power Law regressions (a) First cropping (b) Second cropping>>

The distribution of *B. tabaci* larvae varied among the three-leaf strata of the brinjal plants. This was mainly due to the behaviour of whitefly adults that laid their eggs on the underside of leaves especially on the upper stratum. McAuslane (1995) reported that the eggs were scattered all over the leaf either singly or in cluster. The immature stages lived on the underside of brinjal leaves. The first instar larvae moved some distance around the surface of leaves. They then became sessile and fed on the plant sap until they turned into pupae. Since the shoot of a brinjal plant was the preferential oviposition sites of *B. tabaci*, only adults and eggs were found on the upper stratum of the plant, followed by the first larval instar in the middle stratum, the last larval instar in the lower stratum and lastly the empty pupa on the oldest brinjal leaves. Thus, the major part of the larvae III and larvae IV were concentrated in lower stratum of the plant with leaves of a specific age. By restricting the sampling in this region, efficiency in sampling could be increased. The total number of whitefly larvae of all the strata in the first cropping was lower than that in the second cropping throughout the cropping period (Figure 3). This was because over the duration of the first cropping, the brinjal was planted in a new area. It took a while for the whiteflies to establish their population in this new area. However, in the second cropping, the population flourished immediately after transplanting the crop. The whitefly population from the first cropping immediately moved to the new hosts of brinjal plants in the second cropping which provided a good condition for their oviposition site, shelter and also food sources.

Generally, in both cropping periods, whitefly population started to increase and reached the highest peak during flowering and fruiting stages. The population decreased when the plants reached the end of economic life of four months for brinjal plants. These trends were due to the presence of host plants at initial stage of population growth, and then they reached the highest peak mainly due to availability of food and good shelter. Subsequently, the population started to decrease due to sudden reduction of food and the older leaves became unsuitable for immature whiteflies. In the first cropping, the

population decreased drastically after 5 WAT as compared to the second cropping. There were many factors affecting the population of whitefly such as climate (temperature, rainfall and relative humidity), natural enemies, surrounding area, and host plants. Horowitz et al. (1984) found that the population dynamic of *B. tabaci* in cotton fields were mainly due to climatic factors such as humidity and temperature, but parasitism was not a decisive factor. According to Gerling et al. (1986), an extreme relative humidity of both high and low conditions were unfavourable for the survival of immature stages of whitefly. In the second cropping, the immature whiteflies successfully maintained their population until the end of cropping period probably because they could adapt the surrounding condition. Previous study Gerling et al. (1986) found that if host plants were cultivated continuously in time and space, the population of *B. tabaci* would increase and cause greater damage to host plants grown later in the planting season. In fact, outbreaks of *B. tabaci* in Brazil occurred under such circumstances (Kogan and Turnipseed, 1987). The distribution of immature whiteflies varied significantly among the leaf strata. The upper stratum harboured lower number of whitefly larvae. This part of brinjal plant consisted of young leaves, a preferred place for egg oviposition (Butler et al., 1983). Since only the larvae and pupae were counted in this study, the upper stratum consequently had lower number of whitefly larvae. The population on the upper stratum was highly exposed to the rain which might have a direct effect on the whitefly larvae, which could be brushed off from the under surfaces of the leaves. Furthermore, first and second nymphal stages of whitefly had higher mortality rate (Horowitz et al., 1984). A similar situation was also observed in another study carried out by Hassan (1996). He reported that, each species of whitefly in different agricultural ecosystems could exhibit different patterns of spatial distribution.

The distribution of the whitefly larvae was strongly dependent on the leaf strata during different cropping as the interaction between strata and cropping was different and highly significant. The population pattern of *B. tabaci* changed in the middle stratum. This indicates that the sampling of *B. tabaci* could be represented at the middle part of brinjal plants. According to Von Arx et al. (1984) in Sudan, about 50% of the whitefly population lived on the main stem of cotton leaves (middle stratum) during the period of maximum population density. Ekbom and Xu, (1990) noted that the distribution of *B. tabaci* on plants was far from random, since the insects tend to select both particular plants and parts of the plant. For the sampling of immature stages, it has been difficult to estimate accurately the population density of *B. tabaci*. Population dynamics has traditionally sought to explain spatial distribution, one of the most important characteristics and ecological properties of insect species as the by-product of environmental heterogeneity and reproductive population growth act on random process of movement and mortality (Taylor, 1984). He also noted that field sampling was not complete and cannot be sustained without understanding the underlying spatial distribution. The earlier studies on the distribution of *B. tabaci* larvae on brinjal plants showed that the population of *B. tabaci* were aggregated on all plant strata except upper stratum. In the first cropping, whitefly larvae population was low at the upper stratum and was not aggregated. This could possibly be due to the field experiment located in a new area with very low whitefly infestation. However, in the second cropping the number of whitefly larvae was increased and an aggregated pattern was observed. The upper stratum, which mainly consisted of young leaves, provided a favourable place for whitefly adults to lay eggs confirming similar finding with the previous study carried out by McAuslane (1995). The main reason was that, the larvae emerged a week after the eggs have been oviposited on the under surfaces of the leaves, resulting in low number of *B. tabaci* larvae found in the upper stratum. When the new leaves had appeared, the secondary leaves reached their maturity and the level or stratum changed to the middle stratum. Furthermore, larval and pupal stages had longer life cycle, which was about 23.6 days at 25°C (Coudriet et al., 1985). Hence the larvae and pupae were mainly found at the middle stratum. Otherwise, the number of larvae and pupae were found to be lower at the lower stratum because most of the pupae had been emerged to adults. The physiological growth and development of brinjal plants took a longer duration the middle stratum than upper and lower strata. Some of the factors might influence in the spatial distribution probably due to cultural practices such as pruning the old leaves at the lower stratum which resulted in the number of larvae to be smaller in the lower stratum.

5. Conclusion

This study concludes that the total number of whitefly larvae pests was found to be most abundant at the middle stratum of the brinjal plants. The quantity of food source is one of the major factors that have attracted whitefly in the area. In fact, its flight ability enables them to search food quickly. Thus, this could encourage whitefly to reproduce in great numbers and subsequently cause severe infestation in the fields. There would be more serious damage to the field planted later in the next cropping, if the host plants were cultivated continuously in time and space of a same area.

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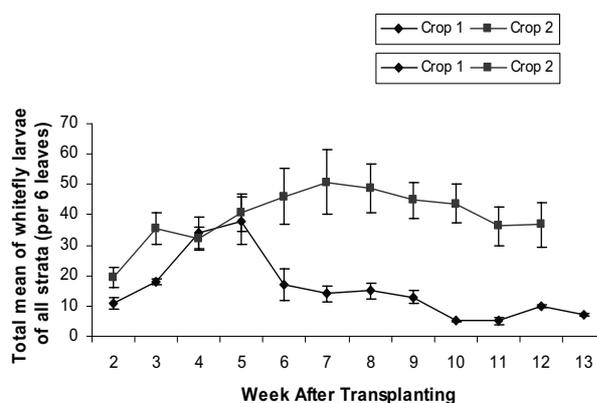


Figure 1. Distribution and abundance of whitefly larvae on each brinjal plant (total mean larvae in three leaf strata) in two cropping periods.

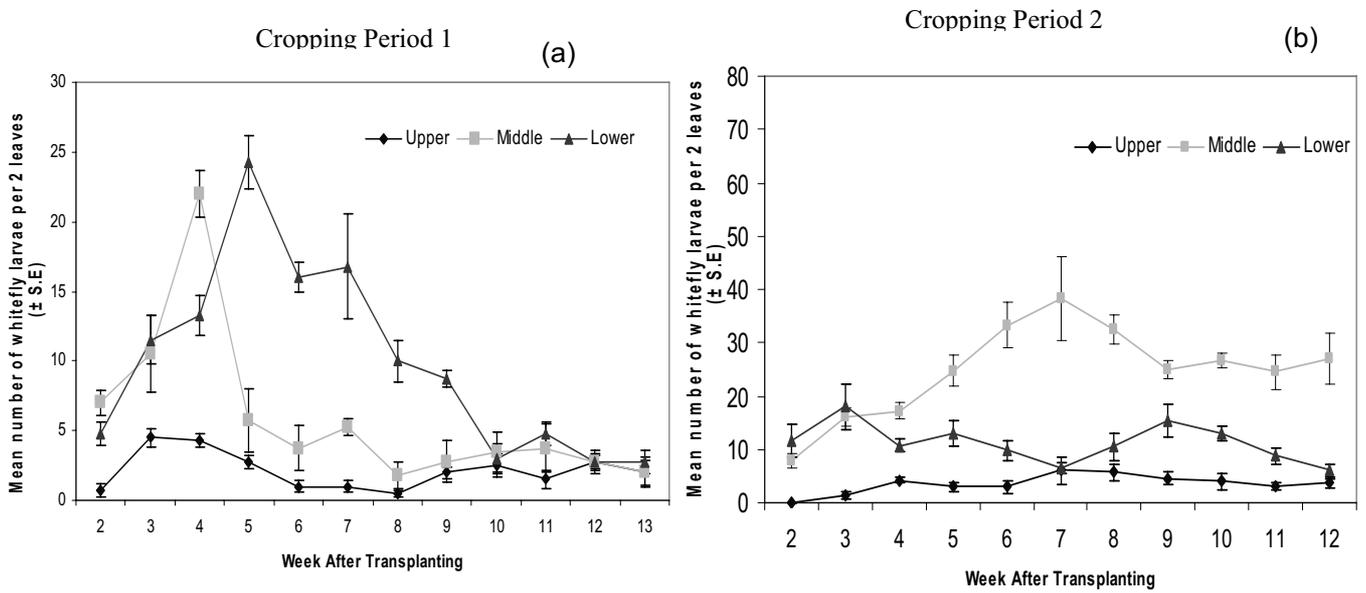


Figure 2. Mean number of whitefly larvae on upper, middle and lower strata of brinjal plant per 2 leaves
(a) First cropping (b) Second cropping.

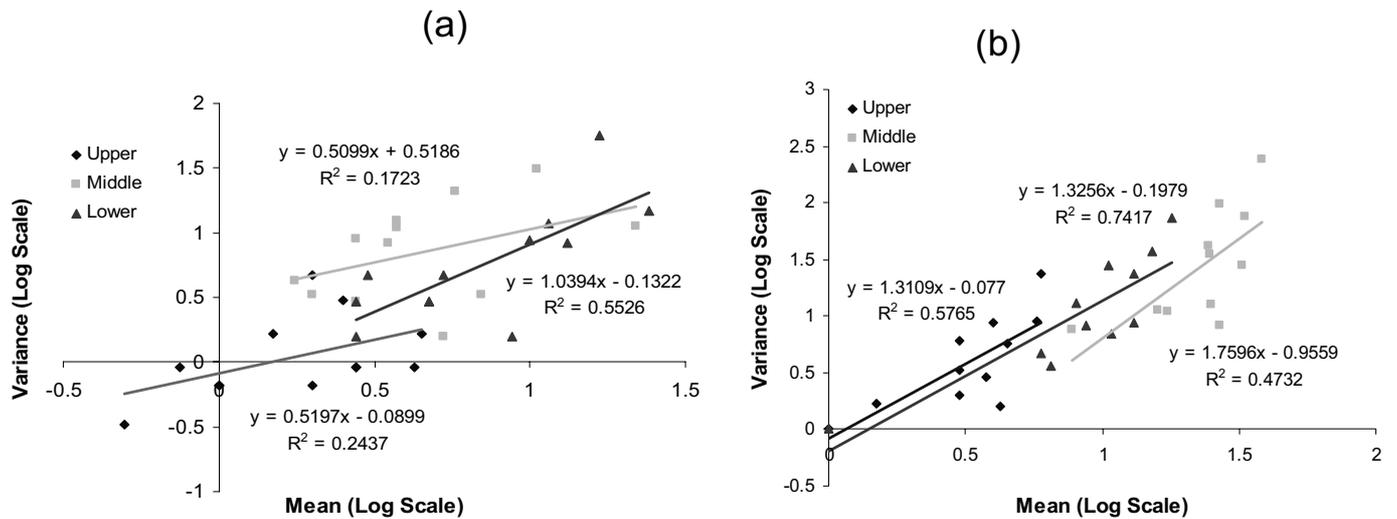


Figure 3. Spatial distribution of whitefly larvae on brinjal with Taylor's Power Law regressions
(a) First cropping (b) Second cropping



Automation and Emerging Technology Development of 2d Seed Sowing Robo

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Abstract

In the current scenario most of the countries do not have sufficient skilled manpower specifically in agricultural sector and it affects the growth of developing countries. So it's a time to automate the sector to overcome this problem. An innovative idea of our project is to automate the process of sowing crops such as sunflower, baby corn, groundnut and vegetables like beans, lady's finger, pumpkin and pulses like black gram, green gram etc to reduce the human effort and increase the yield. The plantations of seeds are automatically done by using DC motor. The distance between the two seeds are controlled and varied by using Microcontroller. It is also possible to cultivate different kinds of seeds with different distance. When the Robo reaches the end of the field we can change the direction with the help of remote switches. The whole process is controlled by Microcontroller.

Keywords: Agricultural Robo, Automation, DC motor and Microcontroller

1. Objective

Automation brings comfort to our life. Automation in its pure sense defines any activity that minimizes human factor to increase productivity at consistent quality. The main fact is that how fast the process is being completed. This project "An Autonomous Agricultural Seed Sowing Robo" aims in fulfilling the lack of man power and automate the process of seed sowing with low cost.

2. Need

Low Cost Automation (LCA), the buzzword in all industrial firms generally involves pneumatic, electrical as well as electronic components. LCA is important in the automation of factories, for example, the electronic component assembly plants. Automation saves a lot of tedious manual work and speeds up the production processes.

Now days we have a problem on lack of man power. So agricultural field we spent more money for both planting especially seed sowing. It consumes more time and also increase the cost with low accuracy. So it is a time to automate the process of sowing.

3. Block Diagram

The block diagram shown in Fig. 1, which consists of selection buttons for choosing the distance between the seeds, Microcontroller unit, power supply unit and mechanical setup (Robo). The power supply unit consists of step down transformer, rectifier circuit, filter and regulator. The robo consists of motors to drive the robo and stepper motor to

control the flow of seeds and DC motor to sow the seeds in the field. The following figure shows the block diagram of our project.

4. Description of Components

4.1. DC Motor

An electric motor is a machine which converts electrical energy to mechanical energy. Its action is based on the principle that when a current-carrying conductor is placed in a magnetic field, it experiences a magnetic force whose direction is given by Fleming's left hand rule. When a motor is in operation, it develops torque. This torque can produce mechanical rotation. DC motors are also like generators classified into shunt wound or series wound or compound wound motors.

4.2 Spur Gear

Gears are used to transmit the power from one shaft to another shaft or between a shaft and a slide. This is accomplished by successively engaging teeth. Spur gears have straight teeth parallel to the axis and thus, subjected to axial thrust due to the tooth load.

4.3 Stepper Motor

The operation of a stepper motor requires the presence of the following elements:

- ✓ A control unit (a micro-processor for example) which supplies impulses the frequency of which is proportional to the speed of the motor. This applies equally to both directions of rotation;
- ✓ A sequencer which will direct the impulses to the various motor coils.
- ✓ A power supply.

<Figure 1>

4.4 Gear Motor

<Figure2>

4.5 Opto Isolator

An opto-isolator (shown in Fig 3) is a device that uses a short optical transmission path to transfer a signal between elements of a circuit, typically a transmitter and a receiver, while keeping them electrically isolated — since the signal goes from an electrical signal to an optical signal back to an electrical signal, electrical contact along the path is broken.

<Figure 3>

A common implementation involves a LED and a phototransistor, separated so that light may travel across a barrier but electrical current may not. When an electrical signal is applied to the input of the opto-isolator, its LED lights, its light sensor then activates, and a corresponding electrical signal is generated at the output. Unlike a transformer, the opto-isolator allows for DC coupling and generally provides significant protection from serious over voltage conditions in one circuit affecting the other.

4.6 Power Supply Unit

The Power supply unit shown in Fig 4, which consists of filters, rectifiers and voltage regulators. Starting with an AC voltage, a steady DC voltage, is obtained by rectifying the ac voltage then filtering to a dc level and Finally Regulation is usually obtained from an IC voltage regulator unit, which takes a dc voltage and provides a somewhat lower dc voltage, which remains the same even if the input dc voltage varies or the output load connected to the dc voltage changes.

<Figure 4>

4.6.1 Block Diagram For Power Supply

A block diagram containing the parts of a typical power supply and the voltage point in the unit is shown below in the diagram 5.

<Figure 5>

The ac voltage, typically 230v is connected to transformer, which steps the ac voltage down to the level for desired dc output. A diode rectifier provides a full wave rectified Voltage that is initially filtered by a simple capacitive filter to produce a dc voltage. This resulting dc voltage usually has some ripple or ac voltage variation. A regulator Circuit can use this dc input to provide a regulated that not only has much ripple voltage. But also remain the same dc values even if the input dc voltage changes. This voltage Regulation is usually obtained using one of a number of popular voltage regulation IC Units.

4.6.2 Transformer

A transformer is the static device of which electric power in one circuit is transformed into electric power of the same frequency in another circuit. It can rise or lower the voltage in a circuit but with a corresponding decrease or increase in current. It works with the principles of mutual induction. In our project we are using step down transformer for providing that necessary supply for the electronic circuits.

4.6.3 Rectifier

The full wave rectifier conducts during both positive and negative half cycles of input AC input; two diodes are used in this circuit. The ac voltage is applied through a suitable power transformer with proper turn's ratio. For the proper operation of the circuit, a center-tap on the secondary winding of the transformer is essential. During the positive half cycle of ac input voltage, the diode D1 will be forward biased and hence will conduct; while diode D2 will be reverse biased and will act as open circuit and will not conduct. In the next half cycle of ac voltage, polarity reverses and the diode D2 conducts, being forward biased, while D1 does not, being reverse biased. Hence the load current flows in both half cycles of ac voltage and in the same direction. The diode we are using here for the purpose of rectification is IN4001.

4.6.4 Filter

The filter circuit used here is the capacitor filter circuit where a capacitor is connected at the rectifier output, and a DC is obtained across it. The filtered waveform is essentially a DC voltage with negligible ripples, which is ultimately fed to the load.

4.6.5 Regulator

The output voltage from capacitor is more filtered and finally regulated. The voltage regulator is a device, which maintains the output voltage constant irrespective of the change in supply variations, load variations and temperature changes. Hence IC7805 is used which is a +5v regulator.

4.7 Relays

A relay is an electrically operated switch. Current flowing through the coil of the relay creates a magnetic field which attracts a lever and changes the switch contacts. The coil current can be on or off so relays have two switch positions and they are double throw (changeover) switches. Relays allow one circuit to switch a second circuit which can be completely separate from the first. For example a low voltage battery circuit can use a relay to switch a 230V AC mains circuit. There is no electrical connection inside the relay between the two circuits; the link is magnetic and mechanical.

4.8 PIC Microcontroller

The microcontroller that has been used for this project is from PIC series. PIC microcontroller is the first RISC based microcontroller fabricated in CMOS (complementary metal oxide semiconductor) that uses separate bus for instruction and data allowing simultaneous access of program and data memory.

The main advantage of CMOS and RISC combination is low power consumption resulting in a very small chip size with a small pin count. The main advantage of CMOS is that it has immunity to noise than other fabrication techniques.

4.8.1 PIC (16F877)

<Figure 6>

The above Fig.6 shows the architecture of PIC Micro controller 16F877. Various microcontrollers offer different kinds of memories. EEPROM, EPROM, FLASH etc. are some of the memories of which FLASH is the most recently developed. Technology that is used in pic16F877 is flash technology, so that data is retained even when the power is switched off. Easy Programming and Erasing are other features of PIC 16F877.

4.8.2 Special Features of PIC Micro controller

The Core Features are

- High-performance RISC CPU
- Only 35 single word instructions to learn
- All single cycle instructions except for program branches which are two cycle
- Operating speed: DC - 20 MHz clock input

DC - 200 ns instruction cycle

- Up to 8K x 14 words of Flash Program Memory,
Up to 368 x 8 bytes of Data Memory (RAM)
Up to 256 x 8 bytes of EEPROM data memory

- Power-up Timer (PWRT) and Oscillator Start-up Timer (OST)
- Watchdog Timer (WDT) with its own on-chip RC Oscillator for reliable operation
- Programmable code-protection
- Power saving SLEEP mode
- Selectable oscillator options
- Low-power, high-speed CMOS EPROM/EEPROM technology
- Fully static design
- In-Circuit Serial Programming (ICSP) via two pins
- Only single 5V source needed for programming capability
- In-Circuit Debugging via two pins
- Processor read/write access to program memory
- Wide operating voltage range: 2.5V to 5.5V
- High Sink/Source Current: 25 mA
- Commercial and Industrial temperature ranges
- Low-power consumption:
 - < 2mA typical @ 5V, 4 MHz
 - 20mA typical @ 3V, 32 kHz
 - < 1mA typical standby current

The Peripheral Features are

- Timer0: 8-bit timer/counter with 8-bit pre scalar
- Timer1: 16-bit timer/counter with prescaler, can be incremented during sleep via external crystal/clock
- Timer2: 8-bit timer/counter with 8-bit period register, prescaler and postscaler
 - 10-bit multi-channel Analog-to-Digital converter
- Synchronous Serial Port (SSP) with SPI. (Master Mode) and I2C. (Master/Slave)
- Universal Synchronous Asynchronous Receiver Transmitter (USART/SCI) with 9- bit address detection.

5. Working

<Figure 7>

This part explains how the actual process is being done. The working of the project is explained below as follows: At The First Stage we should fill the seeds inside the container. Then select the button for distance between the seeds. When the power supply is given to the robo its start to move in the field. The time taken to reach the distance is feed into the microcontroller when it reaches the distance it will stop the robo by OFF the geared motor with the use of relay. Then the stepper motor is activated to control the flow of seeds which is kept inside the container after the flow of seed it will stopped by using relay. Finally the DC motor is activated to sow the seeds inside the field at the depth of 1 to 1.5 inches. Then the DC motor is stopped and Geared motor is activated and the process is repeated. The front part of the robo has only one wheel to easily change the direction. The change in direction is controlled by geared motor with the assistance of remote button. When the robo reaches the end if the field we can change the direction by using remote switches present in the robo. All the operation is controlled using Microcontroller.

5.1 Micro controller Program

```
#include <pic.h>
##include "usart.h"
#include <stdlib.h>
##define seed1delval 5
#define seed2delval 5
#define sowdelval 1500
#define countval 10 //8
```

```

#define changestepdelval 5000
#define rightturndelval 15000
#define rightdelval 5000
#define stepdelayval 18000
#define movfor ((reverse = 0),(forward = 1),(enable_st = 1))
#define movrev ((forward = 0),(reverse = 1),(enable_st = 1))
#define stop (reverse = 0),(forward =0)
// robot pin connections
// #define l_opto (!l_opto1)
// #define c_opto (!c_opto1)
// #define r_opto (!r_opto1)

static volatile bit l_opto @ (unsigned)&PORTB*8+0;
static volatile bit c_opto @ (unsigned)&PORTB*8+1;
static volatile bit r_opto @ (unsigned)&PORTB*8+2;
static volatile bit optoformovement @ (unsigned)&PORTB*8+5;
static volatile bit rswitch @ (unsigned)&PORTE*8+0; // extra switches to turn the front motor manually to left and
right
static volatile bit lswitch @ (unsigned)&PORTE*8+1;
static volatile bit forward @ (unsigned)&PORTA*8+2;
static volatile bit reverse @ (unsigned)&PORTA*8+4;
static volatile bit right @ (unsigned)&PORTA*8+1;
static volatile bit left @ (unsigned)&PORTA*8+0;
static volatile bit enable_st @ (unsigned)&PORTE*8+1;
static volatile bit enable_tr @ (unsigned)&PORTE*8+2;

static volatile bit led1 @ (unsigned)&PORTC*8+3;
static volatile bit led2 @ (unsigned)&PORTC*8+2;
static volatile bit led3 @ (unsigned)&PORTC*8+4;
static volatile bit led4 @ (unsigned)&PORTC*8+5;
static volatile bit buzzer @ (unsigned)&PORTC*8+0;
static volatile bit startswitch @ (unsigned)&PORTA*8+3;
static volatile bit stopswitch @ (unsigned)&PORTE*8+2;
static volatile bit r_switch @ (unsigned)&PORTE*8+0;
static volatile bit l_switch @ (unsigned)&PORTE*8+1;
static volatile bit sowswitch @ (unsigned)&PORTB*8+6;
static volatile bit mode1 @ (unsigned)&PORTB*8+3;
static volatile bit mode2 @ (unsigned)&PORTB*8+4;
static volatile bit mode3 @ (unsigned)&PORTB*8+5;
static volatile bit sowmotor @ (unsigned)&PORTC*8+1;
static volatile unsigned char stepper @ &PORTD;
static bit countcamebit, rbit, lbit;
static unsigned char rotcount;
unsigned char dummy,receiveddata;

```

```
void msdelay(unsigned int);
void mswait (void);
void stepdelay (void);
void stepwait (void);
void rotateclk(void);
void rotateantclk(void);
void turnright (void);
void turnleft (void);
void operation_move (void);
void sowseed1 (void);
void sowseed2 (void);
```

```
unsigned char noofrot;
unsigned char i;
static bit switchbit;
unsigned char seed1delval;
```

```
void main()
{
// robo pins initialisation
    ADCON1 = 6;
    TRISA0 = 0;
    TRISA1 = 0;
    TRISA2 = 0;
    TRISA3 = 1;
    TRISA4 = 1;
    TRISA5 = 1;

    TRISE0 = 1;
    TRISE1 = 1;
    TRISE2 = 1;

    TRISB = 255;

    TRISD0 = 0;
    TRISD1 = 0;
    TRISD2 = 0;
    TRISD3 = 0;
    TRISD4 = 0;
    TRISD5 = 0;
    TRISD6 = 0;
    TRISD7 = 0;
```

```
TRISC0 = 0;
TRISC1 = 0;
TRISC2 = 0;
TRISC3 = 0;
TRISC4 = 0;
TRISC5 = 0;
// TRISC3 = 0;
TRISC7 = 1;

forward = 0;
reverse = 0;
right = 0;
left = 0;
enable_st = 0;
enable_tr = 0;

stepper = 0;

PORTC = 0;
led1 = 0;
led2 = 0;
buzzer = 0;
countcamebit = 0;

// robo initialisation ends

INTCON = 0;
RBPU = 0;
INTEDG = 0;

// usartinit(9600);
// usarttx('H');

seed1delval = 5;

rbit = 0;
lbit = 0;
led1 = 1;
led2 = 0;
msdelay(1000);
led1 = 0;
led2 = 1;
msdelay(1000);
PORTC = 0;
```

```
led2 = 0;

while(!c_opto)
{
  buzzer = 1;
  if(!rswitch)
  {
    if(!r_opto)
    {
      left = 0;
      right = 1;
      enable_tr = 1;
    }
    else
    {
      left = 0;
      right = 0;
      enable_tr = 0;
    }
  }
  else if (!lswitch)
  {
    if(!l_opto)
    {
      right = 0;
      left = 1;
      enable_tr = 1;
    }
    else
    {
      left = 0;
      right = 0;
      enable_tr = 0;
    }
  }
  else
  {
    left = 0;
    right = 0;
    enable_tr = 0;
  }
}
buzzer = 0;
// led1 = 1;
```

```
msdelay(2000);
led1 = 0;
led2 = 0;
led3 = 0;
while(startswitch)
{
if(!mode1)
{
led1 =1;
led2 = 0;
led3 = 0;
seed1delval = 2;
}
else if(!mode2)
{
led1 =0;
led2 = 1;
led3 = 0;
seed1delval = 5;
}
else if(!mode3)
{
led1 =0;
led2 = 0;
led3 = 1;
seed1delval = 7;
}
}

while(startswitch);
// led1 = 1;
msdelay(200);
while(!startswitch);
movfor;
while(1)
{
// while(!RCIF);
// receiveddata= RCREG;
while(r_switch && l_switch &&stopswitch)
{
for(i=0; i< seed1delval; i++)
{
msdelay(1000);
if(!r_switch || !l_switch || !stopswitch)
```

```

        {
            switchbit = 1;
            break;
        }
    }
    if(switchbit)
    {
        break;
    }
    else
    {
        stop;
        sowseed1();
        movfor;
        switchbit = 0;
        for(i=0; i< seed1delval; i++)
        {
            msdelay(1000);
            if(!r_switch || !l_switch || !stopswitch)
            {
                switchbit = 1;
                break;
            }
        }
        if(switchbit = 1)
            break;
        else
        {
            stop;
            sowseed2();
            movfor;
        }
    }

}

if(switchbit)
{
    switchbit = 0;
    if(!r_switch)
    {

        // stop;
        led2 = 1;

```

```
        turnright();
        led2 = 0;
    }
else if(!l_switch)
    {
    // stop;
    led3= 1;
        turnleft();
        led3 = 0;
    }
else if(!stopswitch)
    {
        led1 = 0;
        stop;
        msdelay(200);
        while(!stopswitch);
        msdelay(200);
        while(startswitch);
        led1 = 1;
        msdelay(200);
        while(!startswitch);
        movfor;
    }
}

// if(!r_switch)
// {
//         receiveddata= 'R';
//         operation_move();
//         msdelay(200);
//         while(!r_switch);
//         msdelay(200);
//     }
// else if(!l_switch)
//     {
//         receiveddata= 'L';
//         operation_move();
//         msdelay(200);
//         while(!l_switch);
//         msdelay(200);
//     }
}
```

```
}
```

```
void sowseed1 (void)
```

```
{
```

```
    noofrot = 4;
```

```
    rotateclk();
```

```
    msdelay(2000);
```

```
//    noofrot = 1;
```

```
//    rotateclk();
```

```
    sowmotor = 1;
```

```
    msdelay(sowdelval);
```

```
    sowmotor = 0;
```

```
    msdelay(1000);
```

```
    noofrot = 4;
```

```
    rotateantclk();
```

```
}
```

```
void sowseed2 (void)
```

```
{
```

```
    noofrot = 4;
```

```
    rotateantclk();
```

```
    msdelay(2000);
```

```
//    noofrot = 1;
```

```
//    rotateclk();
```

```
    sowmotor = 1;
```

```
    msdelay(sowdelval);
```

```
    sowmotor = 0;
```

```
    msdelay(1000);
```

```
    noofrot = 4;
```

```
    rotateclk();
```

```
}
```

```
void turnright (void)
```

```
{
```

```
    movfor;
```

```
    receiveddata = 'R';
```

```
    operation_move();
```

```
    msdelay(rightturndelval);
```

```
    receiveddata = 'S';
```

```
    operation_move();
```

```
    movfor;
```

```
    msdelay(rightdelval);
```

```
    receiveddata = 'R';
```

```
    operation_move();
```

```
    msdelay(rightturndelval);
    receiveddata = 'S';
    operation_move();
    movfor;
}
void turnleft (void)
{
    movfor;
    receiveddata = 'L';
    operation_move();
    msdelay(rightturndelval);
    receiveddata = 'S';
    operation_move();
    movfor;
    msdelay(rightdelval);
    receiveddata = 'L';
    operation_move();
    msdelay(rightturndelval);
    receiveddata = 'S';
    operation_move();
    movfor;
}
void operation_move(void)
{
    switch (receiveddata)
    {
        case 'F':
            movfor;
            break;

        case 'B':
            movrev;
            break;

        case 'R':
            rbit = 1;
            while(!r_opto)
            {
                left = 0;
                right = 1;
                enable_tr = 1;
                // if(RCIF)
                // {
                /// receiveddata = RCREG;
```

```
//      continue;
//      }
    }
    left = 0;
    right = 0;
    enable_tr = 0;
    rbit = 1;
    lbit = 0;
    break;
case 'L':
    lbit = 1;
    while(!l_opto)
    {
        right = 0;
        left = 1;
        enable_tr = 1;
//      if(RCIF)
//      {
//          receiveddata = RCREG;
//          continue;
//      }
    }
    left = 0;
    right = 0;
    enable_tr = 0;
    lbit = 1;
    rbit = 0;
    break;
case 'S':
    if (rbit)
    {
        while(!c_opto)
        {
            right = 0;
            left = 1;
            enable_tr = 1;
        }
        left = 0;
        right = 0;
        enable_tr = 0;
        msdelay(400);
        rbit = 0;
    }
    if(lbit)
```

```
        {
        while(!c_opto)
            {
            left = 0;
            right = 1;
            enable_tr = 1;
            }
            left = 0;
            right = 0;
            enable_tr = 0;
            msdelay(400);
            lbit = 0;
            }
        stop;
        break;

    default:
        break;
    }
}
void interrupt intl()
{
    if (INTF)
        {
        rotcount++;
        countcamebit = 1;
        INTF = 0;
        }
}
void rotateantclk(void)
{
    unsigned int i;
    for (i=1; i<=noofrot; i++)
        {
        stepper = 0x10;
        stepdelay();
        stepper = 0x20;
        stepdelay();
        stepper = 0x40;
        stepdelay();
        stepper = 0x80;
        stepdelay();
        }
}
```

```
void rotateclk(void)
{
  unsigned int i;
    for (i=1; i<=noofrot; i++)
      {
        // stepper = 0x0ff;
        // while(1);
        stepper = 0x80;
        stepdelay();
        stepper = 0x40;
        stepdelay();
        stepper = 0x20;
        stepdelay();
        stepper = 0x10;
        stepdelay();
      }
}
```

```
void stepdelay (void)
{
  static unsigned int i;
  for (i = 1; i<=stepdelayval; i++)
    stepwait ();
}
```

```
void stepwait (void)
{
;
}
```

```
void msdelay (unsigned int msdel)
{
  unsigned int i;
  for (i = 1; i<=msdel; i++)
    mswait ();
  i += 2000;    // a dummy instruction for correct time delay;
  i +=20;
}
```

```
void mswait (void)
{
  unsigned int j;
```

```

for (j= 1; j<=414; j++);
}
__CONFIG(HS & WDTDIS & PWRTEN & BOREN & LVPDIS & DUNPROT & WRTEN & DEBUGDIS &
UNPROTECT);

```

6. Conclusion

This project work “An Autonomous Agricultural Seed sowing Robo” has been done on the aim of solving the problem faced by many restaurants where we have taken survey. Thus the project work has been successfully completed.

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www.picallw.com

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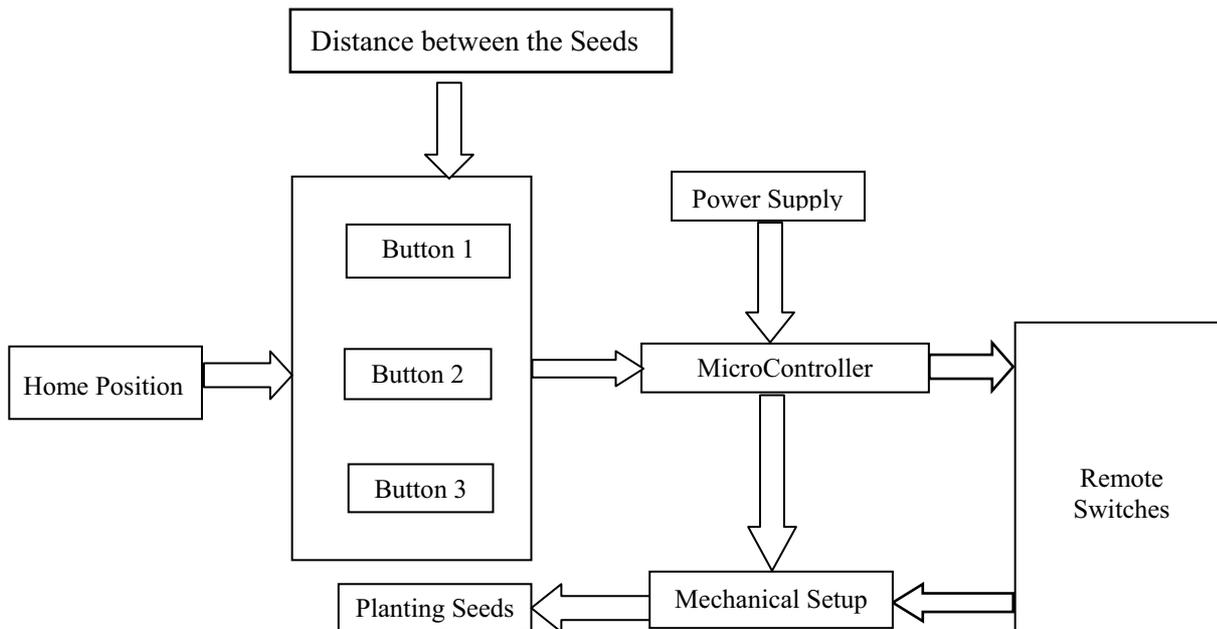
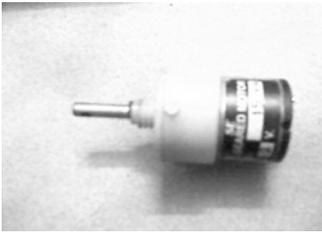


Figure 1. Block Diagram



Specifications:

Type - DC Geared Motor

Voltage - 5V

Current - 110mA

Rpm - 150 RPM

Figure 2. Gear Motor

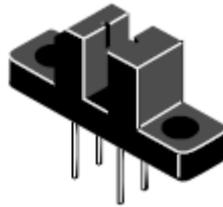


Figure 3. Opto Isolator

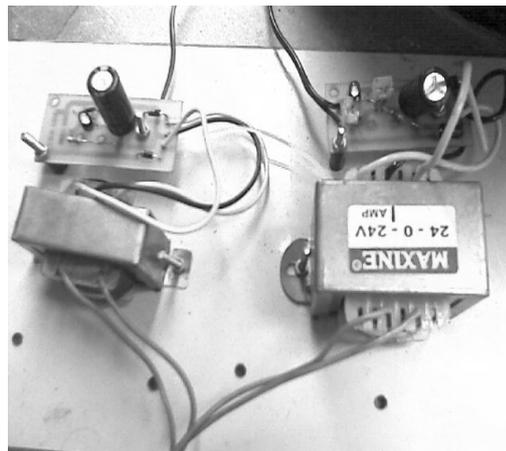


Figure 4. Power Supply

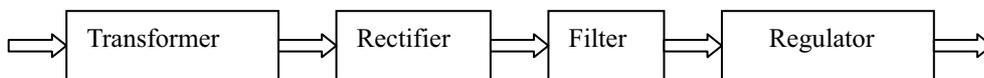
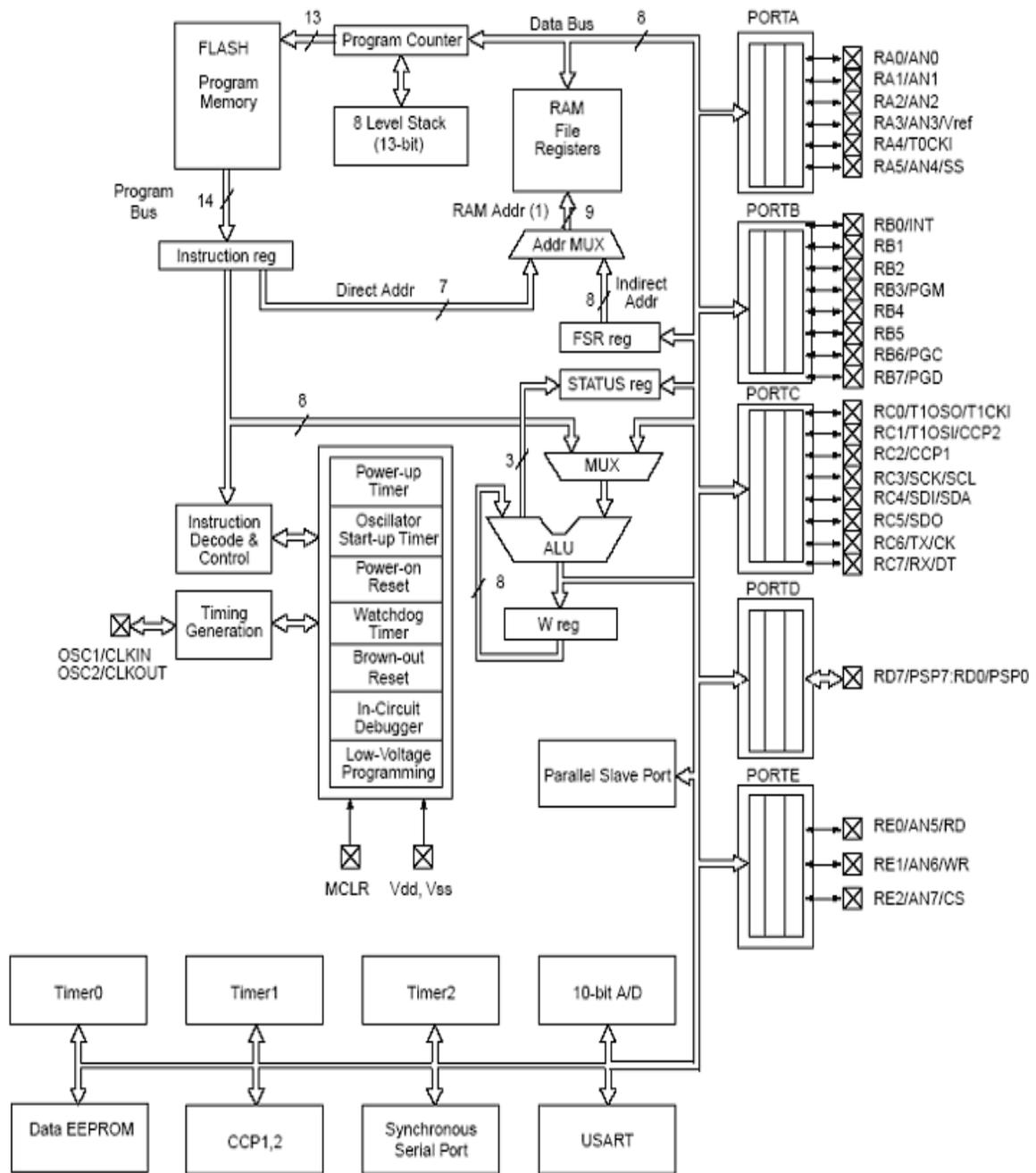


Figure 5. General Block Diagram



Note 1: Higher order bits are from the STATUS register.

Figure 6. Architecture of PIC Micro controller

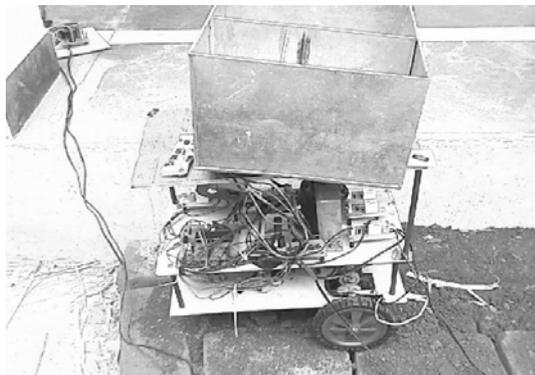


Figure 7. Photographic view of the Experimental Setup



Genetic Analysis of *Scleropages formosus* Golden Asian Arowana Using Microsatellite DNA

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The research is financed by The People's Republic of China Ministry of Agriculture Affairs State(948), Science , Science Research Institute fund of scientific research operational fee technical Program from Chinese Academy of Fishery Sciences Research Institute No. 2130108, Technology fund of Pearl River Fisheries Research Institute, Technology fund of Pearl River Fisheries Research Institute No.2003-3-5, Science and Technology Developing Fisheries Program of liwan district Guangdong Province.No. 20082109029

Abstract

Asian arowana (*Scleropages formosus*) is a highly endangered fish species listed in Appendix 1 of CITES since 1980. In order to explore the population genetic structure of golden Asian arowanas *Scleropages formosus* from the breeding population, of 26 fish sampled were genotyped at twenty microsatellite loci. The average allele number of 14 polymorphic microsatellites was 2.94 loci. The average observed heterozygosity was 0.446 ranging from 0.143 to 1.00, and the gene diversity was quite high (0.78). All these data suggest that middle level of genetic diversity existed in the golden Asian arowana population. Data showed the population somewhat departed from HWE, such as excessive and deficient heterozygote numbers. AMOVA analysis offered evidence of a weak genetic differentiation with 0.51% variation between samples.

Keywords: Asian arowana, Genetic diversity, Osteoglossidae

1. Introduction

Asian Arowana (*Scleropages formosus*), commonly known as the Dragonfish, belong to an ancient family of fishes, the Osteoglossidae, which literally means bony-tongue. Several types of *S. formosus* with different color patterns inhabit separate regions of Southeast Asia (Borneo, Sumatra, and Indochina) that were probably connected through freshwater habitats during the Pleistocene glacial ages (Goh, 1999). It is acquired a special status in Japan and some East Asian countries as a very popular but extremely expensive aquarium fish. Due to its popularity and great demand, Asian Arowanas have been fiercely hunted in its native habitat for profits, leading to its inclusion among species threatened with extinction of the population of these fish in the wild listed in CITES appendix I since 1980 (Dawes, 1999). However there has been not any systematic work or genetic assessments about Asian arowana population.

Due to their exceptional variability and relative ease of scoring, microsatellites are now generally considered the most powerful genetic markers. Microsatellites, or simple sequence repeats (SSR), have many attributes making them excellent for scientific studies, such as abundant polymorphisms, codominant heredity and easy detection. These features provide the foundation for their successful application in a wide range of fundamental and applied fields of biology and medicine, including forensics, molecular epidemiology, parasitology, population and conservation genetics, genetic mapping and genetic dissection of complex traits (Chistiakov, 2006; Waldbieser, 2001; Triantafyllidist, 2002). In this study, the highly variable microsatellites of green Asian arowana provide a perspective on the diploid nuclear structure of golden arowana and provide reference for further genetic study of this species.

2. Material and methods

2.1 Samples and DNA isolation

Fin clips of twenty-sixth breeding adult (>six years) golden Asian arowanas samples from Guang zhou tiny-lake aquatic organism technology co., ltd housed which were originated from Malaysia collected in 2001, were collected, and kept in absolute ethanol. Genomic DNA was extracted from a golden Asian arowana individual using a simple and cost-effective method (Yue, 2001). DNA was suspended to be used as the template DNA in polymerase chain reactions.

2.2 PCR amplification

Twenty microsatellite primers described (Yue, 1999) were used to amplify the genomic DNA of golden Asian arowana. Details of all microsatellite loci and PCR condition are given (Note 1). PCR amplification for microsatellites was performed on a MWG-BIOTECH thermal cycler in a total volume of 20 µl. Reactions contained 10×PCR buffer 2.0 µl, MgCl₂ (25 mmol/L) 1.0 µl, dNTPs (10 mmol/L each) 0.4 µl, each primer (10 µmol/L) 1.0 µl, *Taq* DNA polymerase 0.15 U, and DNA template 50 ng. Amplification was carried out using 4 min of initial denaturation followed by 30 cycles of 30s of denaturation at 94°C, 30s annealing at the temperature detailed in table 1 and 30s extension at 72°C with a final extension period of 7 min at 72°C. PCR products were run on a 4% (w/v) 1×TBE horizontal metaphor agarose gel at 250 V for 2 hours.

2.3 Data analysis

The bands of electrophoretic patterns were analyzed using Alpha Ease FC soft and artificial adjustment. The number of alleles, effective number of alleles, observed heterozygosity (*Ho*), expected heterozygosity (*He*) was computed from the microsatellite genotype data using Pop-gene (version 3.2) software. The Polymorphism Information Content (*PIC*) was calculated with the formula as follow (Botstein, 1980):

$$PIC = 1 - \sum_{i=1}^n p_i^2 - \sum_{i=1}^{n-1} \sum_{j=i+1}^n 2p_i^2 p_j^2$$

where n was the number of alleles at one locus; *P_i* and *P_j* were the frequencies of the *i*th and *j*th alleles at one locus, *j*=*i*+1. Departure from Hardy-Weinberg equilibrium (heterozygote deficiency or excess) was calculated for each locus

and each population with GENEPOP v.4.0 (Rousset, 2008) according to Weir and Cockerham's FIS (Weir, 1984), and was tested using the Markov chain method (10000 dememorization steps, 1000 batches, and 5000 iterations) to obtain unbiased estimates of the exact p value. Arlequin software version 3.11 was used to do analysis of molecular variance (AMOVA) (Excoffier, 1992).

3. Results

A total of 20 novel microsatellites were characterized using 26 golden Asian arowana individuals. 17 primer pairs (85%) produced clear and stable bands from the fish after optimizing the PCR conditions, 16 (80%) were polymorphic. All others were abandoned as they were unable to produce specific, clear, stable or polymorphic bands (Note 3). For instance, SSRLY8 only produced one allele. The total number of alleles from the 16 primer pairs in golden Asian arowana was 47, ranging from 2 to 5 with locus D33, D38 and D72 being the most polymorphic with 5 alleles, and markers SSRLY1, SSRLY3, SSRLY4, SSRLY9, SSRLY9, SSRLY10, D42, D85 and D92 being the least polymorphic, displaying only two alleles. The average allele number of the 16 polymorphic markers was 2.94 loci.

Heterozygosity (H), or gene diversity, can highlight the genetic variations of many loci in a population. It is thus considered to be a suitable parameter to estimate the genetic variation of a population. The observed heterozygosity (H_o) ranged from 0.1429 to 1.00 (Note 2). Expected heterozygosity (H_e) varied between 0.1429 in D42 and 0.8242 in D72. Mean intrapopulation diversity was different among loci, both in terms of number of alleles per locus and heterozygosity values (Note 2).

The polymorphic information content (PIC) is an index for analysis of the polymorphism of an amplified product. The PIC of the sixteen loci in golden Asian arowana population ranged from 0.124 to 0.865, and the average PIC was 0.432. According to the protocol of Botstein (1980). The PIC was over 0.432 in all cases, except with SSRLY6, D33, D38, D72 and D85 ($PIC > 0.5$), meaning that these locus were middling polymorphic and could be used to calculate the genetic diversity of golden Asian arowana population, which indicates medium genetic diversity in golden Asian arowana population.

Significant departures from Hardy-Weinberg equilibrium were tested using the parameter d . Heterozygote excess was found at loci SSRLY1, SSRLY2, SSRLY10, SSRLY11, SSRLY12, D33, D38, D42 and D92 in golden Asian arowana population. ($d < 0$). (Note 2)

AMOVA analysis with Arlequin software showed faint differentiation with 0.51% variation in population. This could be caused by gene flow between populations, because few migrants per generation are sufficient to eliminate genetic evidence of stock structure, and that is common in marine species (Waples, 1998).

4. Discussion

N_e , H_o , H_e and PIC are parameters of genetic variations which were 2.94, 0.446, 0.4945 and 0.432, respectively. Data showed the levels of genetic diversity were relatively middle in the population, compared to previously published surveys of population variability. Both the average observed heterozygosity (H_o) and average expected heterozygosity (H_e) were 0.446, 0.4945 and was higher than the mean heterozygosity of 13 species of fresh water fish calculated (DeWoody, 2001). Meanwhile, our results were relatively lower to the report (Yue, 2006), which analyzed the genetic population structure of a red Asian Arowana population with microsatellite markers and found H_e was 0.51–0.95, which suggest that high level of genetic diversity

The Hardy-Weinberg departure value (d) is a fixed index, describing the departure of a locus in a population from Hardy Weinberg equilibrium (HWE). It suggests the population is close to HWE when the value of d is closer to zero. $d > 0$ means the population lacks heterozygotes whereas $d < 0$ indicates heterozygote excess. The value ranges between -1 and 1. But there's no recognized standard. As shown (Note 2), nine loci with heterozygosity deficiency were deviated the HWE severely in gold population. Though Artificial selection, population substructuring, shortage of samples, mutation, low levels of polymorphism and inbreeding may lead to deviations from HWE, the presence of inbreeding caused by over-fishing and environmental could provide a reasonable explanation for the lack of agreement with HWE in the populations we analyzed. According to geological habitat that several of arowana strains diverged from late Pliocene to middle Pleistocene, gold Asian Arowana lived in Bukit Merah Lake of Malaysian and Sumatra of Indonesia (Dawes, 1999) and all the arowana strains are highly endemic to certain river systems and no hybridization could have occurred in hatcheries (Yue, 2000). On the other hand, Asian Arowana were fiercely hunted in its native habitat, causing declination of the population of these fish in the wild. So overfishing and habitat deterioration diminish the population size, and lead to reproductive isolation of unattached groups, which led to decreasing gene flow among the populations.

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Table 1. The amplification conditions and results of 20 microsatellite primers in *Scleropages formosus* Asian Arowana

Primer	Primer sequences(5' → 3')	Product size(bp)	Tm(°C)
SSRLY1	F:GAATGCTTAAAGTGGCAGTGAA R: TAACACAGGGCGTAAGGCCAG	174-216	55
SSRLY2	F:GCTTAAACCCATTACAGACAGG R:AAAGTGGTTTTGCATGAAGAAA	171-229	55
SSRLY3	F:AAGGGAGCAGCAGTTAGGTAGCAG R:AGAGGAAATGTTAATTCACCACGG	196-246	55
SSRLY4	F:GACTGGCGTCCCGTCCTG R:TATGCTCTTTCCCATTGACACTAA	224-256	57
SSRLY5	F:CTTGCGCCCTGTGTTGC R:TTACCAGCAGAAAAGGCCTT	127-176	57
SSRLY6	F:GTGTCAGTATAGTGAATCTGTAG R:ATCTCATTATGCTGCCATTGTCA	154-196	57
SSRLY7	F:GTTTGTCCCTCCATGCACTGAGAG R:CCAACAAAACCATGTGGCAATCAC	154-223	52
SSRLY8	F:AGCACCCCTGTTACTGGAAGAGA R:TTCTCAAAGCAAAAAGCATCACACT	236-292	55
SSRLY9	F:TATTACCATGCGCCCAGCACAC R:AGTCCTGCTTCTGGCTCACCCA	130-138	52
SSRLY10	F:AGCTGACACTTTGAAGCACT R:AAGAGTCGCTGAATTAGCAC	217-238	52
SSRLY11	F:CAGTGGTTGCACACTTACAG R:TATTTTCATCATGCCGACTTT	194-244	55
SSRLY12	F:GTTTCTTCTAGGTGCTCTGGTTTC R:GGATGAGTGACCCAGTGTAAGTAG	223-294	55
D11	F:TGGTTTCCACCTACAGTCCAAAGA R:GTTACGAGTACTGGCCCAATGG	154-170	50
D33	F:GTTCTTCTAGGTGCCGCGTTTC R:CTACTTACACTGGGTCATCATCC	190-216	50
D38	F:TTGGGGTCATGCCACTGG R:CAATAAATACCAAACAGGGAACC	179-216	53
D42	F:AGGAACATCACTGACAACACT R:TGGACTAACTAGGAGCACAT	145-201	50
D72	F:AGCAGGTTAATTTGGAGACT R:CGACCCTGTATGGGACAAG	105-144	55
D85	F:GTTCCACAGGGGCTGAGAAAAT R:GAGGACGGAACAAAAGCATTGG	140-166	50
D92	F:AGTCGCACACCACCACCTCAG R:TCAGCGATAACCCACACCT	190-220	50
D94	F:CAGCAGCACTGACACGGGTTTCG R:TCGCAGGCTGATTAAGGTGTG	194-246	50

Table 2. Allele frequencies, heterozygosity and polymorphism information content

Locus	Allele frequencies					No.of allele	<i>Ho/He</i>	<i>PIC</i>	D-value
	P1	P2	P3	P4	P5				
SSRLY1	0.6429	0.357				2	0.7143 (0.4945)	0.353	-0.5556
SSRLY2	0.7857	0.142	0.071			3	0.4286 (0.3846)	0.332	-0.2000
SSRLY3	0.6429	0.357				2	0.4286 (0.4945)	0.354	0.0667
SSRLY4	0.7143	0.285				2	0.2857 (0.4396)	0.324	0.3000
SSRLY6	0.143	0.429	0.286	0.143		5	0.2857 (0.7473)	0.653	0.5882
SSRLY9	0.7143	0.286				2	0.2857 (0.4396)	0.325	0.3000
SSRLY10	0.7143	0.286				2	0.5714 (0.4396)	0.325	-0.4000
SSRLY11	0.7857	0.214				2	0.4286 (0.3626)	0.280	-0.2727
SSRLY12	0.071	0.071	0.857			3	0.2857 (0.2747)	0.247	-0.1200
D11	0.0714	0.785	0.143			3	0.1429 (0.3846)	0.326	0.6000
D33	0.0714	0.142	0.286	0.143	0.357	5	0.8571 (0.8022)	0.733	-0.1507
D38	0.0714	0.142	0.286	0.357	0.143	5	1.0000 (0.8022)	0.715	-0.3425
D42	0.0714	0.928				2	0.1429 (0.1429)	0.124	-0.0769
D72	0.0714	0.285	0.286	0.143	0.214	5	0.7143 (0.8242)	0.746	0.0667
D85	0.2143	0.285				2	0.2857 (0.4396)	0.865	0.3000
D92	0.8571	0.142				2	0.2857 (0.4396)	0.215	-0.1667
mean						2.94	0.446 (0.4945)	0.432	

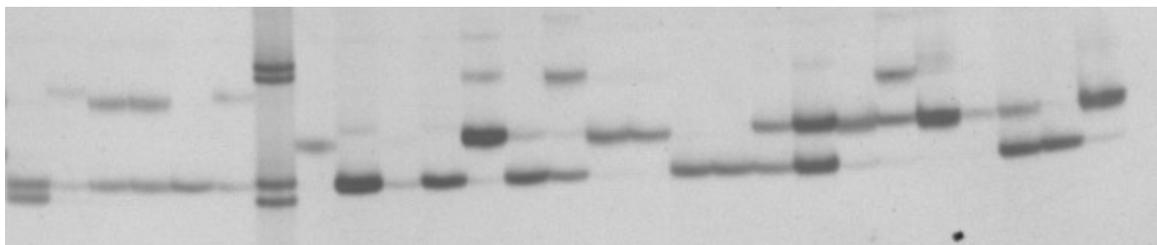


Figure 1. Electrophoregram of microsatellite primer D33 amplified in Golden Asian arowana



Genotypic Variation in Responses of *Citrus* spp. to Arbuscular Mycorrhizal Fungi

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The research is financed by the Thailand Research Fund and Commission on Higher Education

Abstract

Thailand is part of Southeast Asia that covers the center of diversity of citrus species, where various species of the genus are widely grown. One of the most common is tangerine (*Citrus reticulata*), which is commonly grown by grafting on rootstocks of different tangerine varieties or other citrus species. The objective of this study is to investigate responses of some *Citrus* spp. seedlings to arbuscular mycorrhizal (AM) fungi, and thus their potential as rootstocks. The experiment was done with four tangerine varieties, Cleopatra, Fremont, Ocean and Sainamphung; and four other citrus species, lime (*C. aurantifolia*), pomelo (*C. maxima*), sweet orange (*C. sinensis*) and Troyer citrange (*Citrus sinensis*×*Poncirus trifoliata*), in pots for five months. Roots of non-inoculated plants were not infected with AM fungi, while inoculated plants were heavily infected with AM fungi, and contained 14-28 AM spores per 10 g of rhizosphere soil. Most of the citrus responded positively to AM fungi, but with different magnitudes among the varieties and species. Lime and pomelo seedlings were fast growing compared to other citrus species. Total dry weight and N, P, K and Mg contents were increased most strongly by AM fungi in lime, pomelo and tangerine varieties Ocean, Fremont and Sainamphung, but little or none in Cleopatra, Troyer and sweet orange. Lime was the most outstanding in the response to AM fungi, followed by Ocean tangerine and pomelo. The potential of lime, pomelo and Ocean tangerine as rootstock for tangerine should be further investigated.

Keywords: Arbuscular mycorrhizal fungi, *Citrus* spp., Response

1. Introduction

Southeast Asia is generally considered the center of diversity of citrus (Moore, 2001). Various kinds of citrus plants are grown in all regions of Thailand, one of the most widely grown is tangerine (*C. reticulata*) especially the variety Sainamphung. Tangerine is commonly grown by grafting on rootstocks of different tangerine varieties or other citrus species. Tangerine variety Cleopatra and a hybrid citrange or Troyer (*Citrus sinensis*×*Poncirus trifoliata*) are used widely as rootstock in Sainamphung tangerine orchards in Thailand. Arbuscular mycorrhizal (AM) fungi are mutualistic associations with plant roots. They improve the nutritional status of plants resulting in increased growth of the host plants, and they can also improve soil structure (Douds and Millner, 1999). AM fungi are an important part of sustainable agricultural systems. Youpensuk et al. (2008) reported that twenty-two species of AM fungi were found in tangerine

orchards of Chiang Mai province and they increased growth of air layered tangerine variety Sainamphung especially in pots applied only N without P fertilizer. The objective of this study is to investigate responses to AM fungi of seedlings of tangerine varieties and other common citrus species, and thus their potential as rootstocks.

2. Materials and Methods

2.1 Preparing citrus seedlings

Tangerine (*C. reticulata*) varieties examined in this experiment were Cleopatra, Fremont, Ocean and Sainamphung, and the other common citrus plants were lime (*C. aurantifolia*), pomelo (*C. maxima*), sweet orange (*C. sinensis*) and a hybrid citrange or Troyer (*Citrus sinensis* × *Poncirus trifoliata*). The plants were grown from seeds that had outer seed coats were peeled off before germination in sterile soil in plastic trays and watered twice a day.

2.2 The responses of *Citrus* spp. seedlings to AM fungi

One month old seedlings were transplanted into drainable plastic pots containing 6 kg sterile soil, with one seedling per pot. The soil was a sandy clay loam with pH of 6.0. The soil contained 0.90 g/kg total N, 4.1 mg/kg available P, 53.0 mg/kg extractable K, and 18.5 g/kg organic matter. Spores of mixed AM fungal species were collected from the rhizosphere of *Citrus* spp. in northern Thailand. For inoculated treatments, three hundred spores of mixed species of AM fungi were inoculated to the planting hole in each pot. All treatments had four replications. Seedlings were watered once a day. Five months after transplanting, shoot height was measured for each plant. Shoots were separated from roots at the soil surface and dried at 70°C for three days to evaluate for shoot dry weight. Soil in each pot was divided into two subsamples. Roots were washed from each one soil subsample and dried at 70°C for three days to evaluate for root dry weight. After drying, shoot and root samples were ground and analyzed for N contents in citrus plants by Kjeldahl method. Dry ashes of the samples of citrus plants were evaluated for P by molybdovanado-phosphoric acid method, and evaluated for K and Mg by atomic absorption spectrophotometer.

2.3 Assessment of root colonization and spore density of AM fungi

Soil and root samples from the second subsamples of each pot were used for assessment of root colonization and spore density of AM fungi. Fifty g of soil sample from each pot was used to assess spore density and identification of the AM fungi. The soil samples were wet sieved through 750, 250, 100, and 53 µm mesh sieves. The sieved soil on each 250, 100, and 53 µm mesh was centrifuged at 2000 rpm for 5 min and floating particles removed. The soil was suspended in 50% sucrose and centrifuged one min at 2000 rpm. After centrifugation, spores in the supernatant were poured over the finest sieve and washed with water to remove the sucrose before vacuum filtration on filter paper with gridlines. Spores on filter paper were kept in Petri dishes and counted under a stereomicroscope. The spores of AM fungi were identified according to morphological characteristics of AM fungal descriptions (Schenck and Perez, 1988; INVAM website, 2008).

Root samples were washed with tap water and cut into about 1 cm in length, cleared in 10% KOH at 121°C for 15 min, washed over a sieve with tap water, and stained with 0.05% trypan blue in lactoglycerol at 121°C for 15 min. The stained root segments were randomly picked with fine tip forceps and mounted on slides. Thirty pieces of root segments from each sample were assessed root colonization of AM fungi according to the method of McGonigle et al (1990) under compound microscope.

2.4 Statistical analysis

The data were analyzed with SPSS software program for analysis of variance (ANOVA). Duncan's Multiple Range Test at $P \leq 0.05$ was used to determine significances of treatment means.

3. Results and Discussion

Roots of non-inoculated plants were not infected with AM fungi. Percentage of root colonization of AM fungi in roots of inoculated citrus plants was very high from 75 – 96% (Table 1). Spore densities in the pots were about 14-28 spores per 10 g soil. Spores production may not correlate to percentage of root colonization it depends on AM fungal ability to produce spores in each soil condition (Smith and Read, 1997; Youpensuk et al., 2006). Twenty species of AM fungi were found in pots of inoculated citrus plants after five months of inoculation (Table 2). They were in three genera of *Acaulospora* (7 species), *Glomus* (12 species) and *Scutellospora* (1 species). The most AM species frequently found in all pots of inoculated citrus plants were *Acaulospora scrobiculata*, *Glomus etunicatum* and *G. mosseae*. This was similar to the report of Youpensuk et al. (2008) which found that *G. etunicatum* and *A. scrobiculata* were the most frequently found in tangerine orchards in Chiang Mai province. In this experiment, some 13-14 species of AM fungi were found in the rhizosphere of tangerine. There were some difference in the AM fungal species among the different varieties of tangerine, such as *G. aggregatum* was found only in the tangerine variety Cleopatra but *A. rugosa* was not found in this variety while it was found in the other varieties, and *A. delicata* was found in only tangerine variety Ocean. There were 2-3 more AM fungal species in the rhizosphere of the other citrus species. The most abundant in AM fungal species was in the hybrid citrange or Troyer (Table 2). Bever (2002) reported that although AM fungal species can associate with all host but they have host-specific differences in their sporulation growth rates. Although they were not very different in

number of AM fungal species and percentage of root colonization but significant variation in responses to AM fungi were found between the tangerine varieties and among the citrus species (Table 3). Inoculation by AM fungi generally increased plant height in the citrus, but most strongly in pomelo, lime and Ocean tangerine. Inoculation with AM fungi increased shoot dry weight most strongly, by doubling the shoot dry weight or more, in two tangerine varieties, Fremont and Ocean, and also in lime and pomelo, less effect on the tangerine varieties Cleopatra and Sainamphung, but had little effect on sweet orange and Troyer. Root dry weight of AM inoculated lime was three times that in non-inoculated. AM inoculation did not have significant effect on root dry weight of any of the tangerine and other citrus species. Root to shoot ratios of inoculated treatments of all tangerine varieties and pomelo tended to be lower than those of non-inoculated treatments. This may be the result of greater nutrient uptake efficiency by the external hyphae of mycorrhizal roots, which resulted in increased shoot growth of the host plant. Mycorrhizal plants are frequently found to have lower root to shoot ratios than non-mycorrhizal plants (Marschner et al., 1996; Youpensuk et al., 2005). Lime was an exception, as both its root growth was tripled by AM inoculation while the shoot growth was only doubled, which resulted in the root to shoot ratio being increased by AM inoculation.

Comparative responses to AM fungi in these citrus species and tangerine varieties can be seen more clearly by comparing plant total dry weight with and without mycorrhiza (Table 4). Lime responded most strongly to AM fungi with a 115.5% increase in total dry weight, followed closely by tangerine Ocean with 97.7% and pomelo with 91.4% increase in dry weight. Significant, although lower, responses to AM fungi were found in tangerine Fremont (78.4%) and Sainamphung (64.9%). The response to AM fungi was not significant in Cleopatra tangerine, sweet orange and Troyer. The low response to AM fungi in Troyer has been previously reported by Camprubi and Calvet (1996). These authors, however, also reported that rootstocks of sour orange and Cleopatra were more mycorrhizal dependent than Troyer and Swingle citrumelo (*C. paradise* × *P. trifoliata*). The lack of response to AM fungi in sweet orange agrees with the report of Jifon et al. (2002), who reported that sweet orange did not respond to *Glomus intraradices* while sour orange or *C. aurantium* had about 15% of mycorrhizal dependence in high level of P and 70 Pa of CO₂. The results indicated that varieties or species of plants can respond differently to AM fungi. Soil condition also affect to responses of plants to AM fungi such as levels of available P in soil that AM plants more response to AM fungi in low P soil than in high P soil (Graham et al., 1997). In addition to variation in the effect of AM fungi on plant dry weight among the citrus species and tangerine varieties, AM fungi also had different effects on nutrients uptake of the different citrus plants that were sometimes similar to and sometimes different from the effects on dry weight (Table 5). Although AM fungi had little effect on dry weight of Cleopatra tangerine, but it significantly increased N, P and K content of the host plant. Similarly AM fungi significantly increased P content of sweet orange and N and P content of Troyer even though it did not affect their dry weight. The effect of AM fungi on nutrient uptake in some of the more responsive varieties of tangerine and species of citrus was very large. For example, P content of Fremont tangerine, lime and pomelo was increased by more than 300% by AM fungi. The largest increases in the uptake of these nutrients by AM fungi were found in Fremont tangerine, lime and pomelo, while the effect on Troyer was small and on Sweet orange nil. Troyer and Cleopatra are tolerant to *Phytophthora* spp. that cause root rot of plants (Graham and Timmer, 2009). Therefore, they are widely used in Thailand as rootstock for Sainamphung, but their limited response to AM fungi is a cause for concern. Should this response continue into trees in the orchards, it would mean that benefits from AM fungi will be minimal in commercial tangerine orchards. Many experiments reported that AM fungi increased P contents of the host plants. But AM fungi may or may not increase uptake of these N, K and Mg, depending on host plants, species of AM fungi and soil conditions (Marschner and Dell, 1994; Taylor and Harrier, 2001; Rutto et al., 2002). Wu and Xia (2006) reported that level of Mg in leaves of tangerine inoculated with AM fungi was higher in AM seedlings than those in non-AM seedlings under well-watered and water stress conditions. While the levels of K and Ca in leaves and roots were significantly higher in AM seedlings than those in non-AM seedlings only under well-water conditions.

4. Conclusions

All of the tangerine varieties and other citrus species studied were well colonized by AM fungi. Judging by spore morphology a whole range of AM fungi was found in the rhizosphere of these citrus. All of citrus also benefited from association with the AM fungi, but with different magnitudes in different varieties and species. Lime was the most outstanding in the response to AM fungi, followed by Ocean tangerine and pomelo. The limited response to AM fungi in Troyer and Cleopatra tangerine that are used as rootstocks in commercial tangerine orchards suggests a re-evaluation of tangerine rootstocks may be worthwhile. On the other hand the potential of lime, pomelo and Ocean tangerine as rootstock for tangerine should be further investigated.

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Table 1. Means of root colonization and spore densities of arbuscular mycorrhizal fungi in pots of inoculated citrus plants

Species or variety of citrus plant	Mean of root colonization (%)	Spore density (spores/10 g soil)
Cleopatra	75.0b	28a
Fremont	95.5a	14d
Ocean	90.6a	16bcd
Sainamphung	90.6a	23ab
Lime	91.8a	21abc
Pomelo	96.0a	22abc
Sweet Orange	95.1a	21abc
Troyer	94.5a	15cd

Means in the same column followed by different letters are significantly different ($P \leq 0.05$).

Table 2. Species of arbuscular mycorrhizal fungi found in pots of inoculated citrus plants, five months after inoculation with AM fungi

Genera of AM fungi		Species or variety of citrus plants										
	Cleopatra	Fremont	Ocean	Sainumphung	Lime	Pomelo	Sweet Orange	Troyer				
<i>Acaulospora</i>	-	-	<i>A. delicata</i>	-	<i>A. delicata</i>	<i>A. delicata</i>	<i>A. delicata</i>	-				
	<i>A. denticulata</i>	<i>A. denticulata</i>	<i>A. denticulata</i>	<i>A. denticulata</i>	<i>A. denticulata</i>	<i>A. denticulata</i>	<i>A. denticulata</i>	<i>A. denticulata</i>				
	<i>A. lacunosa</i>	<i>A. lacunosa</i>	<i>A. lacunosa</i>	<i>A. lacunosa</i>	<i>A. lacunosa</i>	<i>A. lacunosa</i>	<i>A. lacunosa</i>	<i>A. lacunosa</i>				
	<i>A. longula</i>	<i>A. longula</i>	<i>A. longula</i>	<i>A. longula</i>	<i>A. longula</i>	<i>A. longula</i>	<i>A. longula</i>	<i>A. longula</i>				
	<i>A. morrowiae</i>	<i>A. morrowiae</i>	<i>A. morrowiae</i>	<i>A. morrowiae</i>	<i>A. morrowiae</i>	<i>A. morrowiae</i>	<i>A. morrowiae</i>	<i>A. morrowiae</i>				
	-	<i>A. rugosa</i>	<i>A. rugosa</i>	<i>A. rugosa</i>	<i>A. rugosa</i>	<i>A. rugosa</i>	<i>A. rugosa</i>	<i>A. rugosa</i>	<i>A. rugosa</i>			
	<i>A. scrobiculata</i>	<i>A. scrobiculata</i>	<i>A. scrobiculata</i>	<i>A. scrobiculata</i>	<i>A. scrobiculata</i>	<i>A. scrobiculata</i>	<i>A. scrobiculata</i>	<i>A. scrobiculata</i>	<i>A. scrobiculata</i>			
	<i>G. aggregatum</i>	-	-	-	-	<i>G. aggregatum</i>	<i>G. aggregatum</i>	<i>G. aggregatum</i>	<i>G. aggregatum</i>			
	-	-	-	-	<i>G. australe</i>	-	-	-	-			
	<i>G. caledonium</i>	<i>G. caledonium</i>	<i>G. caledonium</i>	<i>G. caledonium</i>	<i>G. caledonium</i>	<i>G. caledonium</i>	<i>G. caledonium</i>	<i>G. caledonium</i>	<i>G. caledonium</i>			
<i>G. claroideum</i>	<i>G. claroideum</i>	<i>G. claroideum</i>	<i>G. claroideum</i>	<i>G. claroideum</i>	<i>G. claroideum</i>	<i>G. claroideum</i>	<i>G. claroideum</i>	<i>G. claroideum</i>				
-	-	-	-	-	-	-	-	<i>G. clavisorum</i>				
-	-	-	-	-	-	-	-	<i>G. constrictum</i>				
<i>G. etunicatum</i>	<i>G. etunicatum</i>	<i>G. etunicatum</i>	<i>G. etunicatum</i>	<i>G. etunicatum</i>	<i>G. etunicatum</i>	<i>G. etunicatum</i>	<i>G. etunicatum</i>	<i>G. etunicatum</i>				
<i>G. fasciculatum</i>	<i>G. fasciculatum</i>	<i>G. fasciculatum</i>	<i>G. fasciculatum</i>	<i>G. fasciculatum</i>	<i>G. fasciculatum</i>	<i>G. fasciculatum</i>	<i>G. fasciculatum</i>	<i>G. fasciculatum</i>				
<i>G. invernatum</i>	<i>G. invernatum</i>	<i>G. invernatum</i>	<i>G. invernatum</i>	<i>G. invernatum</i>	<i>G. invernatum</i>	<i>G. invernatum</i>	<i>G. invernatum</i>	<i>G. invernatum</i>				
<i>G. macrocarpum</i>	<i>G. macrocarpum</i>	<i>G. macrocarpum</i>	<i>G. macrocarpum</i>	<i>G. macrocarpum</i>	<i>G. macrocarpum</i>	<i>G. macrocarpum</i>	<i>G. macrocarpum</i>	<i>G. macrocarpum</i>				
<i>G. mosseae</i>	<i>G. mosseae</i>	<i>G. mosseae</i>	<i>G. mosseae</i>	<i>G. mosseae</i>	<i>G. mosseae</i>	<i>G. mosseae</i>	<i>G. mosseae</i>	<i>G. mosseae</i>				
-	-	-	-	-	-	-	-	<i>G. multicaule</i>				
								<i>S. heterogama</i>				
Total of AM species	13	13	14	13	15	15	16	17				

Table 3. Effect of AM fungi on height, shoot and root dry weight (DW) of tangerine varieties and other citrus plants, five months after inoculation

Species or variety of citrus plant	Height (cm)	Shoot DW (g/plant)	Root DW (g/plant)	Root:shoot
Cleopatra (M-)	23.67f	1.79e	1.47d	0.82
Cleopatra (M+)	32.73de	2.73de	1.32d	0.48
Fremont (M-)	29.23ef	2.34de	2.06cd	0.88
Fremont (M+)	37.78cd	4.76bc	3.09cd	0.65
Ocean (M-)	29.98e	2.63de	1.69cd	0.64
Ocean (M+)	46.60bc	6.14bc	2.38cd	0.39
Sainamphung (M-)	32.90de	1.84e	1.98cd	1.08
Sainamphung (M+)	33.10de	3.35de	2.95cd	0.88
Lime (M-)	48.80bc	5.02bc	2.45b	0.49
Lime (M+)	65.93a	11.79a	7.31bc	0.62
Pomelo (M-)	39.35bc	4.97b	3.87ab	0.78
Pomelo (M+)	62.55a	11.08a	5.84a	0.53
Sweet orange (M-)	33.85cd	5.44b	3.61ab	0.66
Sweet orange (M+)	44.38bc	5.53b	3.68ab	0.67
Troyer (M-)	51.00bc	3.80cd	2.40cd	0.63
Troyer (M+)	55.13b	3.87cd	2.40cd	0.62
Analysis of variance				
Citrus	***	***	***	
Inoculation	***	***	NS	
Citrus × Inoculation	NS	***	NS	

M-, non-inoculated with AM fungi; M+, inoculated with AM fungi. Means in the same column followed by different letters are significantly different ($P \leq 0.05$). ***, significant at $P \leq 0.001$; NS, not significant.

Table 4. Total dry weight and response to mycorrhizal fungi of tangerine varieties and other citrus plants, five months after inoculation with arbuscular mycorrhizal fungi

Plant	Total dry weight (g/plant)		Response to mycorrhizal* (%)
	Non-mycorrhizal plant	Mycorrhizal plant	
Tangerine			
Cleopatra	3.26cA	4.04cA	23.9
Fremont	4.40cB	7.85bcA	78.4
Ocean	4.31cB	8.52bcA	97.7
Sainamphung	3.82cB	6.30cA	64.9
Other citrus spp.			
Lime	7.47abB	16.10aA	115.5
Pomelo	8.84aB	16.92aA	91.4
Sweet Orange	9.05aA	9.16bA	1.2
Troyer	6.20bcA	6.27cA	1.1

*Dry weight of mycorrhizal plant as percentage of dry weight of non-mycorrhizal plant. Means of total dry weight followed by different letters (lower case in the same column and capital letter in the same row) were significantly different ($P \leq 0.05$).

Table 5. Effect of arbuscular mycorrhizal fungi on nutrient contents in tangerine varieties and other citrus plants, five months after inoculation.

Species or variety of citrus plant	Nutrient content in citrus plants (mg/plant)			
	N	P	K	Mg
Cleopatra (M-)	47.63b	5.98b	47.25b	8.09a
Cleopatra (M+)	70.85a	12.99a	58.67a	10.93a
Fremont (M-)	59.01b	8.33b	109.84b	9.22b
Fremont (M+)	139.00a	38.19a	153.39a	14.25a
Ocean (M-)	67.41b	8.79b	104.31b	8.56b
Ocean (M+)	134.08a	24.22a	142.00a	13.49a
Sainamphung (M-)	47.51b	10.12b	86.54b	9.72b
Sainamphung (M+)	111.46a	30.11a	124.26a	15.47a
Lime (M-)	104.44b	14.46b	154.56b	14.90b
Lime (M+)	239.31a	62.51a	312.07a	37.32a
Pomelo (M-)	95.91b	10.89b	149.80b	20.00b
Pomelo (M+)	201.97a	44.52a	270.66a	34.02a
Sweet Orange (M-)	141.12a	18.45b	220.37a	17.92a
Sweet Orange (M+)	151.03a	28.73a	161.70b	14.92a
Troyer (M-)	71.72b	15.66b	114.22a	12.56a
Troyer (M+)	107.05a	27.28a	114.16a	12.19a

Means of non-inoculated (M-) and inoculated (M+) with AM fungi in the same tangerine variety or citrus species followed by different letters were significantly different ($P \leq 0.05$).



Some Physical and Mechanical Properties of Fennel Seed (*Foeniculum vulgare*)

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Abstract

Determination of physical and mechanical properties of seeds and agricultural products is important in the design of harvesting, handling, and processing equipment. Some physical and mechanical properties of fennel seed were determined as a function of moisture content in the range of 7.78–21.67% d.b. The average length, width and thickness were 58.87, 18.96 and 15.64 mm, at a moisture content of 7.78 %d.b., respectively. In the moisture range from 7.78% to 21.67% d.b., studies on rewetted fennel seed showed that the thousand seed weight increased from 5.5 to 9.2 g, the porosity from 55.91% to 62.21%, the static and dynamic angle of repose from 37.6 to 46.6 and 41 to 53.3, respectively, the coefficient of friction on glass, plywood, and galvanized iron sheet surfaces from 0.55 to 0.74, 0.45 to 0.63, and 0.43 to 0.66, respectively, and deformation on width section increased from 1.68 to 1.86 mm. The bulk density decreased from 413.51 to 352.39 kg m⁻³ and rupture force on both seed length and width sections decreased from 198.93 to 78.68 N, and 600.65 to 186.44 N, respectively, with moisture content in the moisture range of 7.78 to 21.67% d.b. But there was not regular trend for sphericity, true density, and deformation on length section with increasing the moisture content.

Keywords: Angle of repose, Density, Rupture force, Medical plant seed, Moisture content, Porosity

1. Introduction

In the world today, the traditional food, forage and fiber crops are not the only plants of key agricultural and trade significance, but they also include plants whose secondary metabolites are valued for their characteristic aromatic or

therapeutic attributes, or as main natural inputs to the proliferating perfumery and chemical industries. Fennel (*Foeniculum vulgare*) is a plant of the Apiaceae (Umbelliferae) family largely used to impart flavor to a number of foods, such as soup, sauces, pickles, breads, cakes, etc. It is indigenous to the Mediterranean and is cultivated in England, Germany, China, Iran, Vietnam and South America. Diuretic, analgesic and antipyretic activity has also been found in the fennel seed as well as antioxidant activity. The most frequently investigated was the essential oil which showed antioxidant, antimicrobial and hepatoprotective activity (Lucinewton et al., 2005). Present world market of fennel seed is around US\$ 80 million. Worth of Iran export of this product is about US\$ 10 million (Masood et al., 2004).

In order to design equipment for the handling, conveying, separation, drying, aeration, storing and processing of fennel seeds, it is necessary to determine their physical and mechanical properties as a function of moisture content. The properties of different types of grains and seeds have been determined by other researchers such as Singh and Goswami (1966) for cumin seed; Shepherd and Bhardwaj (1986) for pigeon pea; Dutta et al. (1988) for gram seed; Oloso and Clarke (1993) for cashew nuts; Ougt (1998) for white Lupin; Baryeh (2002) for millet; Ogunjimi et al. (2002) for locust bean seed; Amin et al. (2004) and carman (1996) for lentil seed; Coskun et al. (2006) for sweet corn seed; and Cetin (2007) for barbania bean.

However, no published work seems to have been carried out on the physical and mechanical properties of fennel seed and their relationship with moisture content. Hence, this study was conducted to investigate some moisture dependent physical and mechanical properties of fennel seed namely: size, thousand seed mass, sphericity, true density, bulk density, porosity, static coefficient of friction against different materials, angle of static and dynamic repose, and cracking force at safe storage moisture content.

2. Materials and methods

The dry seeds of fennel were used for all the experiments in this study (see Figure 1). The seeds were cleaned manually to remove all foreign matter such as dust, dirt, stones and chaff as well as immature and broken seeds. The initial moisture content of the seeds was determined by oven drying at $105 \pm 1^\circ\text{C}$ for 24 h (Ozarlan, 2002). The initial moisture content of the seeds was 7.78% d.b. The samples of the desired moisture contents were prepared by adding the amount of distilled water as calculated from the following relation (Sacilik et al. 2003):

$$Q = \frac{W_i(M_f - M_i)}{(100 - M_f)}$$

where W_i , is the initial mass of sample in kg; M_i , is the initial moisture content of sample in % d.b.; and M_f , is the final moisture content of sample in % d.b.

The samples were then poured into separate polyethylene bags and the bags sealed tightly. The samples were kept at 5°C in a refrigerator for a week to enable the moisture to distribute uniformly throughout the sample. Before starting a test, the required quantity of the seed was taken out of the refrigerator and allowed to equilibrate to the room temperature for about 2 h (Singh and Goswami, 1996; Coskun et al. 2006). All the physical and mechanical properties of the seeds were determined at five moisture contents in the range of 7.78–21.67% d.b. with four replications at each moisture content.

The following methods were used in the determination of some physical and mechanical properties of fennel seed.

2.1 Size and shape

A vernire caliper was used to measure the axial dimensions of randomly selected 100 seeds; length, width, and thickness. From the average of axial dimensions the geometric mean diameter D_g in mm was determined by using the following formula (Joshi et al., 1993):

$$D_p = (abc)^{1/3}$$

where: a, the length is the dimension along the longest axis in mm; b, the width is the dimension along the longest axis perpendicular to a in mm; and c, the thickness, is the dimension along the longest axis perpendicular to both a and b in mm.

The sphericity was determined using (Mohsenin, 1970):

$$\psi = \frac{(abc)^{1/3}}{a}$$

2.2 Thousand seed weight

Thousand seed weight (TSW) was measured by counting 100 seeds and weighing them in an electronic balance to an accuracy of 0.001 g and then multiplied by 10 to give mass of 1000 seeds.

2.3 Bulk and true densities

The bulk density was determined by filling an empty 250 ml graduated cylinder with the seed and weighed (Mohsenin,

1970). The weight of the seeds was obtained by subtracting the weight of the cylinder from the weight of the cylinder and seed. To achieve uniformity in bulk density the graduated cylinder was tapped 10 times for the seeds to consolidate. The volume occupied was then noted. The process was replicated four times and the bulk density for each replication was calculated from the following relation:

$$\rho_b = \frac{W_s}{V_s}$$

where: the ρ_b is the bulk density in kg m^{-3} ; W_s is the weight of the sample in kg; and V_s is the volume occupied by the sample in m^3 .

The true density was defined as the ratio between the mass of fennel seeds and the true volume of the seeds, and determined using the toluene (C_7H_8) displacement method. Toluene was used instead of water because it is absorbed by seeds to a lesser extent. The volume of toluene displaced was found by immersing a weighted quantity of fennel seeds in the measured toluene (Tavakkoli et al. 2006).

2.4 Porosity

This was calculated from the values of bulk and true densities using the following relationship (Mohsenin, 1970):

$$\varepsilon = \left(1 - \frac{\rho_b}{\rho_t}\right) \times 100$$

where ρ_t is true density and ε is the porosity.

2.5 Angle of repose

The static angle of repose (θ_s) was determined by using the apparatus in Figure 2 consisting of plywood box of 140*160*35 cm and two plates: fixed and adjustable. The box was filled with the sample and then the adjustable plate was inclined gradually allowing the kernels to follow and assume a natural slope (Tabatabaefar, 2003; Heidabeigi et al., 2009). The dynamic angle of repose (θ_d) was determined using a hollow cylinder and then trigonometry rules (Mohsenin, 1970).

2.6 Coefficient of friction

Coefficient of static friction of fennel seed on three surface including wood, galvanized steel, and glass was determined. In order to achieving this end, fennel seeds were put on the surface with changeable slip. When seeds were started to motion, tangent of slip angle was showed the coefficient of friction (Heidabeigi et al., 2009).

2.7 Mechanical properties

Rupture Strength for fennel seed was determined from forces acting on the two sections length and width with speed of load 5 mm/min (see Figure 3). For width section, samples were prepared as a cubic shape. Method of test was putting the seed on desired section and selecting speed of loading and after that applying force till seed fractured. Instron Universal Testing Machine (Model Santam STM-5), that is equipped with a 25 kg compression load cell and integrator, was used for this test. The measurement accuracy was 0.001 N in force and 0.001 mm in deformation (Mohsenin, 1970; Ahmadi et al. 2009).

The individual seed was loaded between two parallel plates of the machine and compressed at the present condition until rupture occurred as is denoted by a bio-yield point in the force-deformation curve. Once the bio-yield was detected, the loading was stopped. The mechanical properties of fennel seed were expressed in terms of rupture required for initial rupture.

2.8 Statistical analysis

The results gained were subjected to statistical analysis using SPSS 13 (SPSS Inc., USA) software and analysis of regression using Microsoft Excel 2003 (Microsoft Corp., USA).

3. Results and discussion

3.1 Seed Moisture Content

The initial moisture content of the seed was found to be $7.78 \pm 0.31\%$ d.b. The four other moisture levels obtained after conditioning the seeds were 10.91 ± 0.53 , 14.83 ± 0.72 , 17.76 ± 1.02 , and $21.67 \pm 0.34\%$ d.b., respectively. The investigations were carried out at the above moisture levels to determine the effect of moisture content on the physical and mechanical properties of fennel seed.

3.2 Size and shape

The variation of the seed length, width, thickness and geometric mean diameter variation with seed moisture content is displayed in Table 1. The table shows that the three axial dimensions increased significantly ($P < 0.05$) with moisture content in the moisture range of 7.78 to 21.67% d.b. The Length increased from 58.87 to 69.04 mm, the width from

18.96 to 21.96 mm, and the thickness from 15.64 to 18.47 mm. Also the geometric mean diameter increased with increase in moisture content. These could be of important consideration in the theoretical determination of the seed volume at different moisture contents.

Similar results of increase are reported by Tavakkoli et al. (2009) for soybean grains and Al-Mahasneh and Rababah (2007) for green wheat.

3.3 Sphericity

The sphericity was found to increase from 43.91 at the moisture content of 10.97% d.b. to a maximum value of 45.1 at the moisture content of 17.76% d.b., after which it decreased with further increase in moisture content. The effect of moisture content on the seed sphericity is presented in Figure 4. The relationship existing between sphericity and moisture content was found to be parabolic and can be expressed using the following equation with a coefficient of determination $R^2 = 0.63$:

$$\psi = -0.0194M^2 + 0.5969M + 40.331$$

Former researches showed that sphericity could be affected by moisture content in different ways. A decrease in the sphericity with increase in moisture content was observed for pigeon pea (Baryeh and Mangope, 2002). Cuskun et al. (2006) showed that sphericity of sweet corn seed is increased linearly by moisture content.

3.4 Thousand seed weight

Figure 5 shows the thousand seed weight variation with seed moisture content ($P < 0.05$). The Figure indicates that the thousand seed mass increases linearly with increase in seed moisture content. The variation can be expressed mathematically as follows with a coefficient of determination $R^2 = 0.96$:

$$TSW = 0.2498M + 3.8355$$

3.5 Bulk and true densities

The variation of bulk density and true density with seed moisture content is depicted in Figure 6. This shows that the bulk density of seed decreased from 413.51 to 352.39 kg m^{-3} in the above moisture range ($P < 0.05$). The relationship existing between bulk density and the seed moisture content can be expressed with the following equation:

$$\rho_b = -15.621M + 431.82, \quad R^2 = 0.97$$

True density decreased from 937.98 to 889.08 kg m^{-3} as the moisture content increased from 7.78 to 14.83% d.b. and increased to 932.62 kg m^{-3} as the moisture content increased to 21.67% d.b. The relationship existing between true density and moisture content was found to be parabolic and can be represented by the following equation:

$$\rho_t = 9.8021M^2 - 62.266M + 996.07, \quad R^2 = 0.82$$

Carman (1996), Gupta and Das (1998), and Visvanathan et al. (1996) found the bulk density of lentil seeds, sunflower seeds, neem nuts to decrease as the seed moisture content increases. Increase of true density as the seed moisture content increases has been found by Gupta and Das (1998) for sunflower seeds, Aviara et al. (1999) for guna seeds, Chandrasekar and Visvanathan (1999) for coffee.

3.6 Porosity

Figure 7 shows the porosity variation with seed moisture content. The Figure reveals that the porosity increases with increase in seed moisture content from 55.91% porosity at 7.78% d.b. seed moisture content to 62.21% porosity at 21.67% seed moisture content. The relationship existing between porosity and seed moisture content was found to be linear and can be expressed using the following equation with a coefficient of determination $R^2 = 0.87$:

$$\varepsilon = 0.4418M + 51.574$$

An increase in porosity with moisture content were reported for chickpea seeds (Konak et al., 2002), and green gram (Nimkar and Chattopadhyay, 2001).

3.7 Angle of repose

Figure 8 shows the variation of both the static and dynamic angle of repose with seed moisture content. From this Figure, it can be seen that both the static and dynamic angle of repose increased linearly ($P < 0.05$) in the moisture range of 7.78 to 21.67% d.b. from 37.6 to 46.6 and from 41 to 53, respectively. This shows that the amount of dynamic angle of repose was higher than static angle of repose at each moisture content. The relationship existing between the static and dynamic angle of repose and moisture content can be expressed using the following equations:

$$\begin{aligned} \theta_s &= 2.26M + 35.5, & R^2 &= 0.99 \\ \theta_d &= 3.19M + 37.93, & R^2 &= 0.95 \end{aligned}$$

These results were similar to those reported by Garnayak et al. (2008) for jatropha seed and Pradhan et al. (2008) for karanja kernel.

3.8 Coefficient of friction

The coefficient of friction of fennel seed increased linearly ($P < 0.05$) for glass surface and logarithmically for both Plywood and galvanized iron sheet surfaces with moisture content and varied with structural surface in the moisture range of 7.78 to 21.67% d.b. (see Figure 9). The maximum value of 0.74 was obtained on the surface of glass and the minimum value of 0.43 was on the surface of galvanized iron sheet. The relationship existing between the coefficient of friction and moisture content can be expressed for different structural surfaces using the following equations:

$$f_g = 0.0124M + 0.4504$$

$$f_w = 0.1723\ln(M) + 0.1129$$

$$f_{gis} = 0.215\ln(M) + 0.0171$$

with coefficients of determination R^2 of 0.93, 0.94, and 0.92, respectively, where f_g , f_w , and f_{gis} are the static coefficients of friction of seed on glass, Plywood, and galvanized iron sheet, respectively.

Similar findings were reported for millet (Baryeh, 2002), pumpkin seeds (Joshi et al., 1993), and karingda seeds (Suthar and Das, 1996).

3.9 Rupture strength

Effect of seed moisture content and force axis on the rupture force and deformation is shown in Table 2. The table shows that the rupture force on both the seed length and width sections decreased with moisture content in the moisture range of 7.78 to 21.67% d.b. Deformation on the seed length section increased in the moisture range of 10.91 to 21.67% d.b. But there was not regular trend for deformation on the seed width section with increasing the moisture content. The relationship existing between the both rupture force and deformation with moisture content can be expressed using the following equations:

$$F_L = -404.55\ln(M) + 1406.9 \quad , \quad R^2=0.98$$

$$F_w = -138.19\ln(M) + 491.61 \quad , \quad R^2= 0.89$$

$$D_L = 0.0015M^2 - 0.0326M + 1.861 \quad , \quad R^2=0.88$$

$$D_w = 0.0021M^2 - 0.0725M + 1.707 \quad , \quad R^2=0.91$$

Where F_L , F_w , D_L , and D_w are rupture force on seed length section, rupture force on seed width section, deformation on seed length section, and deformation on seed width section, respectively.

Tavakoli et al. (2009) for soybean grain reported that the rupture force decreased when increase the moisture content.

4. Conclusions

Some physical and mechanical properties of fennel seed are presented in this research and it can be concluded that:

- (1) In the moisture range of 6.78 to 21.67% d.b. the length, width, thickness, and geometric mean diameter of the fennel seed increased from 58.87-64.09, 18.96-21.96, 15.64-18.87, and 25.95-30.36 mm, respectively.
- (2) Thousand seed weight, porosity, static and dynamic angle of repose, and coefficient of friction on glass surface increased linearly, while bulk density reduced linearly and coefficient of friction on both Plywood and galvanized iron sheet surfaces increased logarithmically.
- (3) There was a parabolic mathematical equation for sphericity, true density, and deformation on both seed length and width sections with changes of moisture content.
- (4) Rupture force on both seed length and width sections decreased logarithmically with moisture content in the above moisture range.
- (5) Results of this research showed that the porosity affects the resistance to airflow through bulk solids. The static coefficient of friction and angle of repose is necessary to design conveying machine and hoppers used in planter machines. When seeds are ground in mills, the rupture force must be known in order to achieve desirable properties without unnecessary expenditure of energy.

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Table 1. Variation of principal dimensions with seed moisture content

Seed moisture content, % d.b.	Length, mm	Width, mm	Thickness, mm	Geometric mean diameter, mm
7.78	58.87 (0.8)*	18.96 (0.17)	15.64 (0.2)	25.95
10.91	59.94 (0.56)	19.29 (0.14)	15.86 (0.11)	26.37
14.83	64.09 (0.63)	21.19 (0.22)	17.71 (0.21)	28.86
17.76	65.3 (0.63)	21.37 (0.17)	18.31 (0.16)	29.45
21.67	69.04 (0.74)	21.96 (0.23)	18.47 (0.15)	30.36

* Values in parentheses are standard deviation

Table 2. Effect of seed moisture content and force axis on rupture force and deformation

		Seed moisture content, %d.b.				
		7.78	10.91	14.83	17.7 6	21.67
Rupture force, N	Length	600.25 (8.41)	417.43 (9.63)	294.26(7.56)	239.48 (5.9)	186.44 (4.9)
	Width	198.83 (13.97)*	189.03 (17.98)	99.39 (8.04)	82.97 (13.21)	78.68 (3.43)
Deformation, mm	Length	1.71 (0.26)	1.68 (0.11)	1.69 (0.52)	1.81 (0.9)	1.86 (0.3)
	Width	1.28 (0.11)	1.15 (0.14)	1.09 (0.47)	1.1 (0.51)	1.12 (0.1)

* Values in parentheses are standard deviation



Figure 1. Fennel Seeds

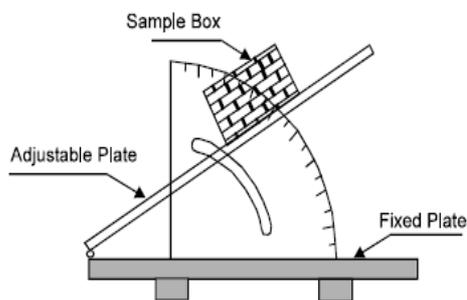


Figure 2. Apparatus to determine angle of repose

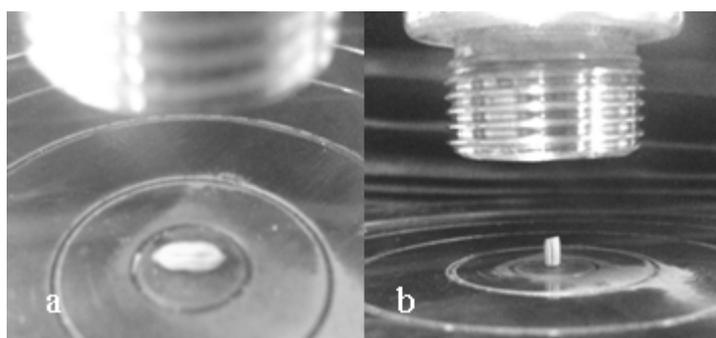


Figure 3. Rupture strength test for fennel seed
 a. force acting on the seed length section b. force acting on the seed width section

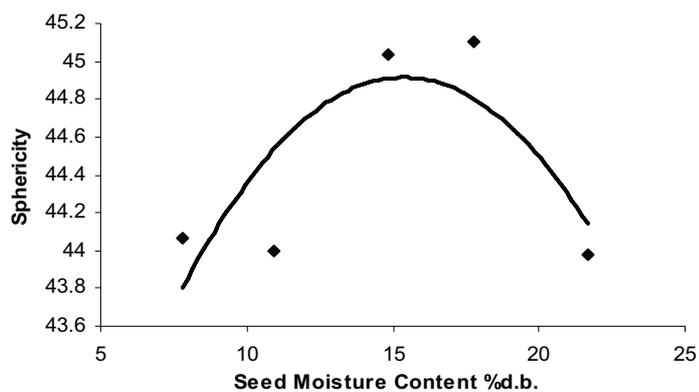


Figure 4. Variation of the sphericity of fennel seed with moisture content

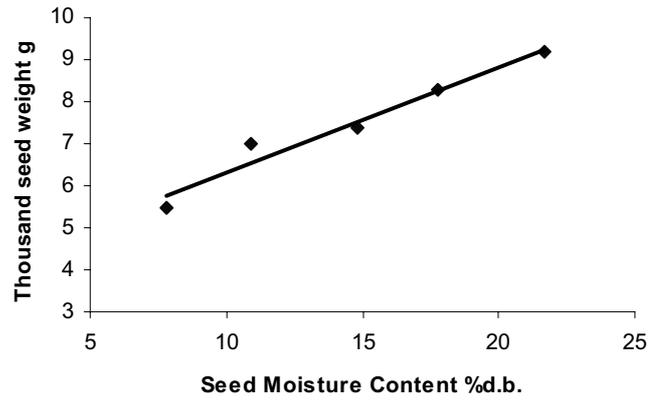


Figure 5. Effect of moisture content on the thousand seed weight

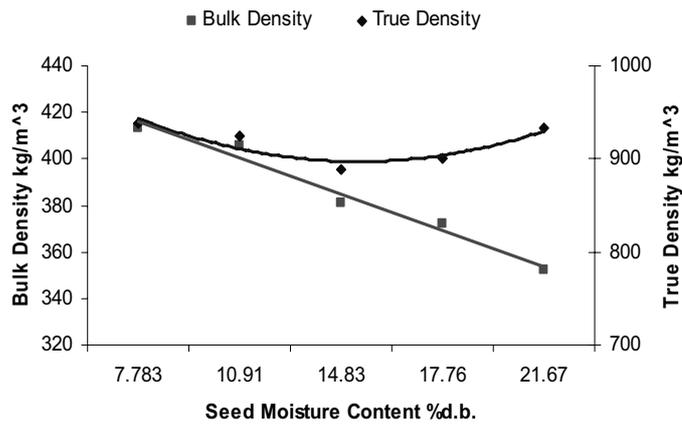


Figure 6. Bulk and true density variation with seed moisture content

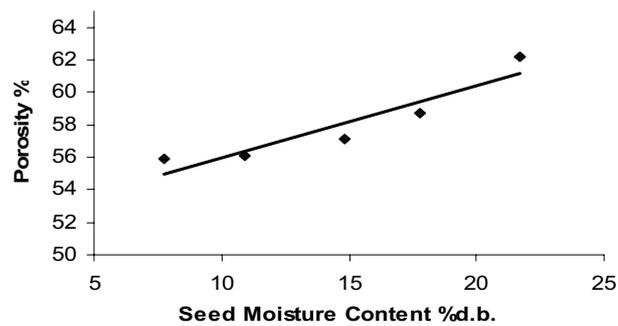


Figure 7. Effect of moisture content on the porosity

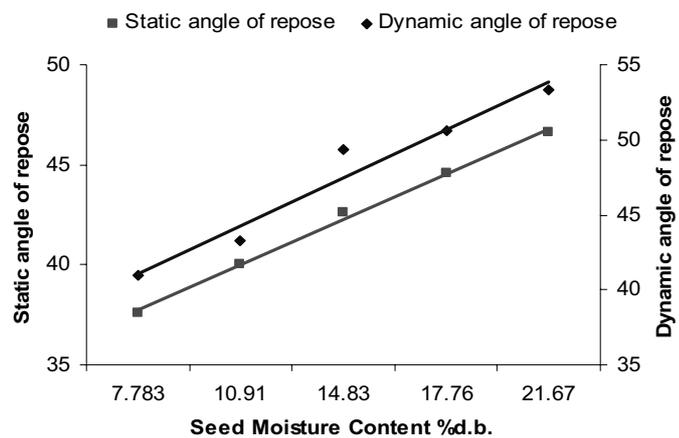


Figure 8. Static and dynamic angle of repose variation with seed moisture content

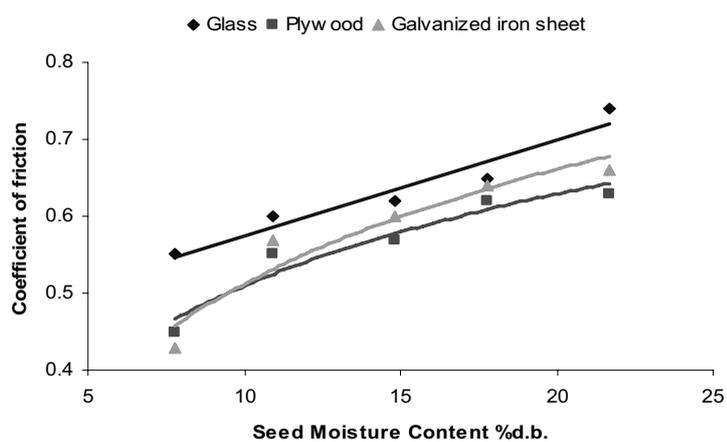


Figure 9. Coefficient of friction variation with seed moisture content



Molluscicide from Tobacco Waste

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Abstract

Tobacco waste can be molluscicide for Golden Apple snail control in paddy field as trialed in 3 experiments e.g. laboratory, green house and field trial. In laboratory found LD₅₀ of tobacco waste on golden apple snail, fish, frog and crab were 5456.25, 687.5, 1562.5 and 5000 Kg/ha respectively. In greenhouse and field trial were found rate of tobacco waste at 1562.5 Kg/ha could kill golden apple snail 100% in two days. Tobacco waste affected to increased electrical conductivity, biological oxygen demand, chemical oxygen demand in water but decreased water pH and dissolved oxygen. Otherwise, tobacco waste can be supplied soil fertility with high organic matter, available phosphorus, exchangeable potassium, calcium and magnesium. Tobacco waste can be increased rice growth and yield without nicotine and acute toxicity inspected.

Keywords: Molluscicide, tobacco waste, Golden apple snail

1. Introduction

Golden apple snail (*Pomacea canaliculata Lamarck*) is native animal of South America. It was imported to Thailand in 1982 for aquarium purposes. It was not prefer for customer then eliminate to river and canal and it wide spread to paddy field. Snail can consumes early growth of rice after planting till 15 day after planting. In Thailand, Golden apple snail outbreak since 1985 within 60 provinces on damaged area 34.375 million ha (Department of Agricultural Extension, 2000). The controlling procedures are man-power and chemical treated with Methaldehyde, tea seed and niclosamide (Bio-Agro Thai, 2007 and Promrangsana, 2007). Tobacco waste from cigarette production process is about 5 ton/day. Normally, tobacco waste is indigenous pesticide for aphid and cutworm control. In this study was studied on quality and rate of tobacco waste on Golden apple snail control. It is possible to reduce chemical import and organic waste garbage pollution.

This project was corroborated between Thailand Tobacco Monopoly, Ministry of Finance and Fertilizer Technology center, Thailand Institute of Scientific and Technological Research in 2007. Tobacco waste from cigarette production process in powder form was tested in laboratory, green house and field to solve out optimum rate of application for Golden apple snail control and affected on rice growth, rice quality and water quality.

2. Materials and methods

2.1 Laboratory trial

- Studied on LD₅₀ of tobacco waste on aquatic animals e.g. fish, crab, frog and golden apple snail.
- The effect of tobacco waste on dissolved oxygen, pH and electrical conductivity.
- Studied on dissolved nicotine and decomposition rate of nicotine at various times and rates of tobacco waste.
- Studied on shelf life of nicotine in tobacco waste at room temperature and freezes dry at 4°C.

- Toxicity testing on mammal e.g. acute oral toxicity, acute dermal toxicity testing and acute dermal irritation (Auletta, C.S. and Acute, 1995; Organization for Economic Co-operation and Development, 1987; Organization for Economic Co-operation and Development, 2002 and United State Environment Protection Agency (EPA), 1998).

2.2 Green house trial

There were 2 trials established in green house at Fertilizer technology center, Klong luang, Phatumthani. First trial was tested on various rates of tobacco waste between 0 to 9375 Kg/ha on rice growth. There were 9 treatments (tobacco waste at 0, 312.5, 625, 1562.5, 3125, 4687.5, 6250, 7812.5 and 9375 Kg/ha) in 4 replications. Second trial was tested on tobacco waste mixed with chemical fertilizer in 7 treatments (control, basal with chemical fertilizer grade 16-16-8 @ 156.25 Kg/ha and top dressing with 46-0-0 @ 31.25 Kg/ha, tobacco waste 625 Kg/ha, tobacco waste 1562.5 Kg/ha, tobacco waste 625 Kg/ha mix with chemical fertilizer grade 16-16-8 @ 78.125 Kg/ha, tobacco waste 1562.5 Kg/ha mix with chemical fertilizer grade 16-16-8 @ 78.125 Kg/ha and tea seed 15.625 Kg/ha mix with chemical fertilizer grade 16-16-8 @ 156.25 Kg/ha) in 4 replications. The data was collected on tobacco waste efficiency to control golden apple snail (mortality), rice damage, water qualities e.g. BOD, COD, DO, pH, electrical conductivity, chemical soil properties e.g. pH, electrical conductivity, organic matter, available phosphorus, exchangeable potassium, exchangeable calcium and exchangeable calcium (Scientific for land development division, Land Development Department, 2004) and affect on rice growth e.g. height, tiller, biomass and yield.

2.3 Field trial

There were 2 trials located in Chainat Rice Research Center, Chainat province. First trial was tested in 6 treatments (control, tobacco waste at 625, 1562.5, 3125, tea seed 15.625 Kg/ha, tea seed 31.25 Kg/ha and copper sulphate 6.25 Kg/ha) in 4 replications. Second trial was tested on tobacco waste mixed with chemical fertilizer in 7 treatments (control, basal with chemical fertilizer grade 16-16-8 @ 156.25 Kg/ha and top dressing with 46-0-0 @ 31.25 Kg/ha, tobacco waste 625 Kg/ha, tobacco waste 1562.5 Kg/ha, tobacco waste 625 Kg/ha mix with chemical fertilizer grade 16-16-8 @ 78.125 Kg/ha, tobacco waste 1562.5 Kg/ha mix with chemical fertilizer grade 16-16-8 @ 78.125 Kg/ha and tea seed 15.625 Kg/ha mix with chemical fertilizer grade 16-16-8 @ 156.25 Kg/ha) in 4 replications. The data were determined as collected in green house trials.

3. Results and discussion

3.1 Laboratory trial

Chemical properties of tobacco waste was contained of pH 5.28, electrical conductivity 19.08 dS/m, organic matter 46.65%, C/N ratio 20.19, total nitrogen 1.34%, total phosphorus 0.34%, total potassium 0.24% and nicotine 1.9 ppm.

3.1.1 The effect of tobacco waste on aquatic animal was found at different rates of tobacco waste affected on Golden apple snail mortality significantly. LD₅₀ of tobacco waste on golden apple snail was 5456.25 Kg/ha. Tobacco waste was affected on fish significantly. The rate of tobacco waste at 1562.5 Kg/ha can killed fish 85% after applied 3 hours and after applied 2 days at the rate of 1250-1562.5 Kg/ha can killed all of fishes. The LD₅₀ on fish is 687.5 Kg/ha. The rate of tobacco waste at 0-937.5 Kg/ha did not found affect on frog living and LD₅₀ was 1562.5 Kg/ha. The application on 0-3750 Kg/ha did not affect on crab and LD₅₀ is 5000 Kg/ha.

The application with different rates of tobacco waste affected on water pH, electrical conductivity and dissolved oxygen significantly. The increasing rate of tobacco waste affected on water pH, electrical conductivity increment but decreased dissolved oxygen.

3.1.2 Nicotine in tobacco waste is 1.90 ppm. The increasing rate of tobacco waste that can increase dissolved nicotine i.e. tobacco waste at 9375 Kg/ha has dissolved nicotine 1.86 ppm after 1 day and at 625 Kg/ha has dissolved nicotine 0.01 ppm. In the other hand, at the same rate of application was found maximum dissolved nicotine after applied 2 days and decreased at 3 days. There was not detected nicotine after applied 4, 7, 15 and 30 days at the rate of tobacco waste 625, 1562.5, 4687.5 and 9375 Kg/ha respectively. That can simulate nicotine prediction model as dissolved nicotine (ppm) = 0.002 (rate of application in Kg/ha) - 0.1037; $r^2 = 0.98$. Decomposition rate of tobacco waste also can simulated model as $0.0023 (\text{day after applied})^2 - 0.1006 (\text{day after applied}) + 0.9644$; $r^2 = 0.88$. Nicotine loss (ppm) = $-0.0409 (\text{day after applied}) + 0.5129$; $r^2 = 0.68$.

3.1.3 Temperature did not affect on nicotine in tobacco waste pellet after treated at 4° C and room temperature during 7 months.

3.1.4 Tobacco waste at the rate of 1562.5 Kg/ha has been tested for acute dermal on Wistar mouse LD₅₀ was more than 15,000 ml/Kg of body weight. The acute skin tested on white mouse (Sprague Dawley specie) after applied 2,000 ml/Kg of body weight 24 hours then observe abnormal symptom at ½, 1, 3 hours to 14 days. The weight of each mouse at 8 and 15 days were not different with control, mouse alive and not found abnormal symptom in visceral organ of gross pathology. Skin irritation tested was not found red and swell symptom on rabbit skin.

3.2 Green house trial

Tobacco waste can be reduced rice damaged by golden apple snail about 14%. After applied tobacco waste was increased pH, electrical conductivity, biological oxygen demand, chemical oxygen demand and nicotine in water but reduced dissolved oxygen. However, every parameter will be neutral at 3 days after applied.

The rate of application more than 625 Kg/ha can be improved rice yield and yield component due to tobacco waste contained of high nitrogen with 1.34% and high organic matter with 46.65%. There was not detected nicotine in rice grain. Before study, chemical soil properties were high available phosphorus, high exchangeable potassium but low organic matter. After application of tobacco waste can be improved organic matter, available phosphorus, exchangeable potassium, exchangeable calcium, exchangeable magnesium in soil.

In second pot trial, tobacco waste was mixed with chemical fertilizer that could be controlled golden apple snail as shown in Figure 1. The application of tobacco waste @ 1562.5 Kg/ha and mixed with chemical fertilizer grade 16-16-8 at 156.25 Kg/ha can be killed golden apple 69% after applied one day and 100% in 2 days. Similarly with tobacco waste @ 1562.5 Kg/ha can be killed golden apple snail 42% after one day and 100% in 15 days. Control treatment rice was damaged more than 50%. Tobacco waste application also increased electrical conductivity, pH, biological oxygen demand and chemical oxygen demand of water immediately after applied but will be neutral in 4 days. There was not detected dissolved nicotine in all treatments at various times. Tobacco waste mixed with chemical fertilizer was induced rice tiller more than chemical fertilizer and control. Tobacco waste can be increased organic matter, available phosphorus, exchangeable potassium, exchangeable calcium and exchangeable magnesium in soil.

3.3 Field trial

The best material which can control golden apple snail in field experiment was tea seed powder at the rate of 31.25 Kg/ha (Figure 2). Tea seed powder could kill golden apple snail 81% at first date after applied and 100% in 3 days. Simultaneously, tobacco waste can be killed golden apple snail 20%, 36% and 56% after used 625, 1562.5 and 3125 Kg/ha at first date and 100% in 2 days. Rice damage after tobacco waste treated was 15.19%, 15.41% and 18.62% in 3125, 1562.5 and 625 Kg/ha respectively and stop damage in two days (Figure 3). The application of tobacco waste @ 3125 Kg/ha can be detected nicotine in water after applied 1 hour with 0.09 ppm and dramatically increase in 2 days with 0.26 ppm. Tobacco waste application was increased electrical conductivity, biological oxygen demand and chemical oxygen demand in water. In the other hand, pH and dissolved oxygen in water were decreased. Tobacco waste can be improved soil fertility i.e. organic matter, available phosphorus, exchangeable potassium, exchangeable calcium and exchangeable magnesium.

In second field trial, different methods on golden apple snail controlling affected on mortality of snail significantly (Figure 4). Tea seed powder @ 15.625 Kg/ha was controlled at 8% on first date and 100% in 45 days. The application of tobacco waste @ 1562.5 Kg/ha or mixed with chemical fertilizer grade 16-16-8 @ 78.125 Kg/ha found rate of control at first date 5% and 6% respectively and 100% in 2 days. In control treatment, rice damaged by golden apple snail was 30.98%, 46.52% and 39.79% at 1, 2 and 3 weeks respectively. Tobacco waste @ 1562.5 Kg/ha mixed with chemical fertilizer grade 16-16-8 @ 78.125 Kg/ha was not found rice damage at 7 days (Figure 5). Tobacco waste application can be increased electrical conductivity, biological oxygen demand and chemical oxygen demand in water. In the other hand, pH and dissolved oxygen in water were decreased. Especially in tobacco waste @ 1562.5 Kg/ha or mixed with chemical fertilizer grade 16-16-8 @ 78.125 Kg/ha were dramatically changed. The application of tobacco waste @ 1562.5 Kg/ha or mixed with chemical fertilizer grade 16-16-8 @ 78.125 Kg/ha or mixed with chemical fertilizer grade 16-16-8 @ 156.25 Kg/ha could increase rice yield and yield component without nicotine detected. From this experiment we were found tobacco waste @ 1562.5 Kg/ha can killed all golden apple snail in 2 days. Simultaneously, tobacco waste can be increased organic matter, available phosphorus, exchangeable potassium, exchangeable calcium and exchangeable magnesium in soil.

4. Conclusion

The application of tobacco waste to control golden apple snail in paddy field was increased water pH and electrical conductivity and decreased dissolved oxygen. That caused by organic material in tobacco waste after applied into water was decomposed by aerobic microorganism and ionic bombard will be occurring (Raenwattana and Jenwanit, 2008).

Temperature was not affected on nicotine in tobacco waste pellet due to vacuum packaging. Nicotine is compound of nitrogen that can react with acid and produce nicotine salt in solid stage which soluble and volatilize at room temperature (Okamoto *et al.*, 2007).

Chemical properties of tobacco waste is pH 5.28, electrical conductivity 19.08 dS/m, organic matter 46.65%, C/N ratio 20.19, total nitrogen 1.34%, total phosphorus 0.34%, total potassium 0.24% and nicotine 1.8 ppm. The optimum rate of tobacco waste to control golden apple snail was 1562.5 Kg/ha. Iida *et al.* (1998) used tobacco waste which has nicotine 2.8% at the rate of 712.5 Kg/ha to killed snail.

Tobacco waste @ 1562.5 Kg/ha has LD₅₀ on acute dermal more than 15,000 ml/Kg of body weight. LD₅₀ on acute skin was more than 2,000 ml/Kg of body weight and irritate on skin was not found red symptom and swell on rabbit skin. Toxicity report LD₅₀ of nicotine was 50 mg/Kg of rat and 3 mg/Kg of mouse and in human was 40-60 mg/Kg (Kenneth, 2007).

That we recommended tobacco waste at the rate of 1562.5 Kg/ha can be killed all Golden Apple snails in 2 days without toxicity, water pollution and nicotine in rice grain. Tobacco waste can be increased soil fertility e.g. organic matter, available phosphorus, exchangeable potassium, exchangeable calcium and exchangeable magnesium.

Acknowledgement

This project could not success without funding from Thailand Tobacco Monopoly, Ministry of Finance.

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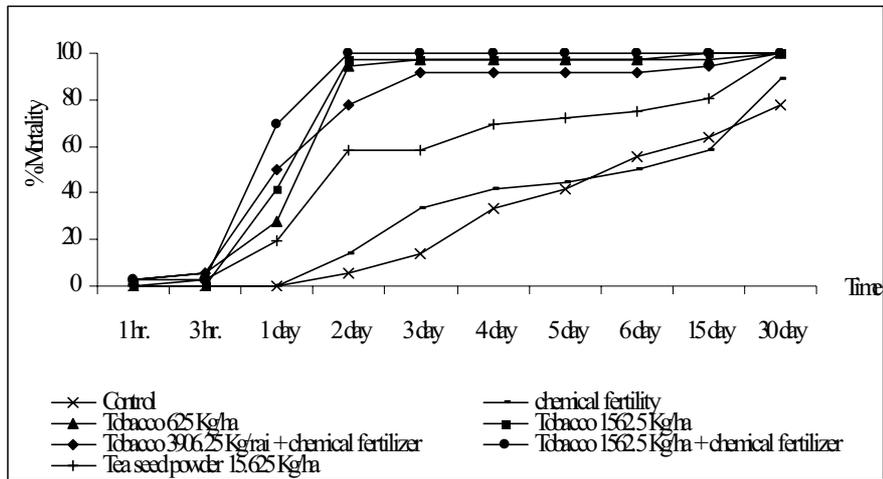


Figure 1. Golden apple snail mortality of each treatment in green house experiment

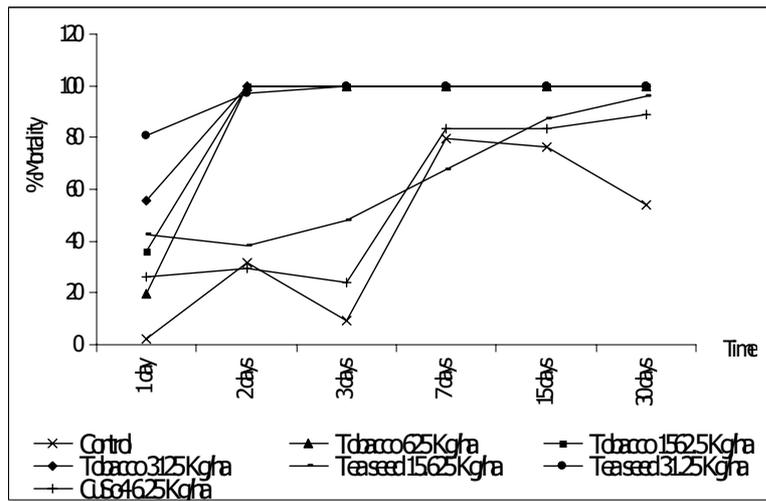


Figure 2. Mortality rate of golden apple snail in first field trial

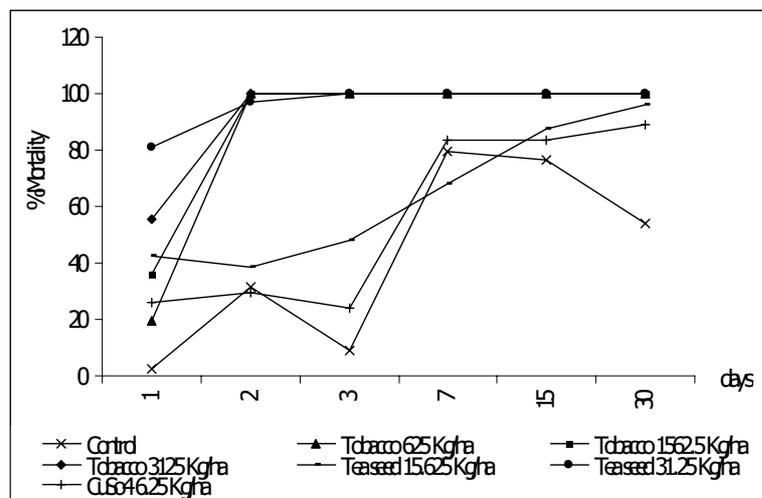


Figure 3. Rice damage rate in first field trial

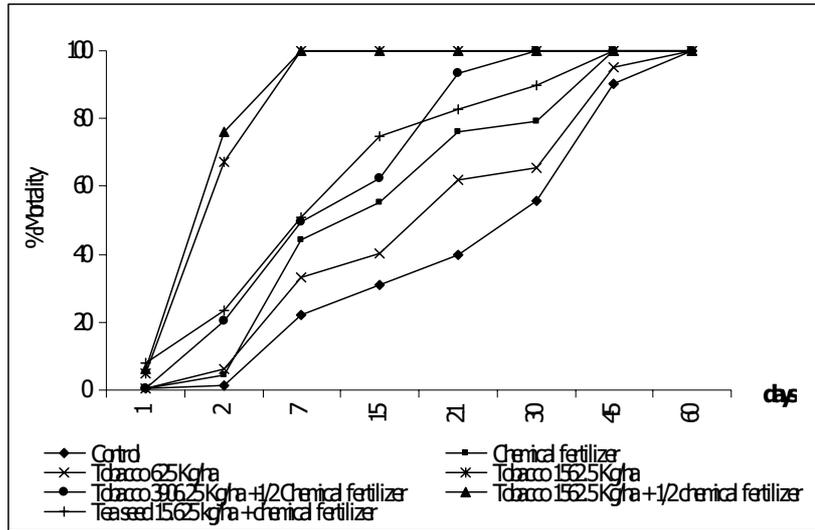


Figure 4. Mortality rate of golden apple snail in second field trial

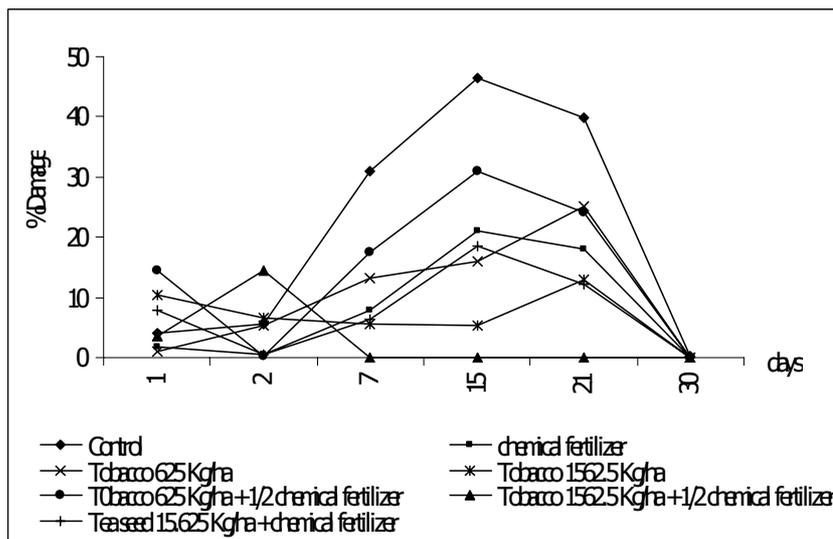


Figure 5. Rice damage rate in second field trial



Treating Infertile Milk Cows by Traditional Chinese Medicine

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Abstract

The infertility of milk cow is a world problem with high incidence rate. Reportedly, in the world dairy industry, the proportion of the infertile milk cow was about 15% in the total mature cows (Donald L B, 1978, P.309), and some others thought this number achieved about 30% according to the statistics (Bulman D C, 1980, P.177-188). In US, there are 12%~19% cows which are eliminated through selection because of sterility and breeding diseases every year, and this number achieves above 40% in all unqualified cows (Jiang, 1990, P.38-41). In China, the infertile rate of mature cows achieves above 25%, and the rate in some cattle farms with imperfect management and bad technical conditions even achieves above 40% (Jiang, 1990, P.38-41). According to the statistics, in a cattle village in the northeast, there were 62 infertile cows in 212 cows, and the infertile rate achieved 29.25%, and in the region of Jinan, Shandong, there were 342 mature cows in 5 collective cattle farms, one civil cattle farm and one cattle breeding village, and the amount of the infertile milk cow achieved 97, i.e. 28.36% of the total amount of mature cows. In the region of Shihezi in Xinjiang, the infertile rate was 22.5%, and the infertile rate in Fujian Province achieved above 10%. In recent years, according to Chinese traditional medicine, Chinese veterinary scientists have accumulated abundant experiences to prevent and cure the infertility of milk cow, and explored the pharmacology of the function of Chinese traditional medicine.

Keywords: Traditional Chinese Medicine, infertility

1. Clinical trial

1.1 About ovarian infertility

The ovarian infertility means female ovarian dysfunction, abnormal estrus cycle and low breeding ability, and it includes ovarian hypo-function (inactive ovary), durable lutein and ovarian cyst. The ovarian hypo-function generally includes delayed ovulation, out of heat in a long term in the post partum or obscure estrus representation. Though some females have obvious estrus, but they don't ovulate, so they have not the ovulation, and the form and character of the ovaries change little by the rectal examination, and the ovarian follicle or the lutein can not be touched, and sometimes one piece of very small luteal vestige can be touched in the one side of the ovary. When the pregnant lutein or the periodic lutein exceeds the normal time limit but still keeps the function, it is called as the durable lutein which can also excrete progesterone, restrain the growth of ovarian follicle, make the estrus cycle stop circulating and induce infertility. The ovarian cyst can be divided into follicular cysts and luteal cysts. The follicular cysts are formed by the epidermis denaturation, ovarian follicle connective tissue hyperplasia and thickening, oocyte death, follicular fluid unabsorbed or increased. The luteal cysts are formed by the epidermis luteinizing of the un-ovulated ovarian follicle wall. According to clinic symptoms, Chinese veterinaries generally divide it into the type of the insufficiency of yang and the type of qi depression to blood stasis, and adopt kidney in vigor-rating, promoting blood circulation by removing blood stasis, enriching the blood and regulating menstruation and other treatment methods. In recent years, Chinese veterinaries have acquired significant developments to apply Chinese traditional medicines and acupuncture to treat the ovarian infertility of milk cows.

(1) Song Dalu used self-made Chinese traditional medicine "promoting pregnant perfusate" to treat 220 cases with the inactive ovary infertility and 209 cases with the durable luteal infertility in five years, and the results indicates the medicine could promote the estrus and ovulation for ill cows effectively, and the estrus rate achieved 94.9%, and the fecundation rate achieved 79.4%.

(2) Jiangsu Academy of Agricultural Sciences used herba epimedii, actinolite, medlar, dodder, psoraleae, rehmannia glutinosa and other Chinese traditional medicines with function of kidney in Vigo-rating and many Chinese traditional medicines with the function of promoting blood circulation by removing blood stasis such as angelica, red paeonia and

motherwort to prepare the compound Xianyang soup which was used to treat the inactive ovary infertility and durable luteal infertility for milk cows, and for cows with the cold symptoms such as white mouth and hypercatharsis, added the nutgrass galingale and cinnamon to warm the kidney, and for the cows with durable lutein symptom, added Xishu, rhizome sparganii, safflower and peach kernel to increase the function of promoting blood circulation and removing blood stasis, and treated 297 ill cows, and the estrus rate achieved 94.6%, and the fecundation rate achieved 86.1% (Wang, 2005).

(3) Chinese Veterinary Research Institute of Chinese Academy of Agricultural Sciences took herba epimedii, actinolite, dodder, angelica, astragalus mongholicus and motherwort to affuse uterus to experimentally treat 141 cases, and the estrus amount was 123, and the pregnant amount was 116, and the conception rate was 82.27%.

(4) Ma Yanfeng et al used the compound epimedii pregnant promoting soup to treat the ill cows with inactive ovary, and the effective rate of estrus and ovulation achieved 80.8%, and the hybridization and fecundation rate was 80.0%, and the effective rate of estrus and ovulation for ill cows with ovary trophy achieved 50.0%, and the hybridization and fecundation rate was 50.0% (Jiang, 1999, 41-43).

(5) The Farming Veterinary Research Institute of Shandong Provincial Academy of Agricultural Sciences used the pregnant promoting formula (composed by motherwort, safflower, atractylodes macrocephala, nutgrass galingale rhizome, alisma orientale, peach kernel, rehmannia glutinosa, angelica and poria) to treat the un-estrus and abnormal estrus of cows combining with *Tristeronum compositum*, and for the 262 tested cows in two test group, the amount estrus cows after treatment achieved 234, and the estrus rate achieved 89.31%, and the fecundation amount after semen transmission for estrus cows achieved 127, and the fecundation rate achieved 54.27% (Ma, 2005).

(6) Wang Hongyan used drugs for regulating blood conditions to treat the infertile milk cows and obtain the curative effects which respectively were 87.5%, 100% and 76.9% (Wang, 2005).

(7) Yang Guolin et al used the point injection of Chinese traditional medicine injection (formula 1) and the uterus perfusion (formula 2) to treat ovarian infertile 232 cows, and the result indicated that the formula 2 (uterus perfusion) had better treatment effect, and this formula was used to treat 141 ill cows, and the estrus rate achieved 87.23%, and the pregnant rate achieved 82.27% (Yang, 1997, P.3-5).

(8) Wu Naike et al applied estrus promoting formula to treat cows with un-estrus or abnormal estrus, and the estrus rate achieved 89.31%, and the fecundation rate after semen transport achieved 54.27%, and the effect was good (Wu, 2000).

1.2 About endometritis

Chinese veterinary thought that the endometritis belonged to humid heat category, and it meant that poisons and dirty things entered into the uterus, which made the blood stasis stagnate in the uterus, and after a long time, stasis became into heat and the disease came on. For the treatment of endometritis, we should adopt the method from inside and outside, i.e. in the interior, regulating the blood conditions, and in the exterior, clearing wet heat and fighting bacterium and diminishing inflammation, and only the method used from inside and outside can acquire good and quick effects.

(1) "Motherwort biochemical formula (composed by 120g motherwort, 75g angelica, 30g Sichuan ligusticum wallichii, 30g peach kernel, 15g rhizome zingiberis and 15g liquorice)" was used for 78 ill cows with unclear lochia after 7 days, 75 cows were cured, and the cure rate achieved 96.15%, and it was used for 66 ill cows with unshed embryonic membrane, 62 cows were cured, and the cure rate achieved 93.94%, and it was used for 42 ill cows with endometritis, 40 cows were cured, and the cure rate was 95.24% (Yang, 2007, P.47-48).

(2) According to Li Chengmin's report, for the milk cows suffering endometritis, "pregnant treasure (composed by motherwort, scutellaria baicalensis, sanling, rhubarb and red paeonia)" was taken orally by one dosage (200g/cow) when the cow was in rut, and after 4~6 hours, transported semen to the cow, and if the milk cows was not pregnant, used the drug and transported semen again. For the ill cows with chronic catarrh endometritis, used 1~2 dosages drug with 5-days-interval, and after estrus occurred, used 1~2 dosages drug and transported semen after 4~6 hours. For the ill cows with chronic purulent endometritis, we used 2~3 dosages drug with the interval of 5~7 days for each dosage. In the test, there were 4953 ill cows where the ill cows with chronic catarrh endometritis were 3242, and the 1~2 dosages "pregnant treasure" were used, and availability rate was 100%, and the cure rate was 98.64%, and after treatment, there were 2823 cows became pregnant through semen transportation, and the fecundation rate achieved 88.27%, and all ill cows with concealed endometrium were cured, and after treatment, there were 495 cows to be pregnant through semen transportation, and the fecundation rate achieved 87.92%, and 905 cows in 1148 ill cows with chronic purulent endometritis were cured after taking 3 dosages drug, and the cure rate achieved 78.8%, and the fecundation rate achieved 63.48%.

(3) According to Xue Zhicheng's report, the soup composed by 70g phellodendron, 40g plantain, 35g pilosula, 50g poria, 50g atractylodes macrocephala, 50g comb grass, 50g motherwort, 40g cuttlebone, 30g liquorice, 40g safflower and 30g red paeonia decocted and affused one dosage one day could cure the ill cows with purulent endometritis.

(4) According to Li Dechang's report, took 120g kuh-seng, 50g rhizome coptidis, 80g scutellaria baicalensis, 80g yellow cypress and pounded to pieces, and decocted them with water for half hour and filtrated, and decocted the dregs for 20 minutes, combined two liquids and concentrated to 500mL, and to prevent deteriorating, seethe the liquid once one day, and the liquid could be used to cure the endometritis of cows, in 21 ill cows, 18 cows were cured after 1~3 dosage, and the cure rate was 85.7%, and except for 5 cows were washed out because of other diseases, 12 cows in other 13 ill cows became pregnant, and the pregnant rate achieved 92.3%.

(5) According to Zhu, Qisu's report, "uterus cleaning oil" was used to treat 33 cows with chronic endometritis, and 25 cows were cured, and the fecundation rate achieved 76%, and the average drug use times were only 1.74 times/cow, and the dosage was 30~50mL/cow once.

(6) The clearing uterus and diminishing inflammation suspensions (mainly composed by bomeol, sodium sulphate, yellow cypress and natural indigo) could be used to prevent the endometritis for post partum cows, in the 231 cows, the fecundation rate achieved 93.4%, and the fecundation rate in the first estrus term was 62.7%, which respectively were enhanced the fecundation rate for 7.0% and 10.2%, and the post partum initial estrus interval and the post partum another fecundation time respectively were enhanced by 20.1 days and 9 days. There were 1307 cows with endometritis to be treated, and the efficiency rate achieved 95.04%, and the fecundation after cure achieved 90.8%. And for 140 cows using Western medicines, the efficiency rate achieved 86.4%, and except for 9 cows washed out after cure, there were 112 cows to be in rut, and the fecundation rate achieved 80.4%, and the efficiency rate and the fecundation rate of the clearing uterus and diminishing inflammation suspensions respectively were enhanced for 8.54% and 10.4% (Yang, 2007, P.47-48).

2. Research of mechanism

At present, the mechanism research about treating the infertility by Chinese traditional medicines has achieved certain level. Jiang Zhaochun et al used "compound Xianyang soup" for 3 cows with durable lutein taken orally, and tested the progesterone content in the blood serum before and after treatment, and before the treatment, the progesterone contents respectively were 3mg/mL, 5.34mg/mL and 5.03mg/mL, and they were in rut respectively in the second, seventh and eighth day, and their progesterone contents all reduced to about 0.1mg/mL, and after estrus, the progesterone contents gradually increased to respectively 0.28mg/mL, 0.86mg/mL and 2.01mg/mL respectively in the second, fourth and fifth day after ovulation. After this formula was applied, the uterus angle exterior diameter, uterus and ovarian weight were obviously higher than the comparison group, Jiang also observed that the growth follicle of ovary increased, and the lutein occurred in the ovary, and the interstitial cells increased, and the endometrium was incrassate, and the uterine cavity became bigger, which indicated that this formula possessed the function to promoting oestrus, ovulation and pregnant for female animals. Chinese Veterinary Research Institute of Chinese Academy of Agricultural Sciences respectively implemented multiple tests including blood test, cell growth of cervices secretion, separation checkup of pathogeny microbe, drug sensitive test, pathology observation, microelement measurement, enzymoimmunoassay latex progesterone test, blood rheology observation, biopsy organization ultrastructure obervation and the function test of "clearing uterus liquid" to hysteromyoma for the "clearing uterus liquid" made by the institute, and studied the anti-inflammation and immunity mechanism function in the "clearing uterus liquid", the effective component separation and picking of the motherwort, and the endometritis animal model copy. The result indicated the various indexes of the cured cases could be rapidly or basically recover to normal levels. The "clearing uterus liquid" possessed many good functions such as anti-inflammation, immunity restriction, bacterium restriction, promoting blood circulation by removing blood stasis, and promoting shrinking uterus.

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Determining and Mapping Soil Nutrient Content Using Geostatistical Technique in a Durian Orchard in Malaysia

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The research is financed by the Universiti Putra Malaysia

Abstract

Soil nutrient are essential for crop growth. Spatial variability of nutrient can be occurred in various scales, between region, field or within field especially in variation in soil properties. Precision farming is a technology currently available for sustainable agriculture. This technology enables farm management is based on small-scale spatial variability of soil and crop parameters in the field. This study was carried out in a Durian Orchard at Bendang Man Agrotourism Project, Sik, Kedah, Malaysia. The objectives of this study are to determine and map soil nutrient content especially Nitrogen, Phosphorus and Potassium (NPK) variability in a durian orchard using geostatistical technique. The NPK was analyzed and mapped by Geostatistic Plus (GS++) to quantify the level of spatial nutrient available and predict nutrient values at unsampled location. Results indicated that NPK ranged from < 0.1 to 1.0 % (N), < 3 to > 45 ppm (P) and 0.8 to >1.4 cmol(+)/kg (K), respectively. Nutrient map showed that the area has less sufficient of N, while P and K were sufficient. This study revealed the potential and ability of geostatistical-variogram in determining and mapping soil nutrient content in a durian orchard. Furthermore NPK map can be used to apply fertilizer to an area, where less NPK content for efficient fertilizer management.

Keywords: Soil nutrient mapping, Precision farming, Soil nutrient analysis, Geostatistical technique, Durian orchard

1. Introduction

Precision farming is a management practice that has been enabled by Geospatial Information Technology (GIT) application and provides the framework within which arable managers can more accurately understand and control more precisely what happens on their farm (Mc Cauley *et al.*, 1997). Precision farming has become increasingly significant in the agricultural operations for the site-specific management. The management and manipulation of farming operation are vital decision-making process in improving crop productivity where there is a need to ensure efficiency in the management of agriculture. Information on soil properties in crop field is very important and useful for fertilizer requirement and also to the specific management of the crop and soil. The availability of nitrogen, phosphorus and potassium (NPK), whether in soils or plants is among of the most of the nutrient studied in precision farming concept (Malek *et al.*, 2007).

Therefore, the general objective of the study was to produce a spatial digital map of NPK variability in a fruit orchard in Bendang Man, Kedah, Malaysia. The specific objectives are to determine and map soil nutrient content especially Nitrogen, Phosphorus and Potassium (NPK) variability in a durian orchard using Geostatistics. Latter, this map will be used for efficient fertilizer management.

2. Methodology

2.1 Soil sampling and analysis

The soil samples were collected at Bendang Man Agrotourism Project, Sik, Kedah. The durian orchard farm has an area of 3.75 ha with latitude 5°51'29"N to 5°51'34"N and longitude 100°49'33"E to 100°49'42"E. A total of 122 soil samples were taken at the base of the standing durian trees. Soil samples were taken for NPK analysis and the location of specific soil samples were identified using a Differential Global Positioning System (DGPS). Each soil was taken at 2 different depths i.e. 0 – 15 cm (top soil) and 15 – 30 cm (subsoil). Samples were then kept in labeled plastic bags and brought back to the laboratory for further treatment and analyses.

The soil samples were air-dried and sieved to pass 2 mm mesh sieve. The analysis carried out were Total nitrogen (by Kjeldahl digestion procedure (Anon, 1995), Total phosphorus (determined by double acid and Mehlich (Ball *et al.*, 1979), and Exchangeable potassium (determined by ammonium acetate (Cole *et al.*, 1968). Semivariograms and kriged map produced from geostatistical software GS⁺, version 3.1. From semivariogram evaluation, the spatial variability of soil properties was generated. Descriptive statistic and variation of NPK nutrient status in soil was analyzed using SPSS tool. GIS software was used in producing map to show the spatial distribution of the NPK content. The classification of the nutrient content is classified according to Department of Agriculture Malaysia (1997).

3. Results and Discussion

The spatial variability map for total N [mol (+)/kg] in the study area is shown in Figure 1. The total N at 15 cm (topsoil) and 30 cm depth (subsoil) ranged between <0.1 and 0.3 %, <0.1 to 1.0 % respectively. According to Soil Survey Staff (1997), these ranges could be classified as very low and low (topsoil) and very low and high (subsoil). The total N in the soil comprises of two forms, namely organic N and inorganic N. However, inorganic N in the soil at any moment was only a small fraction of total N (Lindsay, 1979). About 82.68 % of the study area (0-15 cm) was classified as low level and 17.32 % was very low whereas, for 15-30 cm, 0.1 % was classified high, 2.21% (moderate), 36.50 % (low) and 61.19 % (very low). The low content of the total N in the area were due to denitrified, leached or volatilized N from soil. In general, total N under subsoil was higher than total N at topsoil. At the topsoil, about 17.32% of the study area especially at the central was found having very low N which might be due to high slope (more 10 %). Higher slope usually move away the N to downward direction. However, the variation of total N in the area was not significant due low ranges as shown in Table 1. Meanwhile, the availability of total N is presented in Table 2.

The total P content was more than 45 ppm for topsoil, while for subsoil the content was ranged between ±3 to ±45 ppm (Figure 2). ANOVA table for P was presented in Table 3, while Table 4 showed that availability of P for the topsoil can be classified as very high (99.9 %) to very low (0.1%), respectively. However, the P content for subsoil were less than topsoil ranging between low to moderate, high and very high, which represented 1.05%, 31.7%, 56.08% and 11.17% of the study area. The differences is due to fertilizer is usually applied to the topsoil and P does not leaching easily like NO₃. Moreover, topsoil received nutrients from tree leaves and organic matter by decomposition.

The southern and northern regions of the study area contain high P (25-45 ppm) because the slopes of the areas were low (less than 3%) and P content represent about 56.8 % of the total area. The central and far northern regions represent about 31.7% where P content was moderate (10-24 ppm). The slope in these areas was higher more than 10%. Hence, the higher content of P was occurred in low slope areas. A significant variation of P content was found between the two soil-depths and also in difference slope classes

The spatial content of the exchangeable K in the soil were illustrated in Figure 3. The map showed that the content of exchangeable K for topsoil and subsoil were classified into two classes as high and very high. Significant variations of K between two soil layers were as shown in Table 5. As classification the exchangeable K content were very high in both soil layers, more than 1.4 cmol(+)/kg, where topsoil represent 95.96% and subsoil 87.54% of total study area (Table 6). It noticed that the amount of Potassium in the topsoil much higher than subsoil, due to high organic matter present in the topsoil. Similarly, K was found higher in topsoil than subsoil because K was stable in the topsoil and decomposition of organic matter adds K content in the soil

4. Conclusion

From the study it can be concluded that by using Geostatistical –variogram analysis and spatial interpolation (kriging), there is possible to determine and mapped of NPK distribution in a Durian orchard at Bendang Man Agrotourism Project, Sik, Kedah, Malaysia. In fact, result showed that the NPK variability in soil were spatially ranging from two soil depth from <0.1 and 0.3% (N), <0.1 to 1.0% (N), <3 and >45 ppm (P), <3 to >45 ppm (P), 0.8 to >1.4 cmol(+)/kg (K), 0.8 to >1.4 cmol(+)/kg, respectively. Thus the NPK content in soil analysed in this study revealed that the Durian orchard is poor in N. Therefore it requires more N fertilizer inputs if durian productivity is to be increased. Meanwhile, P and K are sufficient for this site, and the management does not need to add more P and K fertilizers to the orchard.

Acknowledgements

The authors would like to express their sincere thanks to Mr. Wan Sulong Wan Drahman, Manager of Durian Orchard Bendang Man Project (DOBMP) for the permission to use DOBMP as the study site. Thanks are also due to the field Research Assistants of the Forest Geospatial Information and Survey Laboratory, Faculty of Forestry, Universiti Putra Malaysia (UPM) for the field data collection and soil chemical analysis.

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Table 1. Anova for Total Nitrogen

Source of Variation	SS	df	MS	F	P-value	F crit
Sampling locations	0.865459	60	0.014424	1.079648	0.383792	1.534314
Soil layers	0.013562	1	0.013562	1.015106	0.317729	4.001194
Error	0.801612	60	0.01336			
Total	1.680634	121				

Table 2. Classification of total N availability in the study area

Class	N%	Area (%)	
		Topsoil	Subsoil
High	0.6 – 1.0	0	0.1
Moderate	0.3 – 0.6	0	2.21
Low	0.1 – 0.3	82.68	36.5
Very Low	<0.1	17.32	61.19

Table 3. Anova for Phosphorus

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Sampling locations	116341.4	60	1939.024	1.938375	0.005703	1.534314
Soil layers	27462	1	27462	27.45281	0.000002	4.001194
Error	60020.08	60	1000.335			
Total	203823.5	121				

Table 4. Classification of P in the study area.

Class	P (ppm)	Area (%)	
		Topsoil	Subsoil
Very High	> 45	99.9	11.17
High	25 – 45	0	56.08
Moderate	10 – 24	0	31.7
Low	3 – 9	0	1.05
Very Low	< 3	0.1	0

Table 5. Anova for Exchangeable Potassium

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Rows	109.4631475	60	1.824386	6.679857	0.0000000	1.534314
Columns	8.57275082	1	8.572751	31.38851	0.0000007	4.001194
Error	16.38704918	60	0.273117			
Total	134.4229475	121				

Table 6. Classification of Exchangeable K in the study area.

Class	K [cmol(+)/kg]	Area (%)	
		Topsoil	Subsoil
Very High	> 1.4	95.96	87.54
High	0.8 – 1.4	4.04	12.46

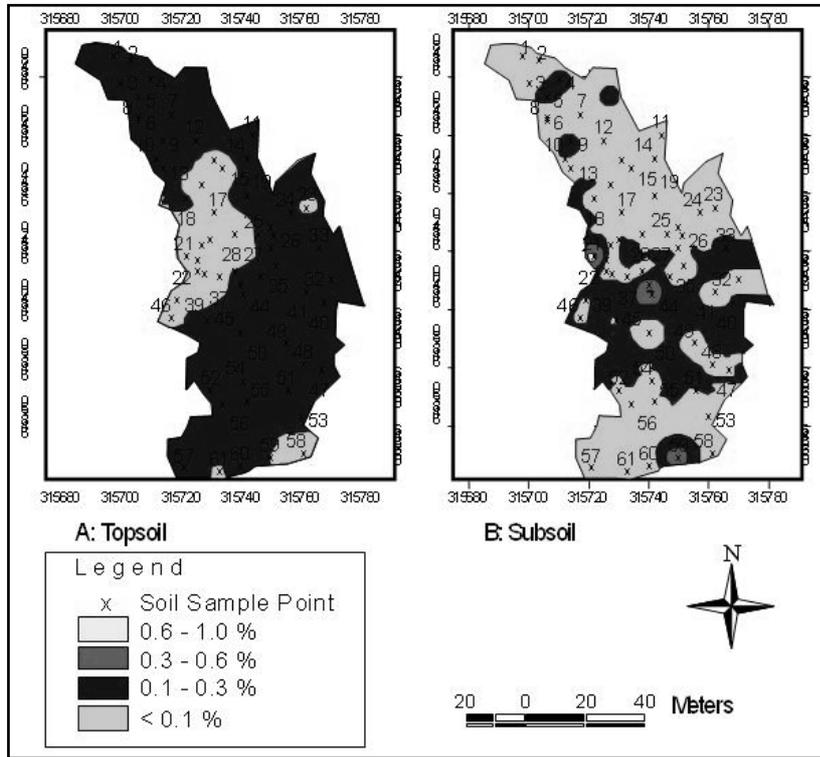


Figure 1. Spatial distribution of total N in soil

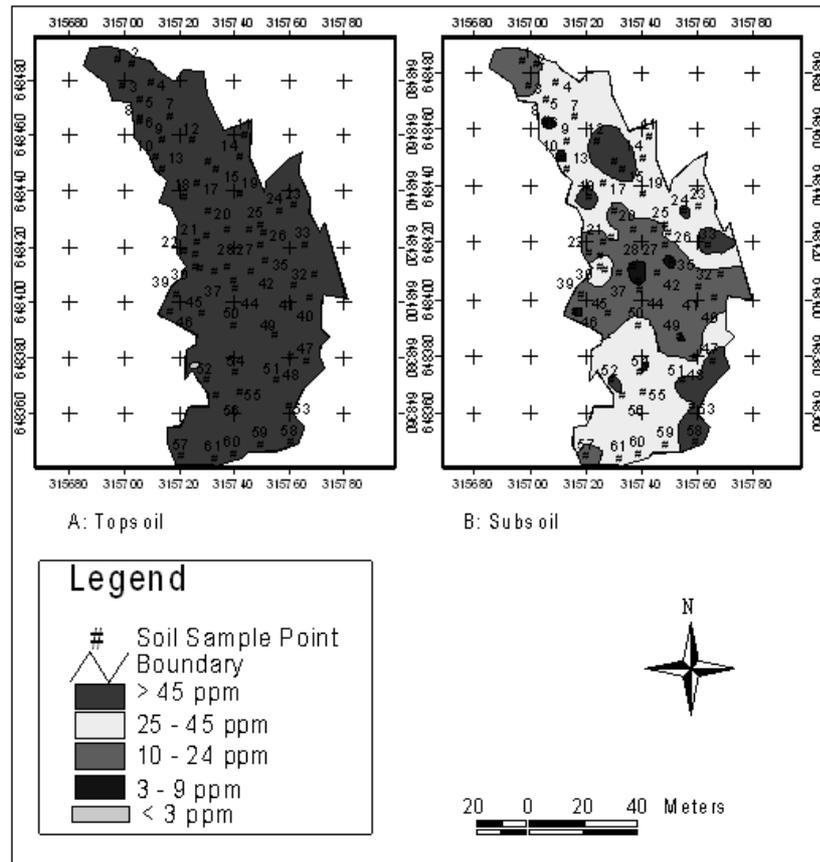


Figure 2. Spatial distribution of P in soil

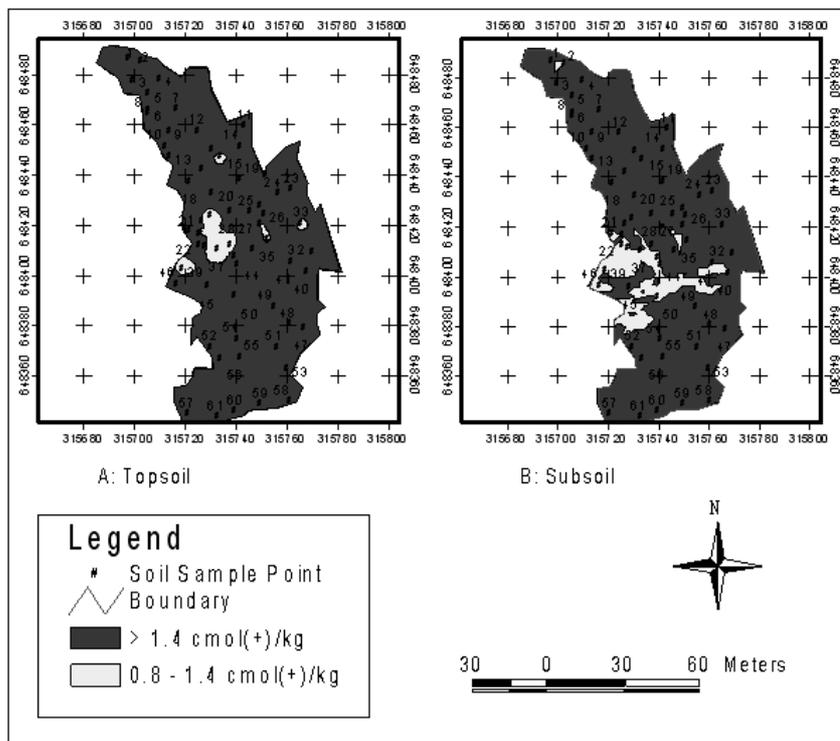


Figure 3. Spatial distribution of Exchangeable K in soil



Effects of Plant Growth Regulator on Endogenous Hormone Levels during the Period of the Red Globe Growth

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Abstract

Objective. The present study was aimed to elucidate the dynamic changes of endogenous hormone levels during the fruit growth and the changing regulation of endogenous hormone levels between normal and abnormal fruits.

Methods. After using exogenous GA₃ and GA₃+6-BA, endogenous hormone levels such as indoleacetic acid(IAA), cytokinin(ZR), abscisic acid(ABA) and gibberellin(GA₃) in normal fruits and rigid abnormal ones were determined by enzyme-linked immunosorbent assays(ELISA), respectively.

Results. The results indicated that the treatments of 20mg/kg GA₃ and the combination of 20mg/kg GA₃+10 mg/kg 6-BA had no remarkable effects on increasing average weight of berries where the vertical diameter of grape fruits was longer than the transverse diameter. Exogenous hormone GA₃ and GA₃+6-BA mixture also affected the endogenous hormone levels. The content of IAA, ABA and ZR was lower in the rigid abnormal fruits than that in the normal ones, respectively, and those levels during the rapid growth period were nearly in accordance with those during the slow growth period.

Conclusion. The fruit growth was closely related to the endogenous hormone levels.

Keywords: Plant growth regulator, Grape, Fruit growth, Endogenous hormone

1. Introduction

Applications of plant growth regulators have been focused by many researches in areas of plant physiology and nutriology(Pan, 1999, PP. 1-2, Amatjit, 2000, PP. 1-16). The regulators have been intensively and extensively applied for agriculture production, and played a vital role in the growth and development of plants. Along with the development of intensive cultivation of fruits, applications of regulators for controlling the growth of fruits have been progressively paid more attention (Ma, 1998, PP. 27-36).

Phytohormone, used as trace signal molecule in plants, has very important significance in regulating all kinds of growth processes and environmental responses, and meanwhile, made great contribution to the agricultural chemical control of crops, fruits and vegetables (Xu, 2006, PP. 433-442). Phytohormone mainly regulates the the plant physiological processes such as growth and development, organ formation and so on. The dynamic changes of endogenous hormone levels during the period of fruit growth have been investigated by many researchers. Meng et al(2005, PP. 6-10) studied

that the relationship between endogenous phytohormones and preharvest fruit drop of apples and found that a higher level of ABA/IAA caused fruit drop seriously. Xia et al (2000, PP. 6-10) found that the fruit growth was regulated by GA and IAA at earlier stage, while ABA did at later stage. Therefore, it would be of great significance for elucidating the time control of fruit growth by endogenous hormones and giving reasonable insight into production practice that better ordered control was undertaken to coordinate the growth and development.

Effects of exogenous plant growth regulators have been investigated more on seedless varieties but relatively less on seedy varieties. Several results showed that GA could promote the increment of grape fruit grains (Kim, 1991, PP. 199-205, Retamales, 1993, PP. 89-94), but some other results showed no effects on seedy fruits (Nosukchal, 1984, PP. 52-59). Though exogenous plant growth regulators could regulate the growth and development of fruits effectively, they could also cause some problems in practice such as rigid and abnormal fruit. Taken together, in the present paper, through the treatment of exogenous GA₃ or GA₃+6-BA mixture applied to red globe grape, 4 hormone levels were investigated in order to observe their effects on the growth and development of fruits and the internal cause of the rigid and abnormal fruits in hormone level.

2. Materials and methods

2.1 Plant material and treatment

Six-year-old trees of the Red Globe (*V. vinifera* L. Cv.) growing at the million mu production base of the red globe grape, 83 Corps, N 5 Division, Xinjiang, were used in this experiment. Grape trees in the garden with strong tree vigor and complete structure have no diseases and pests and similar growth. Tree stands are all small frame with the planting spacing of 1×4m. Supplemental irrigation was provided and standard fertilization practices were followed. Grape trees of similar age and vigor were randomly selected, 4 trees as a block, 3 times replicate, 12 trees for trial altogether. Fruit clusters were processed with micro-jet of GA₃(A) 20 mg/kg and the combination of 20 mg/kg GA₃ +10 mg/kg 6-BA(B) 5 days before (22nd May) and 3 days after (2nd June) blossom respectively, water treatment used as control in the whole trial.

2.2 Methods

2.2.1 Sampling and pretreatment

During the period from 23 days after blossom (19th June, 2007) to fruit maturation (28th August, 2007), normal and rigid abnormal fruits appearing after treatment was randomly sampled once 7 days, 20 fruit grains altogether collected from the upper, central and lower part of the fruit clusters. The samples were bagged with silver paper, quick-frozen by liquid nitrogen, and then stored at -80°C for further investigation.

2.2.2 Separation, purification and determination of endogenous hormone

Accurate 1g sample was grinded into homogenates under ice bath and weak light. Resultant extraction was centrifuged. The supernatant was separated by C-18 solid phase extraction column, transferred into 5ml plastic centrifuge tube, dried by nitrogen gas, eliminated methanol from extractions and then diluted into a constant volume by sample diluents. The endogenous hormone levels in fruits were determined by ELISA as described by He et al (1993, PP. 60-68).

ELISA kit was kindly provided by China Agriculture University as a present. Discarded peels and cores, IAA, ZR, ABA and GA₃ levels were investigated in the flesh of grapes.

2.2.3 Effects of plant growth regulators on the growth of red globe grape

18 fruit grains were marked, 3 ones from the upper, central and lower part of fruit cluster each treatment. The longitudinal and transect diameter of fruits was measured by vernier caliper every 7 days from 19th June on, and accurate to 0.1 mm.

2.2.4 Effects of plant growth regulators on the quality of red globe grape

18 fruit grains were randomly selected to measure fresh weight and soluble solids. Fresh weight was determined by a balance with the precision of 0.1g while soluble solids by a portable sugar analyzer with the precision of 0.2%.

3. Results and analysis

3.1 Effects of plant growth regulators on the growth dynamics of red globe grape

The growth dynamic changes of grape berry was depicted in Figure 1 and 2 and appeared a double S-type curve, after enlargement treatment which could not affect the growth of fruits. The longitudinal diameters of normal fruits were all higher than transect diameter while the difference of longitudinal diameter between B and CK was significantly higher A and CK after both A and B treatment as the same period. During the whole growth process, the longitudinal diameter in A and B was all higher than CK. In mature period, the difference of longitudinal diameter in A and CK was gradually decreased while transect diameter kept stable, and the growth of fruits became slower; the difference of longitudinal diameter in B and CK was significant while that of transect diameter was not significant. The average transects and

longitudinal diameter increased by 0.11cm and 0.10cm in A and decreased by 0.07cm and increased by 0.10cm compared to the CK in the whole period, respectively. The average transect and longitudinal diameter increased by 0.12cm and 0.11cm in B and increased by 0.01cm and 0.20cm compared to the CK in the whole period, respectively. Results showed that fruits were characterized by elongation of grains, oval shape and increment of fruit shape indices when a treatment was involved. As for the rigid abnormal fruits, there was no significance between the transect and longitudinal diameter and the two diameters had almost no increment and were significantly lower than CK (Figure 1 and 2).

3.2 Effects of plant growth regulators on the quality of red globe grape

The increment was significant for the longitudinal diameter while not obvious for transect after the treatment of Plant Growth Regulator. Fresh weight of single and the biggest grain and soluble solid content increased compared to CK. Through analysis of variance with mean separation by Duncan's multiplier range test, there was no great significance in fresh weight of single grain or soluble solid content, but great significance in fresh weight of the biggest grains between B and CK (Table 1).

3.3 Effects of plant growth regulators on endogenous hormone levels in fruits

IAA level was higher in flesh in early growth period. During the whole growth process of normal fruits, IAA level was higher in A than that in B and CK, attained a peak value 23 days after blossom with the content of 1548.90 ng/g FW and then decreased rapidly to a stable level. IAA level was higher in B than that in CK 23 days after blossom, decreased gradually, attained a valley value on 26th July, and then increased gradually to a stable level. IAA content of rigid abnormal fruit was all in an extremely low level in both A and B, where fruits were in a rigid, stagnant stage all the time during the whole growth process (Figure 3).

As for normal fruits, ZR content in A, B and CK was at a higher level which was 1962.87 ng/g FW, 4735.98 ng/g FW and 1283.19 ng/g FW, respectively, in early growth period, and then decreased gradually to a lower level, almost undetectable; As for abnormal fruits, ZA level in A and B was significantly lower than that in CK, and since 23 days after blossom, there was no great difference in ZA level (Figure 4).

ABA level of normal fruits in A, B and CK all attained a peak value 23 days after blossom which was 218.86 ng/g FW, 1079.77 ng/g FW and 890.96 ng/g FW, respectively. Subsequently, ABA level attained a peak value again 58 days after blossom. ABA level increased gradually in late growth period (79 days after blossom). As for abnormal fruits, ABA level was lower during the period of 23 days after blossom, and then increased gradually. ABA level attained a peak value of 926.99 ng/g FW in B on 9th July while 638.84 ng/g FW in A on 25th July, a little later compared to B. Subsequently, ABA level of abnormal and normal in A, B and CK tended to be basically consistent (Figure 5).

GA₃ levels in normal fruits attained peak values of 1424.16 ng/g FW and 1382.96 ng/g FW on 8th and 15th August in A and B during the growth process, respectively, which were higher than that in CK. As for abnormal fruits, GA₃ level was lower in the whole process, and attained peak values of 559.88 ng/g FW and 921.48 ng/g FW 23 days after blossom in A and later in B, respectively. GA₃ level was higher in B than that in A, but attained the peak value later. Subsequently, GA₃ level of tended to be basically consistent with normal fruits (Figure 6).

4. Discussions

Plant growth regulators applied in production promoted growth through boosting cell division and increasing cell volume, which ascribed to comprehensive effects of many hormones. In the present paper, results showed that after treatment of grape swelling agents, the transect and longitudinal diameter of normal fruits increased rapidly, and longitudinal diameter was larger than transect one. Therefore, indices of fruit shape increased which indicated that swelling agents could significantly enhance the longitudinal growth of grape flesh cell, and was not in accordance with the results of Zhao et al (1998, PP. 28-29) and Dai et al (2002) while in line with Wan et al (2004, PP. 13-14) and Wang et al (2004). Meanwhile, as seen from the growth of abnormal fruits, fruits was under stagnation state during the first rapid swelling period and had no changes with no significant difference between the longitudinal and transect diameter in the early stage of growth. IAA, ABA and ZR levels were higher in normal fruits than abnormal ones possibly due to their effects on the regulation of fruits in early growth stage, which was similar to the study of Tao et al (1994, PP. 35-40) on kiwifruit.

Seeds of development was the source of plant growth hormones for normal growth of fruits(Cui, 2006). Through the treatment of A and B, IAA level was higher than CK when GA₃ was involved which indicated that GA could enhance the synthesis of IAA and conformed to the previous studies(Fan, 2004, PP. 728-733, Uilger, 2004, PP. 89-95, Yu, 2003, PP. 125-129). Gong speculated that the volume growth and embryo sac development of some fruits were strongly inhibited, and fruits were under a stage of young fruit in a long term with no edible values, called small green grains, namely rigid abnormal fruits in the present paper. IAA level was lower in abnormal fruits that that in normal ones. Huang et al (1994, PP. 125-129) also speculated that fruit growth depended on the seeds of normal growth, and the outdision of all hormones in seeds stimulated the growth of flesh tissues all around. Therefore, treatments of exogenous

hormones played a vital role in the growth and development of fruits. On the other hands, ovaries grew rapidly once after pollination and fertilization, or otherwise deteriorated, which was due to that embryo and endosperm became the centre of producing IAA after fertilization.

In the present paper, both A and B have promotion effects on ZR, especially in the early stage of fruit growth. ZA level in B was far higher than that in A and CK which conformed that cytokinins mainly produced young fruits. This might be related to the synergistic effects of IAA, for cytokinins could coordinate fruit sink to distribute nutrients in the presence of IAA. As for rigid abnormal fruits, ZA level was consistent and lower compared to CK in the whole process of growth, which indicated that cytokinins was highly associated with the promotion of cell division and extension and the growth and development of fruits, similar to the report of Zhou et al who found that exuberant cell division was closely related to CTK level in the early stage of seedless Litchi fruit growth (Zhou, 1998, PP. 236-240).

High level of ABA in the early stage of grape fruit growth might have stimulatory effect on cell division and meristem activities. ABA attained a second peak level about 58 days after blossom, which was probably associated with transport of substances in cell, sugar accumulation and initiation of fruit afterripening in grape berries. IAA and GA₃ level was low and stable while ABA increased again about 79 days after blossom. This increment of ABA played a more important role in the declining of GA₃/ABA than that of GA₃, which showed that a new balance mainly attributed to ABA among endogenous hormones was confirmed and determined the physiological state and storage characteristics of grapes postharvest. Beeruter found that ABA level was higher in the young fruits growing fast and lower in the fruits growing slowly (1983, PP. 737-743). It was obvious that increment of ABA was beneficial to the promotion of assimilates absorption of metabolic pool cells in line with that ABA level was lower than CK in the early stage of rigid abnormal fruits. Moreover, ABA level was high in grape fruits at high temperature in summer, might due to the functions of ABA on promotion of stomatal closure, reduction of transpiration rate, enhancement of dormancy and high temperature and drought resistance.

In A and B, GA₃ level in normal fruits was obviously higher than that in CK. GA₃ level in abnormal fruits was all higher than that in normal ones 23 days after blossom, decreased subsequently during the period of fruit growth and then became lower than that in normal fruits, which indicated that GA₃ was closely associated with the growth and development of fruits. Ma et al (2007) speculated that effects of GA on the fruit growth was exerted through enhancing the synthesis of IAA, promoting fibrovascular growth and nutrient distribution in combination with IAA, and boosting the enlargement of flesh cells. Hu et al (1997, PP. 36-38) considered that exogenous GA treatment could significantly enhanced the allocation of ¹⁴C photosynthetic product to fruit cluster. Exogenous GA could enhance amylase activities of many plants, resulting in degradation of storage starch in order to provide abundant energy substrates and bond structure, and thus promoted the growth and development of fruits. Exogenous GA could also cause parthenocarpy instead of seeds. It was reported that GA level in ovaries of parthenocarpical grapes and persimmon after blossom was higher than that in seeded fruits (Ma, 1998, PP. 27-36), and was in accordance with our research, which was maybe one causative factor of rigid abnormal fruits.

5. Conclusions

Investigation indicated that the involvement of GA₃ could result in rigid abnormal fruits. Both exogenous GA₃ and GA₃+6-BA mixture could enhance endogenous hormone levels such as IAA, ZA, ABA and GA₃ in normal fruits but had no significant effects on rigid abnormal ones. The causative factors for rigid abnormal fruits in our observation could be listed as follows;

(1) Maybe associate with pollination and fertilization.

(2) As seen from hormone level, IAA, ABA and ZR level which promoted cell division and enlargement in rigid abnormal fruits was lower than that in CK, and was in a stable and lag state which was inadequate for normal growth of fruits in the whole process of growth. Herein, rigid abnormal fruits emerged chronically in the stage of young fruit and did not fall.

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Table 1. Effects of plant growth regulators on the quality of Red Globe berry

	longitudinal diameter	Transect diameter	Single grain weight	Biggest grain weight	TSS
GA ₃	2.69ABb	2.37Aa	10.11Aa	16.23Aa	17.94Aa
GA ₃ +6-BA	2.84Aa	2.50Aa	9.86Aa	18.33Ab	18.29 Aa
CK	2.57Bb	2.46Aa	9.76Aa	15.77 Ab	17.67 Aa

Note: Capital and small letter represent significant at $p \leq 0.01$ and $p \leq 0.05$, respectively

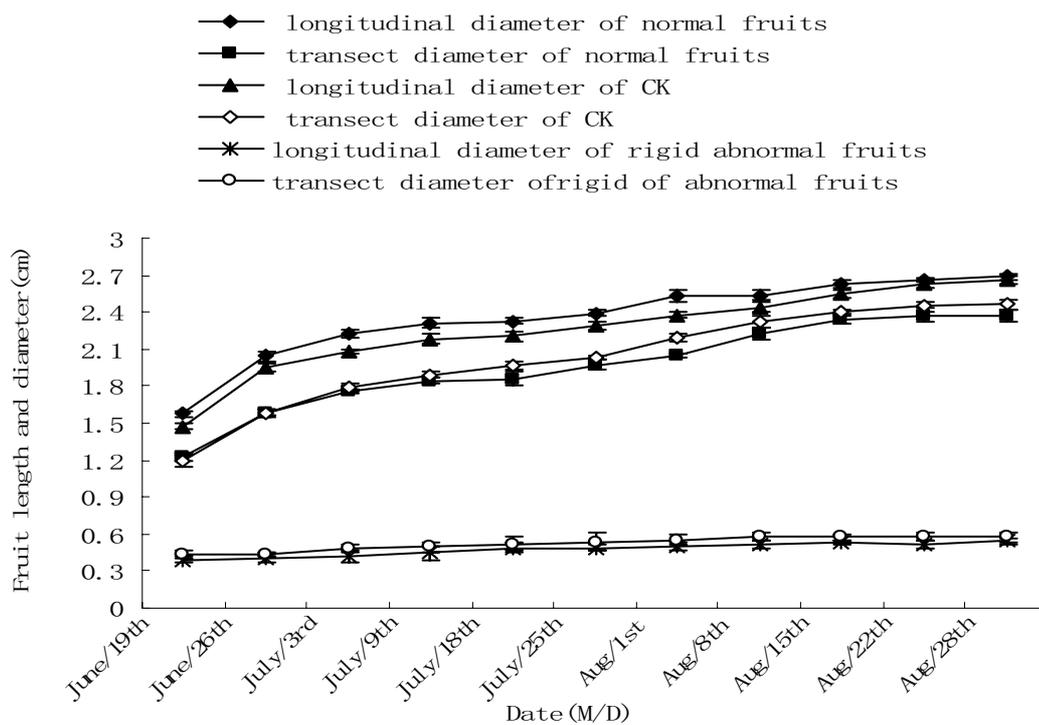
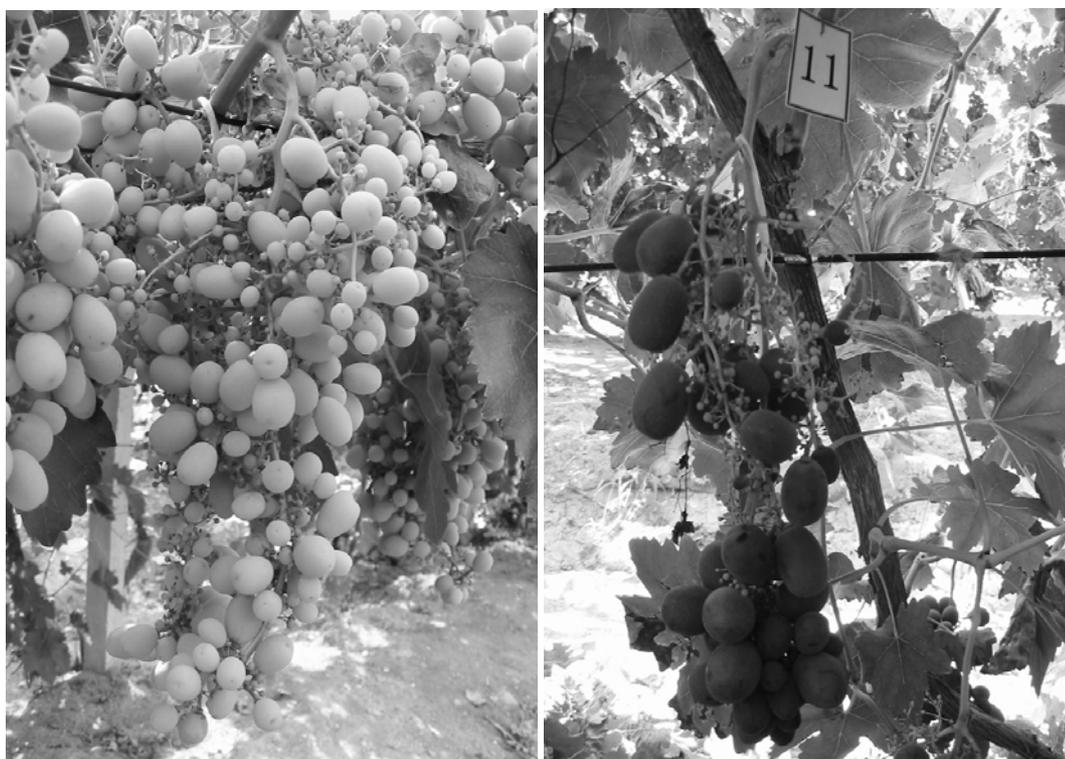


Figure 1. Effects of GA3 treatment on the transect and longitudinal diameter of normal fruit and abnormal ones

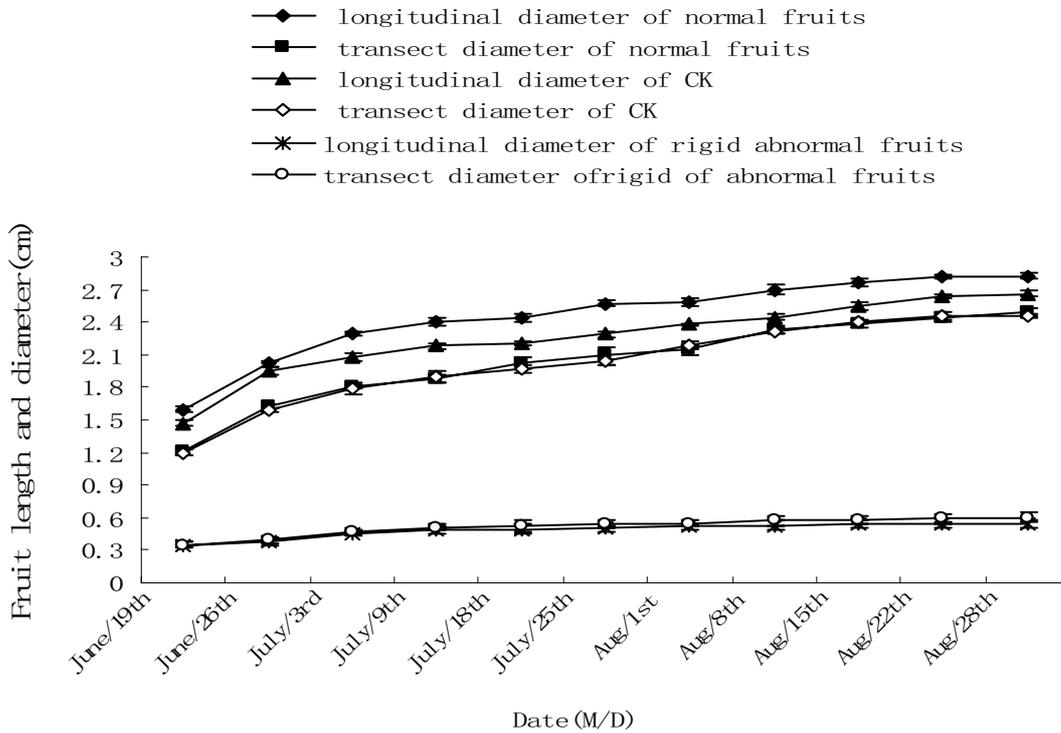


Figure 2. Effects of GA₃+6-BA treatment on the transect and longitudinal diameter of normal fruit and abnormal ones

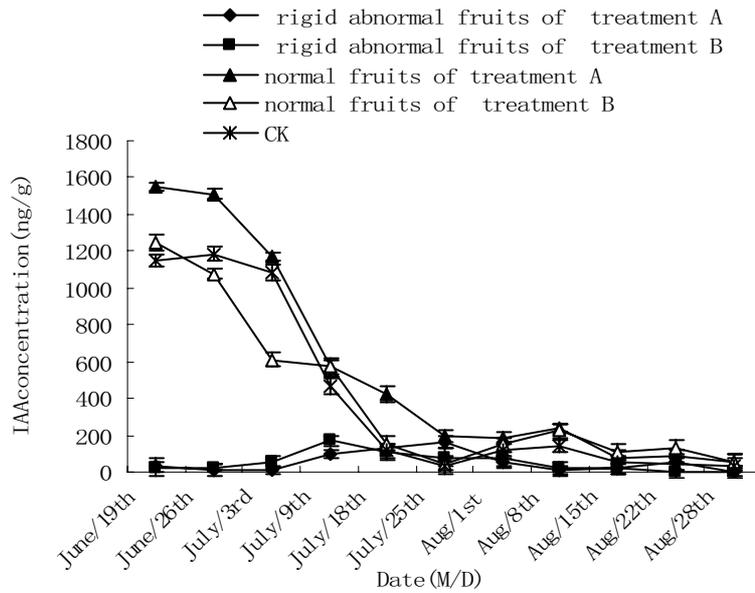


Figure 3. IAA level in fruits after A and B

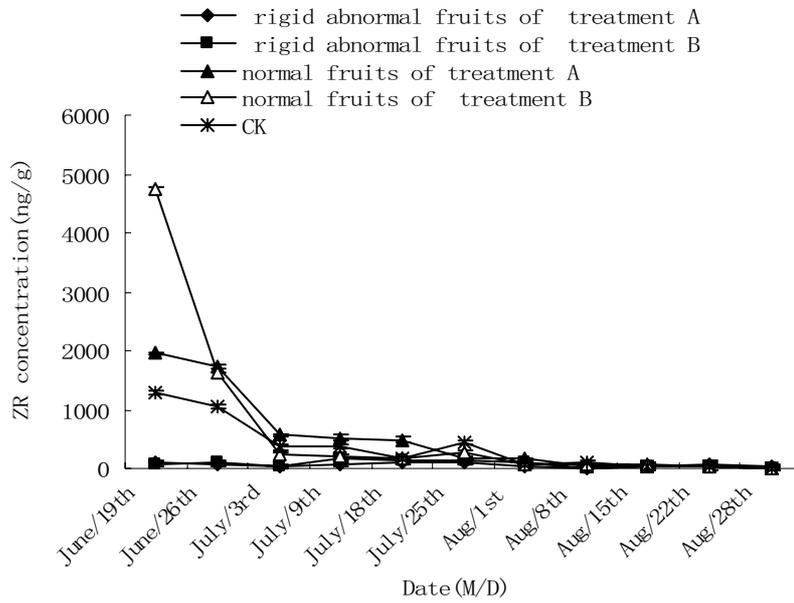


Figure 4. ZR level in the fruit after A and B

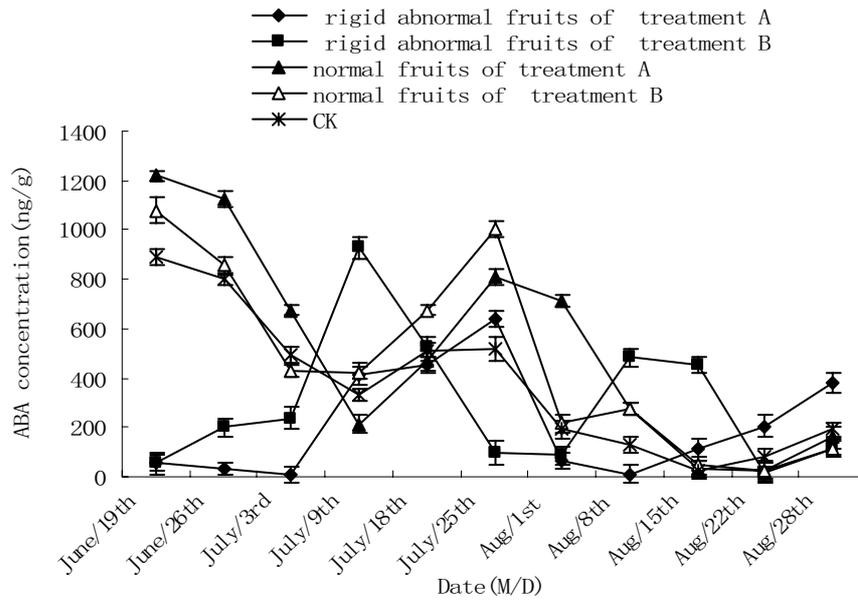


Figure 5. ABA level in fruits after A and B

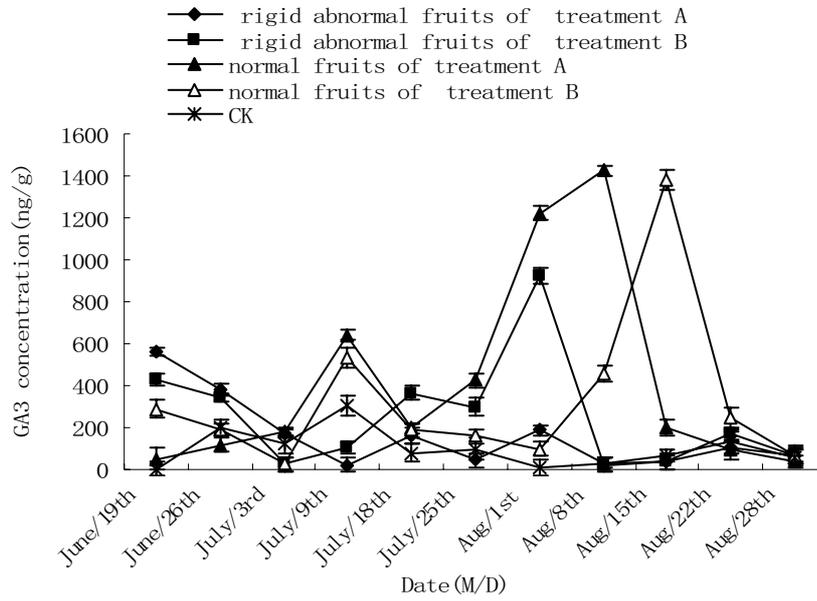


Figure 6. GA₃ level in fruits after A and B



Study Effects of Planting Methods and Tank Mixed Herbicides on Weeds Controlling and Wheat Yield

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The research is financed by University of Tehran (Sponsoring information)

Abstract

The aim of integrated weed management (IWM) is to use of a combination of different practices to maintain weed densities at manageable levels. A field experiment was conducted at Agricultural College, Tehran of University, in Karaj city 2005-2006 by planting wheat, to investigate the response of planting methods and tank mixed herbicides. The experiment was laid out using a split plot arrangement, in randomized complete block design with three replications. Methods of planting were assigned to the main plots; while tank mixed herbicides were kept in the sub-plots. The sub-plot size measured $4.5 \times 4.5 \text{ m}^2$. Row to row distance was kept at 30 cm. Data were recorded on weed density m^{-2} , plant height (cm), spike length (cm), Number of spikes m^{-2} , Number of grains spike $^{-1}$, 1000 grain weight (g), biological yield (kg ha^{-1}), and grain yield (kg ha^{-1}). The data for individual traits were subjected to the ANOVA technique and significant means were separated by the LSD test. The analysis of the data showed that methods of sowing were statistically significant for plant height, No. of grains spike $^{-1}$, 1000-grain weight and biological yield. The herbicides were statistically significant for all the parameters investigated except No. of grains spike $^{-1}$, while the interaction of methods of planting with herbicides could not reach the level of significance in any of the traits examined. Among the methods of planting, line sowing was the best followed by line + broadcast sowing. The herbicide mixtures controlled mixed stands of broadleaf and grassy weeds to the tune of 65 to 74% with a consequent increase in grain yield from 58-107%. Buctril-M + Topik 15 WP, 2,4-D + Puma Super 75 EW and Topik 15 WP were segregated as the top scoring applications by increasing yield to the extent of 107, 104 and 101 %, respectively over the weedy check.

Keywords: Planting, Mix- herbicides, Wheat, Weed control, Grain yield

1. Introduction

Herbicides have increasingly become a key component of weed management programs in developed countries, one being the reason accounts for increased crop yields in these countries. Nearly 61% of total herbicides used worldwide were applied in North America and Europe in 2004, with 15% in Asian countries (Anonymous 2004). Arable lands of Iran received a total amount of 11.1 tons/ha herbicides in 2006, over 5.5 tons/ha being applied in wheat farms (Personal comm. Crop Protection Organization, Iran). Wheat (*Triticum aestivium* L.) belongs to the family Poaceae and is an annual self-pollinated, photoperiodically long day grass. Like other grasses wheat produces tillers depending upon soil fertility and micro- and macro-environment. Wheat is the most important staple food crop for the whole world. Its cultivation is simple and adaptable to a varied soil and climatic conditions. It is also known as the king of cereals. Besides food, wheat is used for livestock and poultry feeds. A large population of the world consumes wheat in a number of ways. Wheat culture both in Tehran State, as well as in the whole country is the backbone of the whole agricultural system. In Iran, wheat was grown on an area of 4.76 million hectares with a grain production of 14.07 million tons, during 2004-2005. The mean country and provincial productions are limited to 3150 kg and 1564 kg ha⁻¹, respectively (.Anonymous, 2004; Baghestani et al , 2005).

During the recent years wheat production has exceeded the requirement of the nation and subsequently the nation has entered into the international wheat export trade. The factors responsible for luxuriant growth and production probably have been the timely availability of fertilizers, higher support prices of wheat and accelerated use of herbicides like Puma super and Topic by the growers. The tempo however, needs to be sustained rather further accelerated, as still there exists a gap between the actual and potential yield of the crop at the farmers' fields. There are several reasons for this gap but the worst one is weed competition with the wheat crop in the field. The weeds use the soil fertility, available moisture, solar radiation and space with crop plants and result in yield reduction. Moreover, the wheat grains contaminated with weed seeds fetch lower prices. As, the nation has entered the international export market, the production of cleaner wheat grains is essential for competition in the international trade. Pervaiz and Quazi (1992) have reported nearly 17.25 % losses caused to the wheat crop by weeds. The losses on annual basis amount to more than Rs. 28 billion at the national level and Rs. 2.00 billion in Iran center (Hassan and Marwat, 2001). Similarly the efficiency of feonaxprop in controlling *Sorghum halepense* L. (johnsongrass) decreased when mixed with 2,4-D or MCPA (Mueller et al. 1989).

The weeds competitive with wheat crop in Tehran State include *Avena fatua*, *Phalaris minor*, *Anagallis avensis*, *Poa annua*, *Cirsium arvense*, *Convolvulus arvensis*, *Ammi visnaga*, *Chenopodium album*, *Fumaria polymorpha*, *Carthamus oxycantha*, *Euphorbia helioscopia*, *Medicago denticulata*, *Melilotus indica*, *Silybum marianum*, *Galium aparine* and *Rumex crispus*. Wheat can be sown by different methods viz., drilling in lines, cross sowing or broadcasting. Each method of planting has a varying impact on weed competition. For wheat cultivation the best method is line and line + broadcast sowing (Gogoi and Kalita, 1995; Code and Donaldson, 1996), because of equidistant spacing of wheat, the wheat is better competitive with weeds. Weed control has been practiced since the time immemorial by manual labour (weeding) or animal drawn implements, but these practices were laborious, tiresome and expensive due to increasing cost of labour. The growing mechanization of farm operations and ever increasing labour wages have stimulated interest in the use of chemical weed control. Chemical weed control is the easiest and most successful alternative method. Although different reports are available on the efficacy of different herbicides in wheat (Ashrafi 2006, Baghestani et al, 2005; Mohibullah and Ali , 1974; Gill and Walia, 1979; Praczyk, et al. 1995; Balyan et al., 1983; Porwal and Gupta (1987); Azad et al. 1997; Khan et al., 1999; Khan et al., 2001; Khan et al., 2002; Hashim et al., 2002; Qureshi et al. 2002; Zand et al. 2007), the herbicide use in Iran is not widely practiced as in the agriculturally advanced nations. The interest around the testing of graminicides (Walia et al., 1998; Ormeno and Diaz, 1998; Brar et al., 1999a; Brar et al. 1999b) indicates the problems posed by grasses. Tank mixing of herbicides is practiced for attaining synergism but, antagonism is also not uncommon in such a mixing (Ashrafi 2006, Williams, 1984; Deschamps, et al., 1990; Augero-Alverde and Appleby, 1991; Augero-Alverde, et al., 1991; Pandey and Singh, 1994; Brar et al., 1999b). Whereas, the studies of Panwar et al. (1996) and Khan et al. (2002) showed synergistic response on combined use of herbicides. Tribenuron, 2,4-D + MCPA and bromoxynil have been widely used for broad-leaved weeds control (Zand et al. 2007).

The instant studies were undertaken to evaluate the efficacy of different herbicides alone and in mixture on dynamics of weeds in wheat planted with different methods with these objectives a) to find out the most economical tank mixture of herbicides for the control of weeds in wheat crop b) to figure out the most suitable planting method for wheat cultivation and c) to evaluate the response of wheat to different planting methods and tank mixture of herbicides.

2. Methods and materials

A field experiment was conducted at Agricultural College, Tehran of University, in Karaj city 2005-2006 to investigate the efficacy of some herbicide mixtures on grassy and broadleaf weeds and their consequent effect on wheat crop. The experiment was laid out in a split plot design with three replications. In a well-prepared soil, the basal dose of NPK was

applied. All the phosphorous and potash were applied at the time of planting while, nitrogen was applied in two split doses. First half with the first irrigation and the remaining half at the early boot leaf stage. Methods of sowing (broadcast, line sowing and line + Broadcast) were assigned to the main plots, while ten herbicides detailed below (Table-1) were kept in the subplots. Each sub-plot size measured $4.5 \times 1.5 \text{ m}^2$. Row to row distance was kept at 30 cm. Wheat variety Zarrin was sown on the 11th November, 2005 at the rate of 100 kg ha^{-1} with broad cast and seed drill and broadcast + drill. The herbicides were applied with a knapsack sprayer during mid-January 2006, after first irrigation, when the soil was in an adequate moisture status. To spray the herbicides successfully all the precautionary measures were adopted so as to avoid any danger of physical exposure to the herbicides. During the course of studies the data were recorded on Weed Density (m^{-2}), Plant height (cm), Spike Length (cm), No. of Spikes m^{-2} , No. of Grains spike⁻¹, 1000 grain weight (g), Biological yield (kg ha^{-1}) and Grain yield (kg ha^{-1}). Standard procedures were adopted for recording the data on all above traits.

The data recorded for each trait were individually subjected to the ANOVA technique by using SAS Computer software and means were separated by using Fisher's protected LSD test. (1997).

3. Results and discussion

An experiment comprising method of sowing and herbicides on wheat was carried out at Agricultural Research, Tehran of University, Karaj city. Data were recorded on weed dynamics and some morphological and agronomic traits of wheat. The data are presented as under:

3.1 Weed density m^{-2}

The analysis of variance showed that method of planting and interactions of method of planting with herbicides were non-significant statistically while, the herbicidal applications were evaluated as significant statistically. It is evident from the data in Table-2 that almost similar weed density m^{-2} was recorded in all methods of planting. However, the highest weed density was recorded in the broadcast sowing (37.2) as compared to line (31.5) or line + broadcast sowing (31.7). All herbicidal combinations although were non-significant among themselves, had a lower density of weeds m^{-2} as compared to the weedy check (88.889). Among the herbicides numerically lowest weeds (23.4) were recorded in Buctri-M + Isoproturon. The interaction of the method of planting with herbicides was non-significant statistically. However, the lowest weed density (20.1) was observed in line sowing treated with Buctri-M + Topik. The treatments involving line sowing in general, had the lowest infestation as compared to the interactions in line + broadcast or broadcast sowing. The highest weed density (102.69) was recorded in the weedy check under broadcast sowing (Table-2). These results are in conformity with Panwar et al, 1995, Pandey and Singh, 1994, Kha et al. (1999), Khan et al. (2002) and Khan et al. (2003) who reported that application of the tank mixed herbicides reduced broad and narrow leaf weeds to a varying degree sometimes approaching 100%. Our findings are however, contrary to the work reported by Ashrafi, 2006, Williams, 1984, Deschamps, et al., 1990, Augero-Alverdo and Appleby, 1991, and Augero-Alverdo, et al., 1991. The variability in findings could be attributed to the different herbicidal combinations tested by those researchers.

3.2 Plant height (cm)

The analysis of variance showed that method of planting and herbicidal applications were statistically significant, while the interaction between method of planting and herbicides was evaluated as non significant statistically. The perusal of data in Table-3 exhibits that Line + Broadcast and Broadcast Sowing were statistically at par with each other, but surpassed the Line Sowing. More plant height (102 cm each) was recorded in Line + Broadcast and Broadcast Sowings. All herbicides although non-significant among themselves had a more plant height as compared to the weedy check (78 cm). Almost all the herbicides gave the equal plant height (103 cm) numerically except Logran Extra + Topik and Logran Extra + Isoprturon (102 cm), which had slightly lower plant height. Earlier workers like Ahmed et al, 1999 have also found that herbicides do not effect plant height and concluded that trait under reference is strictly under genetic control. The difference in findings can be attributed to the different genetic material used and a variance in environmental conditions. The interaction of method of planting with herbicides although non-significant statistically exhibits that the plant height of the treatments involving Broadcast Sowing was generally taller (105 cm) than the other planting methods. The minimum plant height (75 cm) was observed in the weedy check under the Line sowing (Table-3). These results are corroborated with the conclusions of Gogoi and Kalita, 1995.

3.3 Spike length (cm)

The analysis of variance showed that methods of sowing and interaction of method of planting with herbicides were statistically non significant, while the herbicidal applications were detected as significant. It is evident from the data in Table-4 that almost similar spike length was recorded in all methods of planting. However, the longest spikes (10 cm) were recorded in Line and Line + Broadcast sowing. All tank mixed herbicides although non significant among themselves had a more spike length (10 cm) as compared to the weedy check (9 cm). The interaction of method of planting with herbicides was non-significant statistically, yet the highest spike length (11 cm) was observed in Line +

Broadcast Sowing treated with 2, 4-D + Puma Super, 2, 4-D + Topik, Logran Extra + Puma Super, Logran Extra + Topik and in Broadcast 2, 4-D + Puma Super (Table-4). These results are in agreement with the work of Ahmed et al, 1999, who reported that spike length is significantly affected by herbicidal applications.

3.4 No. of spikes m^{-2}

The No. of spikes per unit area is the most important trait contributing to the grain yield in wheat. The data on No. of spikes per m^2 are presented in (Table-5). The statistical analysis of the data indicated that methods of sowing and interaction of method of sowing with herbicides was statistically non significant while, the differences among the herbicides were detected as significant. The data in Table-5 show that almost similar spikes m^{-2} were recorded in all methods of sowing. However, the highest spikes m^{-2} (276.47) were observed in line sowing as compared to Line + Broadcast (269.53) or Broadcast (246.73) sowing. Among the herbicides the highest No. of spikes m^{-2} were recorded in 2, 4-D + Topik (276.67) which however were statistically at par with all other herbicidal applications but statistically higher than the weedy check (199.78) [Table -5]. The interaction of the method of planting with the herbicides although non-significant statistically exhibited that the spikes m^{-2} of the treatments involving Line sowing were generally higher than the other planting methods. Line sowing treated with Buctril-M+Topik (293.00) gave the maximum No. of spikes m^{-2} (Table-5).

3.5 Number of grains spike $^{-1}$

Number of grains spike $^{-1}$ is another important component of yield. Change in number of grains spike $^{-1}$ drastically influences the final yield. The analysis of data showed that the variable method of sowing was evaluated as significant, while the herbicides and interaction of methods of planting with herbicides were statistically non significant. The data in Table-6 exhibits those higher grains spike $^{-1}$ (53.213) were recorded in line sowing. However, it was statistically at par with line + broadcast sowing (50.197) but higher than broadcast sowing (44.2). The herbicidal treatments were statistically non significant, but numerically the highest No. of grains (52.41) were observed in 2,4-D + Topik. Minimum grains (45.00) were recorded in weedy check (Table 6). The interaction of method of planting with herbicides although non significant statistically, exhibited higher grains spike $^{-1}$ (59.569) in the line sowing treated with 2, 4-D+Topik. The lowest grains spike $^{-1}$ (42.333) were recorded in the 2,4-D+Isoproturon under the broadcast planted treatment. These results are corroborated with the results of Balyan et al., 1983, Khan et al, 2001, Khan et al., 2002 and Khan et al, 2003, who concluded that herbicidal applications produce more grains spike $^{-1}$ than the untreated control.

3.6 1000-grain weight (g)

The analysis of data indicated that the methods of planting and herbicides were significant statistically, while the interaction of method of planting with herbicides was statistically non significant. The maximum 1000 grain weight (39 g) was recorded in line + broadcast sowing, but it was statistically higher than the line and broadcast sowing (37 g) [Table 7]. Among the herbicides, the maximum 1000 grain weight was recorded in Buctril-M+Topik and Logran Extra+ Puma Super (39 g) each which however were statistically at par with all other herbicidal applications, but statistically higher than the weedy check (30 g). The interaction of method of planting with herbicides although non significant statistically, showed that the treatments involving line and line + broadcast sowing generally had bolder grain than broadcast sowing. Line sowing treated with Buctril-M+Puma Super (40 g each) had the highest 1000 grains weight (Table -7), while broadcast and line sowings under the weedy check (28 g each) possessed the smallest kernel size. Similar results were also reported by Tanveer et al., 1999, Balyan et al., 1996 and Samunder et al., 1994, who concluded that the herbicides were very effective for weed control and also gave best crop yield.

3.7 Biological yield (kg ha $^{-1}$)

The analysis of variance showed that method of planting and herbicidal applications were statistically significant, while the interaction of method of planting with herbicides was recorded as statistically non significant. The highest biological yield (17763 kg ha $^{-1}$) was recorded in Line sowing, while Line +Broadcast (16739) and Broadcast sowing (15970) were statistically at par with each other (Table -8). Among the herbicides, the highest biological yield (18793 kg ha $^{-1}$) was recorded in 2, 4-D + Puma Super. However, it was statistically at par with all other herbicidal applications except Logran Extra + Topik (15638 kg ha $^{-1}$). The lowest biological yield (10908 kg ha $^{-1}$) was observed in the weedy check. The interaction of method of planting with herbicides although non-significant statistically exhibits that the biological yield of the treatments involving Line sowing was generally higher than the other planting methods. Line sowing treated with 2,4-D + Puma super gave maximum biological yield (23241 kg ha $^{-1}$) than rest of the interactions. The minimum biological yield (10601 kg ha $^{-1}$) was recorded in weedy check under the Line + broadcast sowing. The herbicide 2,4-D + Puma super gave the excellent control of weeds hence consequently it increased the biological yield (Table-8). These results are in a greater agreements with the work of Porwal and Gupta, 1987 and Brar et al., 1999b. They also reported that different herbicides reduced weed and increased grain and straw yield of wheat over the control plots.

3.7 Grain yield (kg ha^{-1})

The data showed that method of sowing and the interaction of method of sowing with herbicides were statistically non-significant while, the differences among the herbicides were detected as significant. The perusal of data (Table 9) exhibited that almost similar grain yield was recorded in all methods of planting. However, the line + broadcast had a slightly higher yield ($4142.78 \text{ kg ha}^{-1}$) as compared to line (4078.39) or broadcast sowing (4088.53). All the herbicides out yielded the weedy check. Among the herbicides the highest yield was recorded in Buctril-M + Topik ($4771.06 \text{ kg ha}^{-1}$). However, it was statistically at par with all other herbicidal applications except Buctril-M + Isoproturon ($3631.82 \text{ kg ha}^{-1}$), 2,4-D + Isoproturon ($3775.64 \text{ kg ha}^{-1}$), and Buctril-M + Puma Super ($4138.53 \text{ kg ha}^{-1}$). The herbicide Buctril-M + Isoproturon was in turn statistically comparable with Buctril-M + Puma Super ($4138.53 \text{ kg ha}^{-1}$), Logran Extra + Topik ($4378.86 \text{ kg ha}^{-1}$), Logran Extra + Isoproturon ($4389.79 \text{ kg ha}^{-1}$) and Logran+Puma Super ($4314.13 \text{ kg ha}^{-1}$) (Table 9). The herbicides Buctril-M+Topik gave an excellent control of weeds hence consequently increased the grain yield. The interaction of the method of planting with the herbicides although non-significant statistically exhibited that the yield of treatments involving line+broadcast sowing was generally higher than the planting with the other methods. Line+Broadcast treated with 2,4-D+Puma Super out yielded ($5115.23 \text{ kg ha}^{-1}$) the rest of the interactions. The lowest grain yield ($2239.68 \text{ kg ha}^{-1}$) was recorded in the weedy check, under line sowing. These findings are in a close conformity with Pandey and Singh 1994, Azad et al, 1997, and Samunder et al, 1994 who reported a differential response of various herbicides on the grain yield of wheat.

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Table 1. Detail of the tank mixed herbicides assigned to the sub-plots

S. No.	Herbicides Mixtures (Trade name)	Common Name	Dose (kg a.i.ha ⁻¹)
.1	2,4-D+ Puma Super 75 EW	2,4-D fecnoxaprop-p-ethyl	0.90 + 0.84
.2	2,4-D + Topik 15 WP	2,4 -D+ clodinafop	0.90 + 0.04
.3	2,4-D + Isoproturan 50 WP	2,4-D + isoproturon	0.90 + 0.63
.4	Buctril-M40 EC+Puma Super 75 EW	(bromoxynil + MCPA) + fenoxaprop-p-ethyl	0.70 + 0.94
.5	Buctril-M 40 EC + Topik 15 WP	(bromoxynil + MCPA) + isoproturon	0.70 + 0.14
.6	Buctril-M 40 EC +Isoproturon 50 WP	(bromoxynil + MCPA) + isoproturon	0.71+ 0.63
.7	Logran Extra 64 WG + Puma Super 75 EW	(triasulfuron+ terbutryn)+ fenoxaprop-p-ethyl	0.16 + 0.94
.8	Logran Extra 64 WG + Topik 15 WP	(triasulfuron+ terbutryn)+ clodinafop	0.16 + 0.04
.9	Logran Extra 64 WG + Isoproturon 50 WP	(triasulfuron + terbutryn)+ isoproturon	0.16 + 0.63
.10	Weedy check (no weeding)	-	-

Table 2. Effect of method of planting and herbicides on weed density (m⁻²) in Wheat

Herbicidal combination	Line sowing	Line Broadcast +	Broadcast	Herbicide Means
2,4-D + Puma Super	22.333	25.667	31.667	26.556 A*
2,4-D + Topic	26.000	24.667	31.000	27.224 A
2,4-D + Isoproturon	27.000	28.333	34.667	30.000 A
Buctril-M + Puma Super	29.000	30.000	34.333	31.111 A
Buctril-M + Topik	20.000	24.667	29.000	24.556 A
Buctril-M+ Isoproturon	25.667	22.333	22.000	23.333 A
Logran Extra+ Puma Super	25.000	28.667	33.667	29.211 A
Logran Extra + Topik	24.667	25.667	24.000	24.778 A
Logran Extra + Isoproturon	23.000	27.000	26.000	25.333 A
Weedy check	82.000	80.000	104.667	88.889 B
Mean	30.467	31.700	37.100	

LSD_{0.05} for herbicides = 11.26

* The means sharing a letter in common do not differ significantly by LSD test at 5% probability level.

Table 3. Effect of method of planting and herbicides on plant height (cm) in wheat

Herbicidal Combination	Line sowing	Line Broadcast +	Broadcast	Mean
2,4-D + Puma Super	100	103	105	103 A *
2,4-D + Topic	101	103	105	103 A
2,4-D + Isoproturon	100	104	104	103 A
Buctril-M + Puma Super	100	104	105	103 A
Buctril-M + Topic	100	104	104	103 A
Buctril-M+ Isoproturon	101	104	104	103 A
Logran Extra + Puma Super	100	104	104	103 A
Logran Extra + Topic	99	103	103	102 A
Logran Extra + Isoproturon	98	104	104	102 A
Weedy check	75	81	79	78 B
Mean	98 B	102 A	102 A	

LSD_{0.05} for sowing methods = 3.348, LSD_{0.05} for herbicides = 2.671, * The means sharing a letter in common in their respective category do not differ significantly by LSD test at 5% probability level.

Table 4. Effect of method of planting and herbicides on spike length (cm) in wheat

Herbicidal Combination	Line sowing	Line Broadcast +	Broadcast	Mean
2,4-D + Puma Super	10	11	11	10 A*
2,4-D + Topic	10	11	9	10 A
2,4-D + Isoproturon	10	10	10	10 A
Buctril-M + Puma Super	10	10	9	10 A
Buctril-M + Topic	10	10	10	10 A
Buctril-M+ Isoproturon	10	10	10	10 A
Logran Extra + Puma Super	10	11	9	10 A
Logran Extra + Topic	9	11	9	10 A
Logran Extra + Isoproturon	9	10	9	10 A
Weedy Check	9	9	9	9 B
Mean	10	10	9	

LSD_{0.05} for herbicides = 0.9822 , *The means sharing a letter in common do not differ significantly by LSD test at 5% probability level.

Table 5. Effect of methods of planting and herbicides on No. of Spikes m^{-2} in wheat

Herbicidal Combination	Line sowing	Line Broadcast ⁺	Broadcast	Mean
2,4-D + Puma Super	275.00	277.00	233.33	261.78 A*
2,4-D + Topic	288.67	281.67	259.67	276.67 A
2,4-D + Isoproturon	278.33	279.33	266.67	274.78 A
Buctril-M+ Puma Super	286.00	263.00	263.00	270.67 A
Buctril-M + Topic	293.00	270.67	262.67	275.44 A
Buctril-M+ Isoproturon	280.00	276.33	272.67	276.33 A
Logran Extra + Puma Super	289.67	283.33	237.33	270.11 A
Logran Extra + Topic	288.00	280.67	230.33	266.33 A
Logran Extra + Isoproturon	280.33	275.67	255.67	270.56 A
Weedy Check	205.67	207.67	186.00	199.78 B
Mean	276.47	269.53	246.73	

LSD_{0.05} for herbicides = 39.46, *The means sharing a letter in common do not differ significantly by LSD test at 5% probability level.

Table 6. Effect of method of planting and herbicides on No. of grains Spike⁻¹ in wheat

Herbicidal Combinations	Line sowing	Line Broadcast ⁺	Broadcast	Mean
2,4-D + Puma Super	52.100	52.00	42.667	48.92
2,4-D + Topic	59.569	50.667	47.000	52.41
2,4-D + Isoproturon	50.767	50.633	42.333	47.81
Buctril-M + Puma Super	54.800	49.667	48.333	50.93
Buctril-M + Topic	51.433	54.133	44.333	49.97
Buctril-M+ Isoproturon	54.867	50.000	43.333	49.29
Logran Extra + Puma super	56.467	48.033	45.333	49.94
Logran Extra + Topic	57.133	51.800	43.000	50.64
Logran Extra+ Isoproturon	48.667	49.333	43.333	47.11
Weedy Check	46.333	46.000	42.667	45.00
Mean	53.213 A*	50.197 A	44.200 B	

LSD_{0.05} for sowing methods = 4.618 , *The means sharing a letter in common do not differ significantly by LSD test at 5% probability level.

Table 7. Effect of method of planting and herbicides on 1000 grain weight (g) in wheat

Herbicidal Combinations	Line sowing	Line Broadcast ⁺	Broadcast	Mean
2,4-D + Puma Super	38	39	38	38 A*
2,4-D + Topic 15 WP	39	39	37	38 A
2,4-D + Isoproturon	38	39	36	38 A
Buctril-M + Puma Super	40	39	37	38 A
Buctril-M + Topic	38	40	38	39 A
Buctril-M+ Isoproturon	39	39	37	38 A
Logran Extra+ Puma Super	38	39	39	39 A
Logran Extra+Topik 15 WP	38	40	37	38 A
Logran Extra+ Isoproturon	37	40	37	38 A
Weedy Check	28	33	28	30 B
Mean	37 B	39 A	37 B	

LSD_{0.05} for sowing method = 1.62LSD_{0.05} for herbicides = 2.628

*The means sharing a letter in common in their respective category do not differ significantly by LSD test at 5% probability level.

Table 8. Effect of method of planting and herbicides on biological yield kg ha⁻¹ in wheat

Herbicidal Combinations	Line sowing	Line Broadcast ⁺	Broadcast	Mean
2,4-D + Puma Super	22222	16872	17284	18793 A*
2,4-D + Topic 15 WP	18922	18930	16173	18008 AB
2,4-D + Isoproturon	19341	17284	18930	18518 AB
Buctril-M + Puma Super	18383	17697	15638	17239 AB
Buctril-M + Topic 15 WP	18107	16872	15226	16735 AB
Buctril-M+ Isoproturon	18930	15638	18107	17558 AB
Logran Extra+ Puma Super	16461	16872	17284	16872 AB
Logran Extra + Topic 15 WP	15638	17284	13992	15638 B
Logran Extra + Isoproturon	18107	19342	16461	17970 AB
Weedy Check	11523	10601	10601	10908 C
Mean	17763 A	16739 B	15970 B	

LSD_{0.05} for sowing methods = 851, LSD_{0.05} for herbicides = 2893, * The means sharing a letter in common in their respective category do not differ significantly by LSD test at 5% probability level.

Table 9. Effect of method of planting and herbicides on grain yield (kg ha^{-1}) in wheat

Herbicide Combination	Method of Sowing			
	Line sowing	Line Broadcast ⁺	Broadcast	Mean
2,4-D + Puma Super	4395.06	5115.23	4568.07	4692.78A*
2,4-D + Topic 15 WP	4802.47	4691.36	4407.59	4633.81A
2,4-D + Isoproturon	3744.86	4084.13	3497.94	3775.64CD
Buctril-M + Puma Super	3781.89	3884.74	4748.97	4138.53BC
Buctril-M + Topic	4494.24	4979.42	4839.51	4771.06A
Buctril-M+ Isoproturon	3827.16	3223.46	3844.86	3631.82D
Logran Extra + Puma Super	4979.42	3859.67	4103.29	4314.13AB
Logran Extra + Topic 15 WP	4320.98	4456.57	4359.02	4378.86AB
Logran Extra + Isoproturon	4198.19	4855.97	4115.22	4389.79AB
Weedy Check	2239.68	2277.36	2400.82	2305.76 E
Mean	4078.39	4142.78	4088.53	

LSD_{0.05} for treatment = 494.7 * The means sharing a letter in common do not differ significantly by LSD test at 5% probability level.



Determination of Soya Plant Population Using NDVI in the Dasht-e-Naz Agri-Industry

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Abstract

Numerous efforts have been made to develop various indices using remote sensing data such as normalized difference vegetation index (NDVI), and vegetation condition index (VCI) for mapping and monitoring of yield estimating and assessment of vegetation health and productivity. NDVI and other indices that derive from satellite images are valuable sources of information for the estimation and prediction of crop conditions. In the present paper, NDVI data of Dasht-e-Naz in Iran in 2006 have been considered for crop yield assessment and estimating. The results showed that there is acceptable relationship between NDVI and Soya plant population. The correlation between NDVI and plant population in high plant population area of field was ($R^2=0.923$) and for low plant population area was ($R^2=0.249$). The crop population models were discussed about high and low plant population in the present paper and could improve in future with the use of long period dataset. Similar model can be developed for different crops of other locations.

Keywords: Soybean, Plant population, Precision farming, Satellite images, Normalized Difference Vegetation Index, NDVI

1. Introduction

Precision farming is a new agricultural system concept with the goals of optimizing returns in agricultural production and environment. This concept involves the development and adoption of remote sensing (Barnes et al., 1996), Geographic Information System (GIS) technology applications, and knowledge-based technical management systems (NRC, 1997). With a refined GIS and spatial knowledge-based management system, farmers should have the ability to appropriately manage field operations at each location in the field, as well as the ability to predict likely yield from early season indicators and distinguish crop type from far. Many studies have focused on variable rate applications (Gotway et al., 1996; Stafford and Miller, 1996) while some are focused on yield mapping (Sudduth et al., 1996). Yield mapping provides not only information about the yield itself, but it allows comparison to field conditions that may explain spatial yield variation.

Remote sensing techniques have a number of advantages over the conventional techniques such as field surveying in vegetation-dynamics studies. Satellite remote sensing has special advantages since it can produce multi temporal images at frequent intervals which facilitate temporal monitoring of vegetation over an area. During the last decade, coarse spatial resolution, high temporal frequency satellite data such as NOAA AVHRR were used extensively to monitor vegetation cover and climatic variability throughout the world (Kulawardhana et al., 2004). The normalized ratio of near-infrared reflectance to red reflectance, called the normalized difference vegetation index (NDVI) has been shown to be a sensitive indicator of biomass and leaf area in several crops, which can be used to distinguish crop. Once a relationship between yield and NDVI is developed, then farmers can predict their yield earlier in the growing season

and therefore, better harvest management, planning the following season's inputs, and other more effective management can be achieved (Chewab and Murdock, 2002).

In the remote sensing, electromagnetic waves are recorded on different platforms. Reflective energy is the great importance and most of system. The rate of electromagnetic reflection in visible spectrum (green, red, and blue frequency) and close infrared depend on the surface of the subject (Jensen, 1996). The manner of reflection of visible spectrums and infrared is shown in figure 1.

Remote sensing has been used to evaluate nutrient deficiencies such as P concentrations of soybean (Milton et al. 1991), S, Mg, K, P and Ca of corn leaves (Al-Abbas et al. 1974), Fe, S, Mg, and Mn in corn, wheat, barley, and sunflower (Masoni et al. 1996) and P uptake of bermudagrass (Sembiring et al. 1998). Reflectance, the ratio of incoming to reflected radiance, can be used to estimate total N and chlorophyll content of fresh plant samples (Yoder and Pettigrew-Crosby 1995). Raun et al. (2001) showed that mid-season sensor reflectance measurements could be used to predict yield potential of winter wheat.

NDVI index is one of the best-known and also best-working indices ever which were first performed in 1974 by Rouse et al. One of the functions of this method is vegetation and plant refreshment and is capable to monitor those levels in different ages. In order to exploitation of vegetation from satellite images, existence or non - existence of satellite band are important. In other words, more correlation between satellite image bands can show more information about satellite image (Drost et al., 1997).

Several works have shown that NDVI is correlated with aboveground net primary production (ANPP) (Paruelo et al., 2000, 2004; Pineiro et al., 2006). Therefore, exploring NDVI interannual variation and its relationship with precipitation contributes to understanding ANPP variation. NDVI is more closely associated with ANPP when the situations being compared largely differ in leaf area and are similar in incident radiation and in radiation use efficiency (Pineiro et al., 2006).

Chewab and Murdock (2002) had applied the NDVI method for gathering amount of nitrogen fertilizer. In another research, using vegetation index from satellite images, cultivated area of soybean and corn were calculated (Drost et al., 1997).

The goal of this research was to predict and evaluate some characteristics of vegetation such as accumulation and plant population of soybean in the Dasht-e-Naz Agri-Industry using vegetation index.

2. Materials and methods

This study employed NDVI to examine the relationship between Soya plant population and the Normalized Difference Vegetation Index (NDVI) in the Dasht-e-Naz agri-industry. The relationship between Soya plant population and NDVI in the Dasht-e-Naz agri-industry was examined using spatial analysis methods. For NDVI interpretation and understanding the accumulation and vegetation populace, first the geographical information of subjected area was studied. Satellite images of Dasht-e-Naz Agri-industry were captured. Some samples were taken from that area and geographical region was determined. Dasht-e-Naze is about 3200 hectares and this area is located between 36°37" latitude and 53°07" longitude. This research was done between years 2005 and 2006. The time of information gathering from the field was obtained by using Orbitron software. Software showed that passing time of IRS satellite was in 2005/09/05 and data were gathered at this time.

Geographical positions of field were determined by using GPS and then population of Soya was investigated by the satellite images. The satellite images of Dasht-e- Naz area were captured by IRS-1C, 1D satellite. The LISS-III and PAN Images of subjected are shown in Figures 2 and 3, respectively.

The raw images were captured from satellite then were processed by PCI-GEOMATICA software. LISS-III images were containing visible bands and their resolutions were about 23.5 meter. For increasing the resolution of images, the PAN and LISS-III images were processed and merged by PCI-GEOMATICA software. Final processed image is shown in the Figure 4. Then the pixel values were exploited from the image in the final process by PCI-GEOMATICA software.

NDVI index was exploited through the below formula (Lillesand anf Kiefer, 1994):

$$NDVI = \frac{B3 - B2}{B3 + B2}$$

where B3 and B2 are spectral reflectance in the near-infrared band and reflectance in the red band, respectively. This is a less measure unit that their range usually is between -1 to 1. Water, snow, clouds or any other non-vegetated scene is represented by a negative number. Low positive numbers near zero indicate rock and bare soil, which reflect near infrared and red at the same level. Increasingly positive numbers indicate greener vegetation (Lillesand and Kiefer, 1994).

In order to determine the ground information, latitude and longitude of plots and measuring points were determined. Then by using a square frame, number of plants per frame was determined. Value of satellite image pixels was extracted

proportionate with measuring points. Table 1 shows the plot number, latitude and longitude of plots, values of bands, NDVI, and plant population.

In order to determine the value of plant population, two regions of plot on the image were selected and pixel value was determined in an 8×9 matrix with 5.8 m² for area of each pixel. The stated pixel in the tables is the center of each matrix. The band values spectrum of two plots are shown in tables 2, and 3. Table 2 shows the values of two bands of high Soya population and another one showed these values about low Soya population. In order to show difference between populations and reduce the sample errors, two samples were selected for each plot.

3. Results and discussion

After gathering information from satellite images and field, the relationship between RGB bands were investigated. Figures 5 and 6 show the correlation between B2 and B3 in high and low plots. Strong relationship between two bands could give us more accurate information. The results showed that there was weak correlation between bands 2 and 3 for low population plots and against that there was strong correlation between those in accumulated fields. The results showed that estimation of plant population in accumulated fields were more accurate than fields with low plant population because in high plant population fields there was good relationship between the bands of satellite images.

The relationship between NDVI indices and Soya population were estimated and are shown in figure 7. A strong positive correlation between NDVI and Soya was found in high population fields. The results showed that the relationship between NDVI and plant population was a liner relationship. The results showed that with increasing the NDVI, plant population is increased. Comparison between the field's data and the results from satellite images showed that there was straight relationship between plant population and NDVI in different field with different plant population but in the field with high plant population, the correlation ratio was very better than that of low plant population.

In order to increase the accuracy of correlation between bands, number of samples from each geographical region was calculated. This leads to the proper show of the information between bands. Figure 8 shows the NDVI of high and low plant population via the number of samples. According to this figure, the NDVI of high plant population was more than low plant population.

The average of NDVI for high plant population and low plant population plots are shown in figure 9. The results showed that there was homogeneous plant population in both samples of high plant population and low plant population and only plant per square meter of plots was different. Based on the results, NDVI index less than 0.2 showed low accumulation and NDVI index with about 0.5 showed high accumulation. It was found that the reason of low plant population of some part of field was because no seed was planted.

4. Conclusions

It can be concluded that using the vegetation index could suitable to estimate the soybean accumulation. And with using this index we could distinguish the region with low and high plant accumulation. Based on the results, NDVI index less than 0.2 showed low accumulation and NDVI index with about 0.5 showed high accumulation. It was found that the reason of low plant population of some part of field was because of not planted seed by planter at planting time.

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Table 1. Observed samples in the Dasht-e-Naz Agri-industrial Company and in the satellite image

Plot number	latitude	longitude	Band (B1,B2,B3)	NDVI	Plant Population (Plant/m ²)
40 (Middle)	53°07'18.7"	36°37'10.5"	37-96-67	0.44	17
40 (Middle)	53°07'18.7"	36°37'10.5"	43-101-72	0.4	17
40 (Middle)	53°07'18.9"	36°37'10.5"	41-98-70	0.41	17
40 (Middle)	53°07'19.1"	36°37'10.5"	45-99-102	0.37	16
40 (Middle)	53°07'19.3"	36°37'10.5"	43-96-71	0.38	16
40 west	53°07'1.4"	36°37'3.3"	38-89-65	0.4	17
40 west	53°07'1.4"	36°37'3.5"	39-90-66	0.39	16
40 west	53°07'1.4"	36°37'3.7"	38-89-65	0.4	17
40 west	53°07'1.4"	36°37'3.8"	39-90-66	0.39	16
40 west	53°07'1.4"	36°37'4"	42-93-69	0.37	16
40 East	53°07'36.1"	36°37'19"	41-95-69	0.39	16
40 East	53°07'36.3"	36°37'19"	43-87-62	0.34	16
40 East	53°07'36.4"	36°37'19"	37-90-65	0.41	17
40 East	53°07'36.7"	36°37'19"	34-88-61	0.44	17
40 East	53°07'36.8"	36°37'19"	34-91-63	0.45	18
39 west	53°07'53.4"	36°37'26.9"	25-92-57	0.57	19
39 west	53°07'53.6"	36°37'26.9"	39-95-60	0.53	19
39 west	53°07'53.8"	36°37'26.9"	31-98-63	0.51	19
39 west	53°07'54"	36°37'26.9"	33-98-64	0.49	18
39 west	53°07'54.2"	36°37'26.9"	32-97-63	0.5	19
39 East	53°08'11.7"	36°37'35"	98-99-66	0.44	17
39 East	53°08'11.9"	36°37'35"	35-94-64	0.45	18
39 East	53°08'12.1"	36°37'35"	38-93-66	0.41	17
39 East	53°08'12.3"	36°37'35"	40-94-68	0.4	17
39 East	53°08'12.5"	36°37'35"	45-97-72	0.36	16

Table 2. Band values spectrums of a high Soya population plot.

B2								
31	34	32	32	31	29	33	32	33
32	34	32	31	33	30	29	32	33
33	34	32	30	32	35	32	32	34
33	33	31	33	35	35	34	33	33
33	33	31	34	×35	35	30	33	32
34	32	32	35	35	35	29	32	32
34	31	32	36	34	32	32	33	31
33	33	33	36	33	27	30	33	30
B3								
92	95	93	93	92	90	94	93	92
93	95	93	92	94	91	91	94	94
94	95	94	92	94	97	94	94	96
94	94	92	95	97	97	96	95	95
94	94	92	95	×96	97	92	95	93
95	93	93	96	96	96	90	93	93
95	92	93	97	95	93	93	94	92
94	94	94	97	94	88	91	94	91

Table 3. Band values spectrums of a low Soya population plot.

B2								
68	69	70	69	70	69	68	69	71
71	69	70	69	70	68	68	69	72
70	69	70	68	69	68	68	69	72
69	68	70	69	69	68	69	70	73
68	68	67	66	×69	68	69	70	72
66	67	63	62	67	69	69	71	72
65	64	61	62	66	68	70	70	71
61	63	61	63	66	68	69	68	67
B3								
86	87	89	88	89	90	89	90	92
89	87	89	88	89	89	89	90	91
89	88	89	89	90	89	90	90	91
90	89	91	91	91	90	91	92	93
90	90	89	88	×69	90	91	92	94
91	91	87	86	90	92	92	94	95
91	88	87	88	91	93	93	93	94
88	89	87	91	92	93	94	93	93

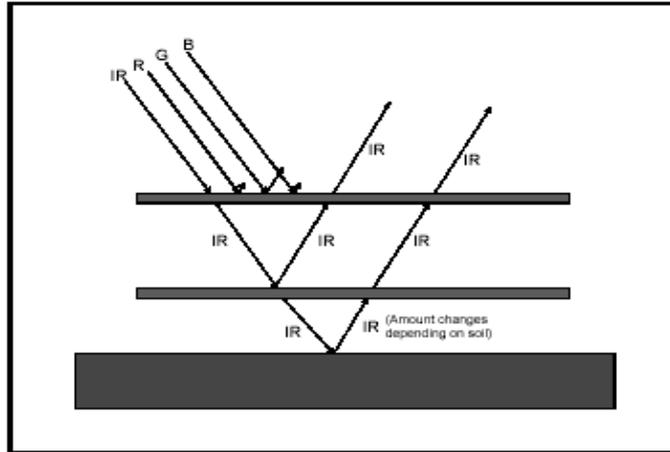


Figure 1. The manner of reflection of visible spectrums and infrared spectrum

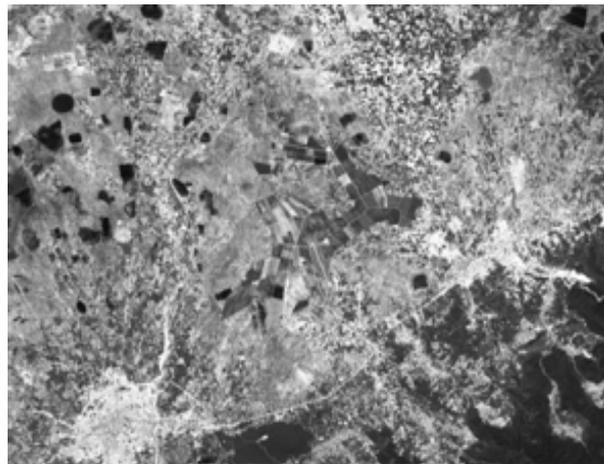


Figure 2. LISS-III image of Dasht-e-Naz

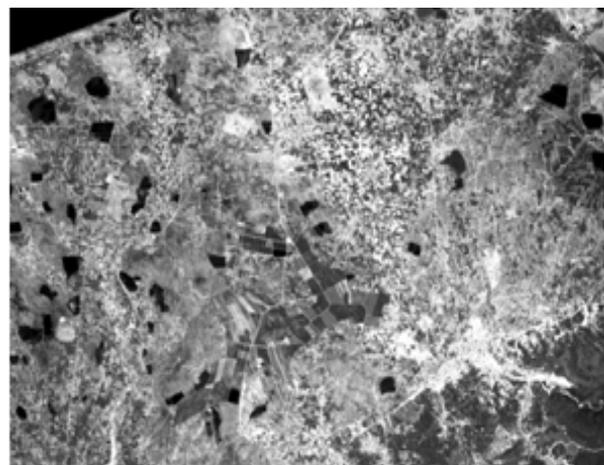


Figure 3. PAN image of Dasht-e-naz

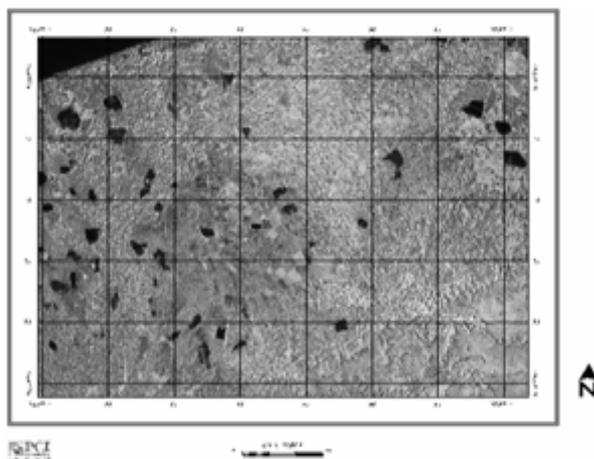


Figure 4. Merged image of Dasht-e-Naz

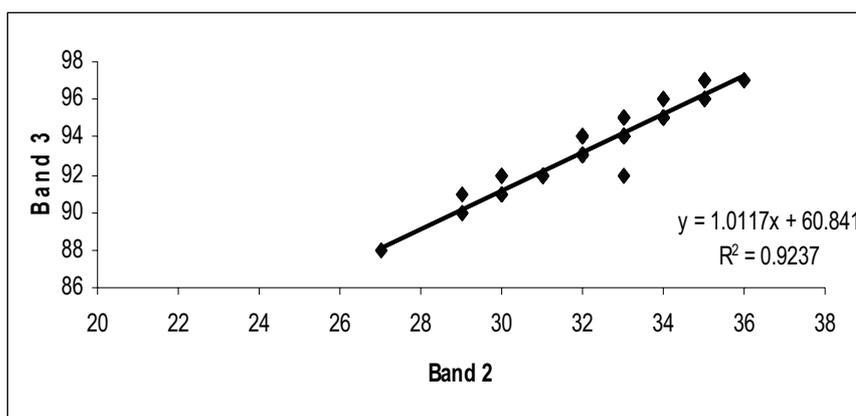


Figure 5. Relationship between bands 2 and 3 for high Soya population

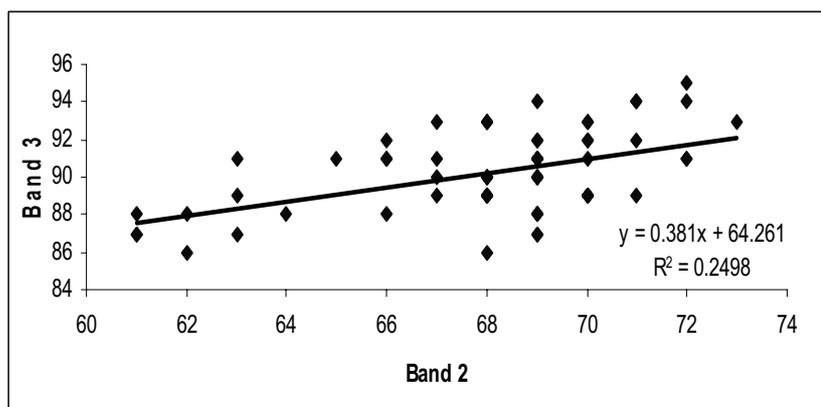


Figure 6. Relationship between bands 2 and 3 for low Soya population

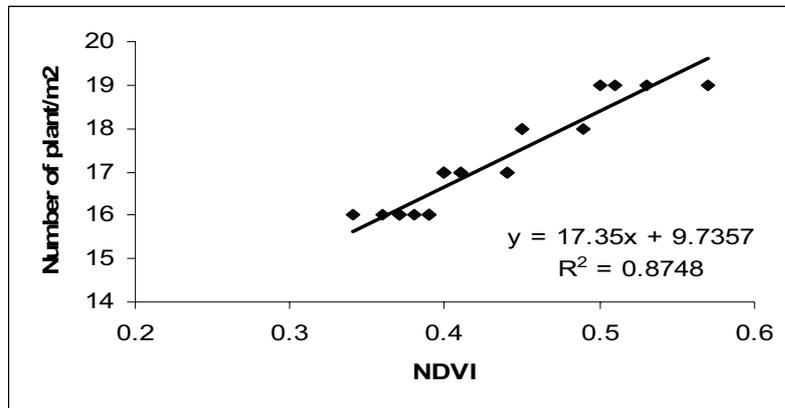


Figure 7. Relationship between NDVI and Soya population for high Soya population

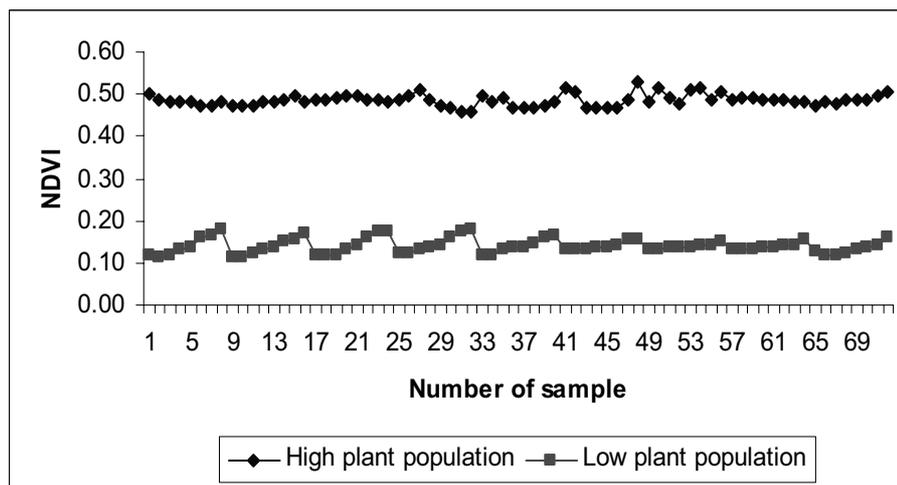


Figure 8. NDVI index for the samples with low and high plant population

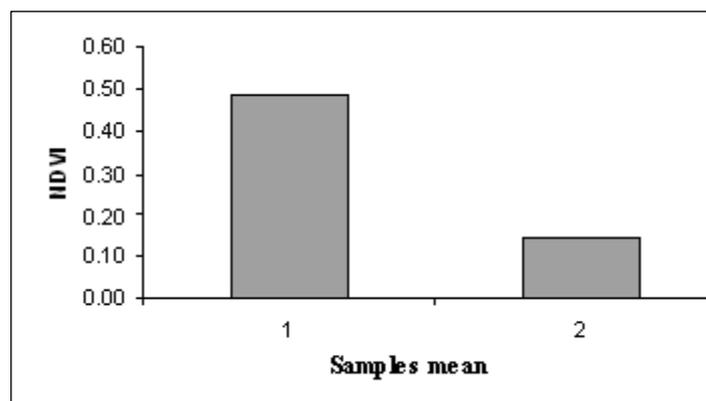


Figure 9. Average of NDVI index for the samples with (1) high and (2) low plant population



Effects of Different Fertilizer Application Level on Growth and Physiology of *Hibiscus cannabinus* L. (Kenaf) Planted on BRIS Soil

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The research is financed by Research University Grant Scheme No. 91049

Abstract

Hibiscus cannabinus L. or Kenaf is one of the most potential annual crop planted throughout the world. Being fast growing and multipurpose, it has been utilized as a substitute of jute and, more recently, as raw product for the production of pulp and paper. With strong and long fiber yield, mass production of Kenaf throughout Malaysia is critical. The utilization of less fertile soils such as BRIS soils is important to increase the Kenaf production throughout Malaysia. Thus, the objective of this study was to determine the effects of different level fertilizer application on growth and physiology of Kenaf planted on BRIS soils. V36 variety was used and planted in three different plots by treatments with fertilizers namely high (1960 kg/ plot), medium (1260 kg/ plot) and low (700 kg/ plot) respectively. Each plot comprises 106,000 trees where trees were planted on 20 lines. There were contrasting results on the effects of fertilizer on growth and physiology of Kenaf in the dry (41 days) and wet season (64 days). Significant effects were only observed for diameter, height, leaf number and area during the wet season. Similar results were also found for biomass. The increasing trends with increasing the rates of fertilizer were observed in the wet season for growth and biomass parameters. The correlation analyses between total aboveground biomass with diameter and height were more pronounced in the wet season. *AGR*,

RGR and E_G calculated from the differences between the dry and wet season readings for aboveground biomass showed that the higher rate of fertilizer recorded the higher values of AGR and RGR . However, no trend was observed for E_G .

Keywords: Kenaf, BRIS soil, Growth, Physiology, Fertilizer

1. Introduction

Hibiscus cannabinus L. or normally known as Kenaf is one of the most potential annual crop planted throughout the world. It is a member of the hibiscus family (Malvaceae) and indigenous to Africa. Kenaf is an annual fibre crop of tropical origin. The plant has been utilized in the cordage and sacking manufacture as a substitute of jute and, more recently, as raw product for the production of paper pulp. The species is grown in the Asia-Pacific region, with India, China, Bangladesh, producing the largest part of the world supplies. Stems of Kenaf plants consist of an inner thick core of short woody fibres 0.5 to 1 mm long, and of an external bark with fibres of 3 to 4 mm long. The bast fibres are of better quality than the core fibres; both, however, can be utilized in various blends for the production of pulp (Petrini *et al.*, 1994).

In Malaysia, It was first introduced in the early 70's and was highlighted in the late 90's as an alternative and cheaper source of material for producing panel products such as fibreboard and particleboard. According to Sellers *et al.* (1993), Kenaf also has a high potential as a board raw material with low density panels suitable for sound absorption and thermal resistance. Being fast growing and multipurpose, Kenaf also is a good carbon sequester and can improve soil fertility. By the potential to be a valuable agronomic crop leading to the production of a number of commodities, effects of plant population, soil fertility, location and cultivar on Kenaf yields have to be studied (Bhangoo *et al.*, 1986; Ching *et al.*, 1992; Webber, 1993a, b). Kenaf variety needed suitable environmental conditions to predict accurately what and how much of a Kenaf commodity can be produced at a given site and geographic location (Ching *et al.*, 1992; Webber, 1993b). With the highly adaptable with all ranges of soils; Kenaf can be potentially planted at Beach Ridges Interspersed with Swales (BRIS) soil which is poor in water holding capacity and nutrient availability.

The total area of BRIS soils spread along the east coast of the Peninsular and the coastal area of Sabah is about 200,000 ha in total with 155,400 ha in Peninsular Malaysia and 40,400 ha in Sabah respectively. BRIS soils contain 82-99% sand particles, mainly quartz, and have a low Cation Exchange Capacity (CEC) of 9.53 meq/100 g with pH 4.3 to 4.4 (Chen, 1985). BRIS soil structure contains mostly of sand particles with having low water-holding capacity (Jensen, 1989) and poor in nutrient. It is well known that water deficit affects every aspect of plant growth, modifying anatomy, morphology, physiology and biochemistry (Hsaio, 1973; Enu-Kwesi *et al.*, 1986).

The information on growth and physiology of Kenaf planted on BRIS soils is almost non-existence and scarce. Thus, the limitation of growth on this type of soil should be studied thoughtfully based on information that Kenaf can adapt to all types of soils. Kenaf can reach at 4 m tall after one rotation (approximately 4 months) at the normal soil which rich in nutrient-water content.

With the increase of utilization, strong and long fiber yield, the mass production of Kenaf throughout Malaysia is critical. Thus, the utilization of less fertile soils such as BRIS soils is important to increase the Kenaf production throughout Malaysia. The information of Kenaf adaptability on BRIS soil is crucial. At the same time, silvicultural practices also need to be implemented on BRIS soil in order to increase growth and productivity of Kenaf. Therefore, the study on growth and physiology are important to determine the successfulness of different level fertilizer application of planting Kenaf on BRIS soil. Hence, the objective of this study was to determine the effects of different level fertilizer application on growth and physiology of Kenaf planted on BRIS soil.

2. Materials and methods

2.1 Study Site

The study was conducted in Setiu, Terengganu at the latitude of 5°36'59.42"N and longitude of 102°44'18.04"E. The location of the study area is about 200 m above sea level. Total cover areas in Setiu are 130,436.3 ha.

The mean relative humidity of the study site is about 70% to 90% (Malaysia Meteorological Department, 2008). The ranges of mean minimum and maximum temperature are 22°C to 37°C respectively. The annual rainfalls in Setiu fluctuated from the lowest 2990 mm to the highest of 4003 mm per year.

2.2 Plant Materials

V36 was used in this experiment and planted on BRIS soils in Setiu, Terengganu. This variety was selected by Lembaga Tembakau Negara (National Tobacco Board) and was planted on 14th February 2008 in three different plots assigned by treatments (levels of fertilizer). Each plot comprises 106,000 plants and were planted in 20 lines.

2.3 Experimental design

The plant samples were randomly selected using quadrat sampling within the plots and generated using random table. In each experiment, 15 samples per treatments were used in both the dry and the wet seasons. NPK with the ratio of 12:12:36 + 2MgO + TE and the micronutrient compositions were Boron, Cuprum, Iron, Manganese, Molybdenum and Zink were

used. The treatments were divided into three different fertilizer levels namely high (1960 kg/ plot), medium (1260 kg/ plot) and low (700 kg/ plot) respectively. Fertilizer was sprayed for 16 second per liter for every plot per day.

2.4 Growth parameters

2.4.1 Sampling procedure of growth parameters

Ten samples per treatment were selected randomly for measurement. Plant height (Ht), diameter (D), number of leaves (LN), leaf area (A_L) and biomass were taken. For plant biomass, plants were divided into components namely leaf mass (M_{Leaf}), stem mass (M_{Stem}), root mass (M_{Root}) and flower mass (M_{Flower}). Total aboveground biomass (M_{AG}) was calculated by summing the values of M_{Leaf} and M_{Stem} . Root to shoot ratio ($Rt:St$) was also calculated by dividing the values of M_{Root} and M_{AG} . In addition, absolute growth rate (AGR), relative growth rate (RGR) and growth efficiency (E_G) were also measured (Muchow, 1979). Plant Ht was measured using a steel ruler and height stick while D (10 cm above the ground) was measured using digital caliper. The Ht was measured from cotyledon level up to the base at the terminal bud. Measurement of collar D was made using a digital caliper and starting point of the measurement was marked using permanent marker pen. Two readings were made at the right angles to each other. The A_L was measured using Model Li-3100 Area Meter, LICOR, inc. Lincoln, Nebraska USA.

For biomass experiment, leaves, stems and roots were dried in oven at 60-70°C for 48 hour until the constant weight was obtained. Total aboveground biomass was calculated by summing the values of stems and leaves dry weight.

The absolute growth rate (AGR), relative growth rate (RGR) and growth efficiency (E_G) were determined after aboveground biomass at first reading (dry season) and second reading (wet season) were obtained using equation as stated below:

$$AGR = \frac{W_2 - W_1}{\Delta T}$$

Where:

- AGR : Absolute Growth Rate
 W_1 : Biomass at first reading
 W_2 : Biomass at second reading
 ΔT : Different in time

$$RGR = \frac{\ln(W_2) - \ln(W_1)}{\Delta T}$$

Where,

- RGR : Relative Growth Rate
 \ln : Longitude
 W_1 : Biomass first reading
 W_2 : Biomass second reading
 ΔT : Different in time

$$E_G = \Delta W / \Delta T \times (1 / A_L) \\ = (W_2 - W_1 / T_2 - T_1) \times (1 / A_L)$$

Where,

- E_G = growth efficiency
 A_L = total leaf area
 W_2 = weight at second reading
 W_1 = weight at first reading
 T_2 = time at second reading
 T_1 = time at first reading

2.4.2 Sampling procedure for gas exchange

Prior to the destructive sampling, the same samples used in the growth measurement were selected in this experiment. For every sample, three measurements were taken using gas exchange analyzer (Model LI-6400, LICOR, inc. Lincoln,

Nebraska USA). This measurement was done to determine the net photosynthesis rate (A_{net}), stomatal conductance (G_s) and transpiration rate (E_L). All the measurements were taken at 0800 to 1100 h to avoid the midday depression in photosynthesis as being recommended by Hiromi *et al.* (1999).

In addition, water use efficiency (WUE) is expressed as the quotient of the diffusive fluxes of CO_2 into the leaf and water vapour out of the leaf during photosynthesis (Farquhar and Richards, 1984). WUE is a fundamental aspect of the physiology of terrestrial plants. The parameter determine the efficiency of plant bring in carbon dioxide for photosynthesis without losing much water out of its stomata. It is the ratio of carbon dioxide intake to water lost through transpiration. All the gas exchange measurements were carried out on the fully expanded and non-senescing leaves at the fifth node from the apex.

2.5 Statistical analysis

Data of growth and physiological of Kenaf were analyzed using analysis of variance (ANOVA) (SPSS version 16.0) to estimate the treatment variations while Duncan's Multiple Range Test (DMRT) were used to detect the significant grouping among the treatments. In addition, AGR , RGR and E_G were analyzed using mean separation test. Correlation for height, diameter and aboveground biomass were analyzed using SigmaPlot version 10.0.

3. Results

3.1 Growth Parameters

3.1.1 Diameter and Height

There were contrasting results found for diameter and height of *Hibiscus cannabinus* L. grown in two different seasons between treatments. A highly significant difference between treatments was found in the wet season for both parameters (Table 1). However, the result showed clearly that both parameters were not significantly different in the dry season (early stage of planting).

The mean values of diameter ranged from as low as 8.54 mm (High) to 10.12 mm (Low) in the dry season and from 12.62 mm (Low) to 17.46 mm (High) in the wet season. Meanwhile, the mean values of height ranged from 79.4 cm (High) to 89.6 cm (Medium) and 178.3 cm (Low) to 240.4 cm (High) in the dry and the wet season respectively. In the wet season, the values of both parameters increased with increasing the level of fertilizer but no increasing patterns were found in the dry season (Table 2).

3.1.2 Leaf Number and Leaf Area

Significant differences were only found in the wet season for leaf number (LN) and leaf area (A_L). Significant difference at $p \leq 0.01$ was found for LN and at $p \leq 0.05$ for A_L (Table 3).

The mean values of LN and A_L for both seasons were shown in Table 4. In the dry season, there were no increasing trends with increasing the level of fertilizer found for LN but the increasing trend was only found in the wet season. Similar results were also found for A_L . However, DMRT grouping shown that medium and high application of fertilizer did not differ to each other with only small fraction of differences between them for both LN and A_L . The differences were about 1% and 4% for LN and A_L respectively.

3.1.3 Biomass

Table 5 shows the analysis of variance (ANOVA) for the plant biomass for two seasons. The plant biomass was divided into leaf mass (M_{Leaf}), stem mass (M_{Stem}), root mass (M_{Root}) and flower mass (M_{Flower}). Total aboveground biomass (M_{AG}) was calculated by summing the values of M_{Leaf} and M_{Stem} . Root to shoot ratio ($Rt:St$) was also calculated by dividing the values of M_{Root} and M_{AG} . Significant differences between treatments were found at $p \leq 0.05$ for M_{Leaf} and M_{root} in the wet season only. However, significant differences were found for the rest of the parameters in both seasons.

Mean values for biomass components were shown in Table 6 for two seasons. Biomass for stem, flower and aboveground biomass increased with the increasing of fertilizer level. Meanwhile, the DMRT grouping for biomass components for both seasons shows inconsistent results except biomass for root, flower and aboveground stem.

Based on the correlation analyses conducted using SigmaPlot 10.0 (Systat Software Inc.), there were no strong correlations between diameter and height with total aboveground biomass for every treatment except for height in the high level of fertilizer in the dry season (Figure 1). However, the results of correlation analyses between growth parameters and aboveground biomass were more profound in the wet season (Figure 2). Both height and diameter showed high correlation except for correlation between aboveground biomass and height at high level of fertilizer showed low correlation ($R^2 = 0.47$).

In addition, the true growth performance of *H. cannabinus* can be determined based on absolute growth rate (AGR), relative growth rate (RGR) and growth efficiency (E_G) of biomass. The values for AGR , RGR and E_G were only able to be calculated based on the mean values of biomass in both time of measurements (dry and wet) due to the samples were destructed. The increasing trends with increasing the level of fertilizer were found for AGR and RGR but no specific trend

was observed for E_G (Figure 3). The substantial increased of AGR was observed from the low to the medium level of fertilizer application and the degree of increment was starting to reduce from the medium to the high level of fertilizer application. Growth efficiency from the medium level of fertilizer application is low because it has higher reading in leaf area. The values of increment relative to the final biomass or relative growth rate were found to increase constantly from the low to the medium and to the high rate of fertilizer application. The value was found higher in the low level of fertilizer application followed by the high and medium level of fertilizer application for E_G .

3.2 Gas exchange

The analysis of variance (ANOVA) revealed that photosynthesis rate (A_{net}) and water use efficiency (WUE) were significantly different at $P \leq 0.05$ in the dry season while contrasting results were found in the wet season where only A_{net} was not significant (Table 7).

In the dry season, the mean value of net photosynthesis (A_{net}) was found higher in the high level of fertilizer application followed by the medium and the low level of fertilizer application. Similar results were also observed for water use efficiency (WUE), where the mean values increased with increasing the level of fertilizer i.e. 3.96 (low), 4.22 (medium) and 4.43 (high) in respectively. In the wet season, the mean value of stomatal conductance (G_s) was found higher in the high level of fertilizer application followed by the medium and the low level of fertilizer application. Similar results were also found in transpiration rate (E). However, the mean value of water use efficiency (WUE) was only found significantly higher in the medium level of fertilizer (Table 8).

4. Discussion

4.1 Growth parameter

Diameter and height during the dry season showed that there were no significant differences found between treatments with regards to the level of fertilizer concentrations. This is due to the poor growth performance of plants when the water level in soil was inadequate. Plants need sufficient amount of water to survive and keep their cells in good conditions at the early stage of development in order to produce new tissues and cells. In addition, the readings were taken after 41 days of planting, thus the differences in terms of performance is too soon to be detected. According to Muchow and Wood (1980), Kenaf plants under continuous irrigation vigorously increased plant height from the beginning of the experiment to its termination at the 10th week. Height increased slowly with the plants under stress, and severe stress had the most detrimental effect on height increment. In terms of biomass component, roots biomass showed outstanding performance compared to aboveground biomass. This is due to the need of more water to absorb at the dry season. The second measurement was taken during the wet season. The readings for both parameters showed significantly different between the level of fertilizer. The high and medium levels of fertilizer were obviously different with the low level where the different in terms of concentration between medium and low was 560 kg/plot. Thus, the medium level of fertilizer should be considered as most economic silvicultural practices to produce the best level of growth performance so far.

Leaf number and leaf area were not significantly different among treatments in the dry season and the mean values were also low. The plants might reduce the numbers of leaf to deter loss of water during the dry season through the process of transpiration. According to Charles-Edwards *et al.* (1983), daily increment in aboveground growth, depends on the proportion of incident light intercepted by the crop and the crop potential daily photosynthetic integral is obviously a function of the daily radiation integral. This might due to the early stage of planting where the differences in terms of growth were relatively not varied and the effect of fertilizer also still at the early stage. According to Tanner and Sinclair (1983), at the early stage of trees development, the differences were not significant. This phenomenon was also reported by Gartlan (1986). In contrast, leaf number and leaf area were significantly different for every level of fertilizer rate in the wet season. The high and medium levels of fertilizer were significantly affecting the number of leaves and leaf area. This will enhance plant to absorb light and produce food for the plants during the process of photosynthesis. Thus, this will also influence the level of plant water usage.

Biomass component during the dry season showed no differences between the parameters. However, during the wet season, leaf mass and root mass revealed the significant differences between the levels of fertilizer. Referring to the leaf mass at high and medium level of fertilizer, the results were better than low rate of fertilizer. This is due to the atmospheric and soil water content at the wet season is higher than the dry season. This phenomenon will influence plants to increase their leaf mass to enhance the respiration, transpiration and photosynthesis process. The high potassium (K) is needed for the process of respiration. This will encourage the plant stomata opening and energy production for photosynthesis process. This condition will influence the plant nutrient requirement at the maximum level.

For instance, in natrophilic (salt tolerant) plants, the K requirements can be lower. Plant requirement for K varies with the stage of growth, with the highest uptake rates often attained during the vegetative stage (Marschner, 1986). Negative correlation between increasing leaf K concentration and the concentrations of Calcium (Ca) and Magnesium (Mg) in *Hibiscus rosasinensis*, is also consistent with the reports that high K concentrations in the soil cause a decline in the uptake of Mg and Ca in cereals (Mengel and Kirkby, 1987). However, K is known to have a positive effect on the growth

and dry matter accumulation of plants (Mengel and Kirkby, 1987). However, it is similar that the chemical analysis in the soil and also in the plant could not be carried out due to the time constraint and equipment limitation.

4.2 Gas exchange

Net photosynthesis was not significantly different during wet season. This could be due to the requirement for light at the early stage of growth is more profound compared to the matured plants. High level of fertilizer applied was highly affected net photosynthesis, transpiration rate and water use efficiency. A common response to water stress is stomatal closure, which reduces both flux of CO₂ and water vapour. Alternatively, stomates may remain open while turgor is maintained through osmotic adjustment. Stomatal conductance and transpiration rate in Kenaf progressively declined with age in the adequately watered control. All the levels of stress brought stomatal conductance and transpiration to zero. Kenaf was also observed to roll its leaves during drought. These two mechanisms could be described as drought tolerance by dehydration postponement (Kramer, 1983), equivalent to drought avoidance by Levitt (1980). In wet season, stomatal conductance, transpiration rate and water use efficiency was significant but not for net photosynthesis rate. Wet moisture in the air might be affect stomatal conductance that reduce water through vapour to balance physiological plants.

4.3 Relationship between growth parameters and physiological attributes

Stem development and elongation are the critical components of the growth process (Schulze and Chapin, 1987). Physiological efficiency of any particular fibre species is manifested in the increment of plant height and increase in basal diameter. These parameters which result from the respective activities of apical growth and intercalary growth are generally considered dependable yield components of a bast fibre crop. Water deficit during dry season was observed to have significantly reduced height and diameters in growth parameter independent to the levels of fertilizer. In addition, when water supply is limited, turgor pressure approaches the yield threshold, and growth depends on the rate of water supply. This phenomenon was proven during the wet season where results of diameter and height were highly significant. Levels of fertilizer also contribute to the growth performance where high level of fertilizer gave better growth performance and otherwise. In addition, water use efficiency showed that the optimum level of fertilizer is medium compared with high and low. The alteration of these growth parameters under water deficits (in dry season) is due in part to the role of water in turgidity maintenance necessary for cell enlargement (Kramer, 1983). Cell division also decreases with increased water deficits, because cells apparently must attain a certain size before they can divide. As there is no direct method for assessing the fibre yield from a standing crop, plant height and basal diameter are considered as the general guiding criteria for efficient production of fibres in a particular species (Maiti and Chakravarty, 1977). It can therefore be concluded that drought affects the efficient production of fibres in Kenaf.

Leaf growth is the most sensitive of plant processes to water deficits and is frequently inhibited in field crops (Hsaio, 1973; Schulze and Chapin, 1987). For a given location and growth duration, the amount of light intercepted is primarily dependent on leaf area development, and these have been shown to be directly linked to leaf turgor. There were variable results between wet and dry seasons. During the dry season, water deficit condition adversely affected number of leaves, leaf mass and leaf area due to poor leaf expansion and defoliation. Plants usually grow straight and unbranched in dense stands in allocation energy for transform food to growing stem mass. At the same time during dry season, net photosynthesis rate and water use efficiency have significant different.

In the wet season, only net photosynthesis rate was not significant between other physiological parameters. This might due to plant need to reduce net photosynthesis because the leaf number and area were low. However, at this period, growth efficiency gave the highest performance at the low level of fertilizer. In addition, wet moisture in the air might be affect stomatal conductance that reduce water through vapour to balance physiological plants and at the same time photosynthesis rate was reduced. During the wet season, medium level of fertilizer showed outstanding performance compared to low and high level of fertilizer. Based on the growth performance and physiology characteristics, medium level of fertilizer is the best rate to produce good stands of Kenaf at BRIS soils compared to low and high level of fertilizer.

5. Conclusion

Based on the growth attributes for 41 (dry season) and 64 days (wet season) of planting, the effect of fertilizer on the growth traits were varied and more profound at the age of 64 days of planting. The results couldn't be treated at a general statement of V36 performance on BRIS soils due to the number of site replication where the research conducted was at one particular place (Setiu, Terengganu) instead of all BRIS soil sites in Malaysia. In terms of gas exchange characteristics, the changes of stomatal conductance, transpiration rate and water use efficiency were greater after 64 days of planting. However, the photosynthesis rate is otherwise. The study should be repeated to verify the findings. In addition, the knowledge on gas exchange can support for better understanding of physiology of Kenaf on its water requirements and its ability in light conversion in carbonaceous molecules influencing the biomass production and, indirectly, the carbon sequestration activity in different rate of fertilizer application. In addition, knowledge on Kenaf's carbon balance,

as a response of assimilation and respiration rate to environmental conditions, is fundamental for understanding and assessing crop growth and productivity.

Acknowledgement

We have to remember Mr. Mohd Khairil Saufi for his help during the field experiment. This project wouldn't be reality without supports from Lembaga Tembakau Negara for providing land and plant materials. The project was funded by UPM under the competitive grant UPM/RUGS/91049.

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Table 1. The summary ANOVA of diameter and height parameters of *Hibiscus cannabinus* L. with regard to the level of fertilizer

Season	Fertilizer	Diameter (<i>D</i>)			Height (<i>Ht</i>)	
		df	Mean Square	F Value	Mean Square	F Value
<i>DRY</i>	Fertilizer	2	3.66	0.95 ^{ns}	146.07	1.23 ^{ns}
<i>WET</i>	Fertilizer	2	60.52	5.92**	9873.43	9.74***

*Significantly different at $P \leq 0.01$ and $P \leq 0.001$

Table 2. The mean values of diameter and height of *Hibiscus cannabinus* L. with regard to the level of fertilizer in the dry and the wet seasons

Parameter	Level of fertilizer		
	Low	Medium	High
	<i>DRY</i>		
<i>D (mm)</i>	10.12 ± 1.02 ^a	9.91 ± 0.96 ^a	8.54 ± 0.59 ^a
<i>Ht (cm)</i>	81.4 ± 5.81 ^a	89.6 ± 5.79 ^a	79.4 ± 2.04 ^a
	<i>WET</i>		
<i>D (mm)</i>	12.62 ± 1.06 ^b	15.77 ± 1.05 ^a	17.46 ± 0.92 ^a
<i>Ht (cm)</i>	178.3 ± 10.57 ^b	217.7 ± 10.43 ^a	240.4 ± 9.14 ^a

Mean values followed by the same letter in the same row do not differ statistically as per Duncan's Multiple Range Test at $P \leq 0.05$.

Table 3. The summary of ANOVA for leaf number and leaf area parameters of *Hibiscus cannabinus* L. with regard to the rate of fertilizer

Season	Fertilizer	Diameter (<i>D</i>)			Height (<i>Ht</i>)	
		df	Mean Square	F Value	Mean Square	F Value
<i>DRY</i>	Fertilizer	2	966.47	2.16 ^{ns}	34944.5	0.63 ^{ns}
<i>WET</i>	Fertilizer	2	4969.3	6.50**	2037083	3.64*

*Significantly different at $P \leq 0.05$ and $P \leq 0.01$

Table 4. The mean values of leaf number and leaf area of *Hibiscus cannabinus* L. between the rate of fertilizer in the dry and the wet seasons

Parameter	Level of fertilizer		
	Low	Medium	High
	<i>DRY</i>		
<i>LN</i>	62.6 ± 14.9 ^a	44.0 ± 6.4 ^a	35.4 ± 2.4 ^a
<i>A_L</i>	620 ± 147 ^a	524 ± 88.3 ^a	453 ± 65 ^a
	<i>WET</i>		
<i>LN</i>	47.4 ± 6.5 ^b	84.7 ± 8.0 ^a	87.2 ± 11.1 ^a
<i>A_L</i>	705 ± 124 ^b	1410 ± 253 ^a	1546 ± 298 ^a

Mean values followed by the same letter in the same row do not differ statistically as per Duncan's Multiple Range Test at $P \leq 0.05$.

Table 5. ANOVA of biomass parameters of *Hibiscus cannabinus* L. between the level of fertilizer in the dry and the wet seasons

Parameter	DRY			WET	
	df	Mean Square	F Value	Mean Square	F Value
M_{Leaf}	2	2.81	0.65 ^{ns}	357.71	5.07*
M_{Stem}	2	10.12	1.23 ^{ns}	2946.81	1.31 ^{ns}
M_{Root}	2	0.52	0.16 ^{ns}	307.47	3.41*
M_{Flower}	2	-	-	195.15	3.18 ^{ns}
M_{AG}	2	18.89	0.84 ^{ns}	10174.9	2.54 ^{ns}
$Rt:St$	2	0.02	1.92 ^{ns}	0.03	1.41 ^{ns}

*Significantly different at $P \leq 0.05$

Table 6. The mean values of biomass parameters of *Hibiscus cannabinus* L. between the level of fertilizer in the dry and the wet seasons

Parameter	Level of fertilizer		
	Low	Medium	High
<i>DRY</i>			
M_{Leaf}	4.74 ± 1.33 ^a	3.80 ± 0.77 ^a	3.26 ± 0.48 ^a
M_{Stem}	7.09 ± 1.27 ^a	7.92 ± 1.71 ^a	5.15 ± 0.64 ^a
M_{Root}	2.39 ± 0.57 ^a	2.46 ± 0.48 ^a	2.98 ± 1.17 ^a
M_{Flower}	-	-	-
M_{AG}	11.83 ± 2.50 ^a	11.72 ± 2.47 ^a	8.41 ± 1.12 ^a
$Rt:St$	0.20 ± 0.01 ^a	0.21 ± 0.03 ^a	0.32 ± 0.08 ^a
<i>WET</i>			
M_{Leaf}	6.37 ± 1.16 ^b	17.88 ± 3.40 ^a	14.95 ± 2.87 ^a
M_{Stem}	50.05 ± 18.47 ^a	76.62 ± 12.85 ^a	82.16 ± 13.02 ^a
M_{Root}	8.74 ± 2.42 ^b	18.69 ± 3.63 ^a	17.95 ± 2.83 ^a
M_{Flower}	2.12 ± 1.46 ^b	4.26 ± 2.21 ^{a,b}	10.61 ± 3.38 ^a
M_{AG}	61.70 ± 22.19 ^b	107.05 ± 20.83 ^{a,b}	123.23 ± 16.58 ^a
$Rt:St$	0.17 ± 0.01 ^a	0.20 ± 0.02 ^a	0.16 ± 0.001 ^a

Mean values followed by the same letter in the same row do not differ statistically as per Duncan's Multiple Range Test at $P \leq 0.05$.

Table 7. Analysis of variance of gas exchange of *Hibiscus cannabinus* L. between the level of fertilizer in the dry and the wet seasons

Parameter	DRY		WET	
	Mean Square	F Value	Mean Square	F Value
<i>Anet</i>	273.97	16.92*	9.65	0.39 ^{ns}
<i>G_S</i>	0.03	0.94 ^{ns}	0.09	5.57*
<i>E</i>	3.96	2.40 ^{ns}	9.21	9.22*
<i>WUE</i>	1.68	3.90*	4579.66	7.50*

*Significantly different at P ≤ 0.05

Table 8. The mean values of gas exchange of *Hibiscus cannabinus* L. between level of fertilizer in the dry and the wet seasons

Parameter	DRY			WET		
	Low	Medium	High	Low	Medium	High
<i>Anet</i>	11.34 ^b	12.07 ^b	17.07 ^a	13.39 ^a	12.48 ^a	12.35 ^a
<i>G_S</i>	0.27 ^a	0.24 ^a	0.21 ^a	0.27 ^b	0.34 ^a	0.38 ^a
<i>E</i>	2.79 ^a	2.55 ^a	2.03 ^a	2.53 ^b	3.29 ^a	3.60 ^a
<i>WUE</i>	3.93 ^b	4.22 ^{ab}	4.43 ^a	33.46 ^b	45.49 ^a	33.46 ^b

Mean values followed by the same letter in the same row do not differ statistically as per Duncan’s Multiple Range Test at P < 0.05.

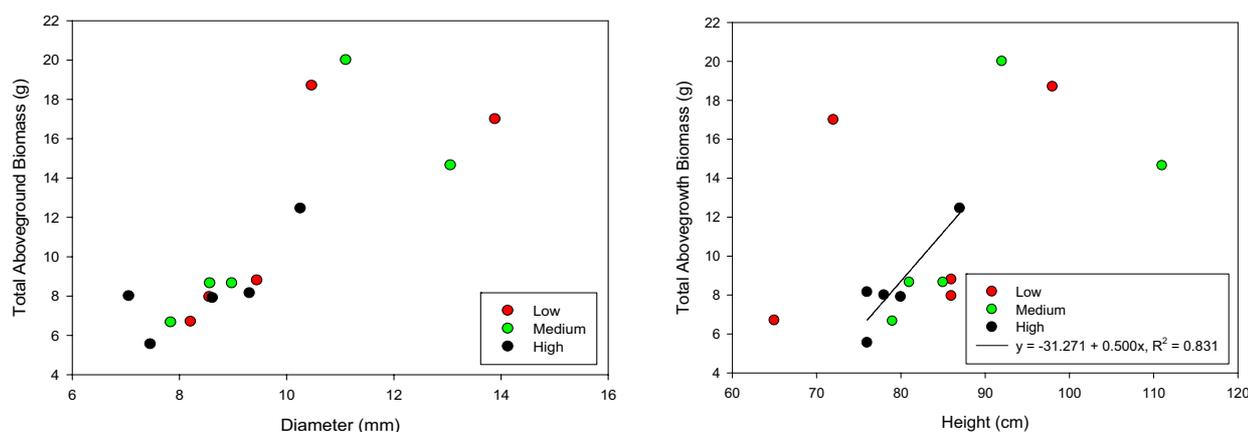


Figure 1. Correlation between total aboveground biomass with diameter and height in the dry season

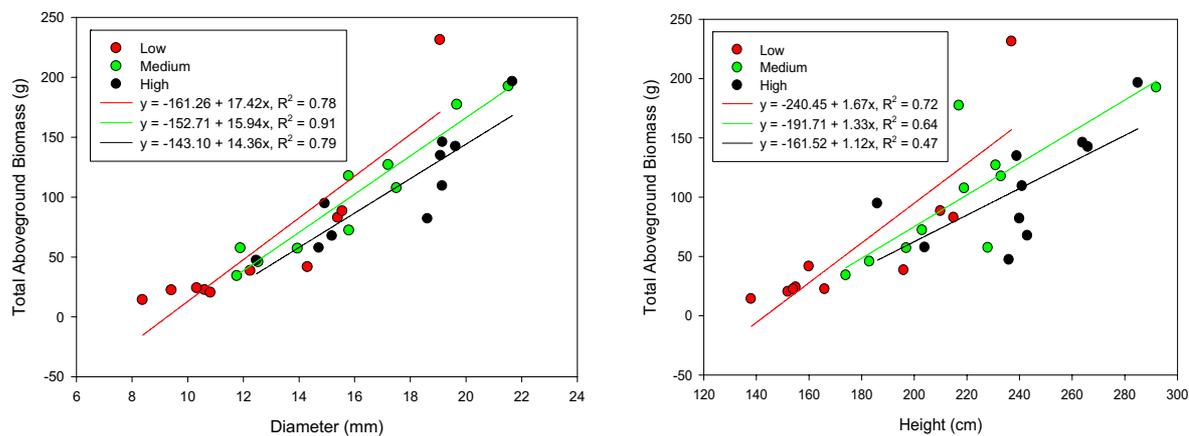


Figure 2. Correlation between total aboveground biomass with diameter and height in the wet season.

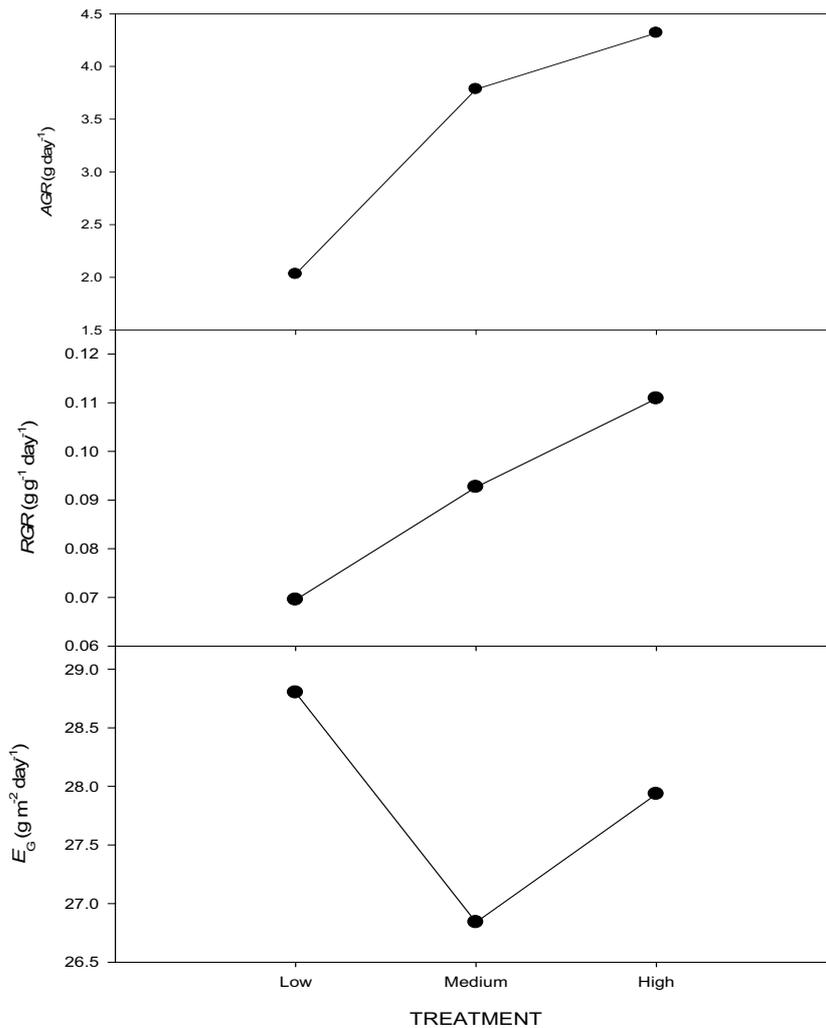


Figure 3. The summary of AGR, RGR and E_G parameters of *Hibiscus cannabinus* L. at the different levels of fertilizer in the dry and the wet seasons



The Effect of Acidification and Magnetic Field on Emitter Clogging under Saline Water Application

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Abstract

Up to day, drip irrigation systems have reached to a high level of technology. But, these systems are not able to show their potential benefits, due to various reasons. Emitter clogging can affect distribution uniformity and the system performance, which has direct relationship with water quality. In this study five types of emitters with different nominal discharges, with or without self-flushing system and with or without pressure compensating system were evaluated under three management schemes; untreated well water (S1), acidic treated water (S2) and magnetic treated water (S3) in order to reduce chemical clogging. Flow reduction rate, statistical uniformity coefficient (Uc), emission uniformity coefficient (Eu) and variation coefficient of emitters' performance in the field (Vf) were monitored. The emitter performance indexes (Uc and Eu) decreased during the experiment due to emitter clogging. The Uc and Eu values in different management schemes confirmed that the acidification has better performance than the magnetic water in order to control emitter clogging and keep high distribution uniformity. Regarding to Vf values, the priority of untreated and treated water was as $S2 > S3 > S1$ for each emitter.

Keywords: Drip irrigation system, Discharge, Emitter, Acidification, Magnetic treatment

1. Introduction

Drip irrigation systems are designed to deliver a certain amounts of water and nutrient to the plant. As water becomes more limited in arid and drought prone areas, these systems are used more widely. There are numerous advantages using drip irrigation systems. However, these advantages could be nullified by emitter clogging, which is directly related to the quality of the irrigation water (Dehghanisani et al., 2007; Gilbert et al., 1979). Therefore, drip irrigation systems need a high maintenance. The greatest problem and concern dealing with these systems is emitter clogging. Nakayama and Bucks (1981) found a significant reduction in uniformity when 1-5% of emitters were completely clogged even with 2 to 8 emitters/plant. Partial or complete clogging drastically reduces water application uniformity (Nakayama and Bucks, 1981) and consequently decreases irrigation efficiency and crop production. Bucks and Nakayama (1982) proposed an irrigation water quality classification for potential clogging hazard. They classified the hazard into three main categories of physical, chemical and biological clogging that play major roles in the clogging process. Physical clogging may be caused by factors such as suspended inorganic materials (sand, silt, clay or plastic particles) and organic materials (plant and animal residuals, etc.). Biological clogging is due to organic sediments plus iron or hydrogen sulfide that accumulated

in emitters and laterals (Dehghanisani et al., 2005; Tajrishy et al., 1994). Chemical clogging is contributed to precipitation of calcium carbonate that is common in arid regions with waters rich in calcium and bicarbonates. Iron deposits (ocher) have created severe clogging primarily in the United States. The soluble and reduced form of Iron (ferrous ion or Fe^{+2}) is presented in groundwater of many places. Manganese might be oxidized and transform into particulate form that cause emitter clogging.

Chemical clogging, through salt precipitation, is very difficult to be controlled. The general recommendation to prevent chemical clogging is to lower the water pH, by acid injection, to a value such that salt precipitation does not occur.

Water passing through magnetic field might get new properties and is called magnetic water. Magnetic treatment (MT) of hard water is currently used to prevent scale formation on hot surfaces, in particular in heat exchangers, as well as in domestic equipments. This treatment process has been developed to substitute chemical water treatment methods employing chemical product that might be harmful to the environment and human health.

Comprehensive experimental researches were carried out on modification of CaCO_3 precipitation process by MT. According to the literature, the efficiency of magnetic treatment depends on numerous parameters. For example, Chibowski et al. (2003) and Barrett and Parsons (1998) found out that the magnetic treatment applied on hard water decreased the quantity of scale formation on the wall. The principle of this phenomenon has not been understood well. Various contradictory hypotheses have been suggested that attributed these to the Lorentz forces $\vec{F} = q \cdot \vec{v} \times \vec{B}$ exerted either on moving ions or on charged solid particles (Higashitani et al., 1993). The magnetic field (MF) would be able to disturb the double ionic layer surrounding the colloidal particles and their zeta potential (Gamayunov, 1983; Higashitani and Oshitani, 1998; Parsons et al., 1997).

The objective of this study was to investigate the effectiveness of three management schemes in decreasing chemical clogging and evaluate the change in discharge rate and uniformity of laterals equipped with different type of emitters.

2. Materials and methods

2.1 Experimental location

The climate of Iran is one of the greatest extremes due to its geographic location and variation in topography. The summer is extremely hot with temperatures in the interior rising possibly higher than anywhere else in the world; certainly over 55°C has been recorded. Iran with an area of 165 million hectares has 37 million hectares arable land of which only 8 million hectares are irrigated, 6 million hectares are rain-fed, and 4.5 million hectares remain in the form of fallow land. Annual rainfall ranges from less than 50 mm in the deserts to more than 1600 mm on the Caspian Plain. The average annual rainfall is 252 mm and approximately 90% of the country is arid or semiarid. Overall, about two-thirds of the country receives less than 250 mm of rainfall per year. Most of the rain falls during the winter season, particularly in northern part of country. In the central and southern part of Iran, the annual rainfall range from 0 to 200 mm.

About half of the area is irrigated by groundwater, including spring water. Over extraction from ground water resources and limited rainfalls have led receding ground water table and poorer water quality over time. From the total 8.1 million ha of irrigated lands in Iran; 7.6 million ha (95%) are under surface irrigation and 0.4 million ha (5%) under the pressurized irrigation. With limited renewable water resources and annual rainfall, particularly during the past two decades, drip irrigation systems have been introduced in agricultural regions of Iran to increase water use efficiency. However, water quality reduction over time has led emitter clogging in most of drip irrigation systems.

2.2 Experimental layout

A field study involving a drip irrigation system was conducted in Hasanabad, Iran ($35^\circ 25'\text{N}$, $51^\circ 10'\text{E}$) from August to November 2007. Five different type of emitters (see Table 1), differing in the nominal discharge, pressure compensating and non-pressure compensating were used (Dehghanisani et al., 2005, 2007). The in-line (EM3 and EM5) and on-line emitters (EM1, EM2 and EM4) were 0.5 m apart. Water pressure at the inlet was 0.10 MPa. Figure 1 illustrates a schematic layout of the drip irrigation system, including three similar subunits, in the field. Three management schemes with application of non-treated water (S1), treated water with acid injection (S2) and magnetic water (S3), each in a subunit, were evaluated for the proposed emitters. Each subunit consisted of 5 laterals with 50 m length and 0.5 m apart, was connected to the sub-main pipe of the drip irrigation system. Each lateral was equipped with a flow meter. No crops were grown in the field during the study. The irrigation system was automated to irrigate 8 hours per day (9:00 am to 17:00 pm) over the 12 weeks in summer of 2007. All subunits were connected to a control station equipped with a pump, backflow-prevention device, screen and silicone filters and pressure gauges.

Sulfuric acid was injecting into subunit 2 continuously in order to lower pH from 8.18 to 6.5 and decrease the possibility of CaCO_3 precipitation. The flow rate of emitters was measured every two weeks and according to ASAE EP405 method (ASAE Standards, 2003b). Twenty five catch-cans were placed under the emitters along each lateral. Water samples were

collected every two weeks and electrical conductivity, pH, Cat ions such as calcium, sodium and magnesium and anions such as carbonate, bicarbonate and sulfate were measured.

2.3 Emitter performance

Laboratory tests were performed to determine the manufacturing coefficient of variation (V_m) of new emitters using Solomon's equation (1979):

$$V_m = 100 \frac{Sq_e}{\bar{q}_e} \quad (1)$$

Where Sq_e and \bar{q}_e are the standard deviation and the average of emitter discharge (Lh^{-1}), respectively. The V_m values ranged from 3.05 to 7.69 percent (Table 1) which attribute a good quality class to emitters, according to ASAE standard method of EP405.1 (ASAE Standards, 2003a). The emitters with a given size and $V_m < 5$ percent were selected to ensure minimum manufacturing variation in discharge in the field.

In the second test of this study, 50 emitters with a given size, selected on the basis of least variation (Equation 1) were fitted to a lateral in order to assess emitter discharge performance at five different pressure heads. The emitter discharge performance is characterized by the k and x parameters of Karmeli's (1977) Equation:

$$q_e = k h^x \quad (2)$$

Where q_e is discharge from individual emitter (Lh^{-1}), h is entry water pressure (m H_2O ; 1 m H_2O = 0.098 \times 105 Pa), k is emitter discharge coefficient and x is emitter discharge exponent, which characterizes the flow regime and q vs. h relationship.

2.4 Statistical analysis

Three statistical factors were used to assess the emitter performance in the field: uniformity coefficient (Uc), emission uniformity coefficient (Eu) and the coefficient of variation of emitters (Vf) along a lateral in the field.

The statistical uniformity coefficient (Uc) provides a measure of deviation from average conditions on each lateral. It was computed using the modified form of Wilcox and Swailes (1947) equation proposed by Bralts et al. (1987) for drip irrigation system:

$$Uc = 100 \left(1 - \frac{Sq}{\bar{q}} \right) \quad (3)$$

Where, Sq is standard deviation of emitter discharge (Lh^{-1}) and \bar{q} is average of emitter discharge in the field on a given lateral (Lh^{-1}).

The emission uniformity coefficient (Eu) was computed using Capra and Tamburino (1995) equation:

$$Eu = 100 \frac{\bar{q}_{1/4 \min}}{\bar{q}} \quad (4)$$

Where $\bar{q}_{1/4 \min}$ is the mean of low quarter of emitter discharge (Lh^{-1}). Uc and Eu assume a different meaning, the former showing deviation from average conditions, and the latter showing the conditions of the least watered plants as compared with that of the average watered plants.

The coefficient of variation of the emitters' performance along a lateral line in the field (Vf %) shows the variation of emitter discharge at a constant pressure head. It was computed using Bralts' (1986) equation:

$$Vf = \sqrt{\left(\frac{Sq}{\bar{q}} \right)^2 - x^2 \left(\frac{S_h}{h} \right)^2} \quad (5)$$

Where S_h the standard deviation of pressure is head (m H_2O), and h is the mean pressure head (m H_2O). Other parameters were already defined in equation 3.

3. Results and discussion

3.1 Water quality

Table 2 shows some chemical characteristics of the non-treated (well) water and treated (magnetic) well water. This table indicates that the magnetic field does not change the chemical properties of the well water significantly. Figure 3 illustrates the variation of pH and EC of the well water during the field experiment. The pH values were higher than 8 all over the experiment, indicating a feasible precipitation of carbonate calcium in the system and consequently emitter clogging.

Equilibrium equations (6, 7 and 8) were used to assess the possibility of calcium sulfate precipitation in the system. If multiplication (product) of calcium and sulfate ions (mol/l) is greater than KSP, it may be resulted in precipitation of calcium sulfate. Value of KSP for calcium sulfate in 25°C is 2.4×10^{-5} ($\text{mol}^2 \cdot \text{l}^{-2}$).



$$\frac{[\text{Ca}^{2+}][\text{So}_4^{2-}]}{\text{CaSo}_4} = K \quad (7)$$

$$[\text{Ca}^{2+}][\text{SO}_4^{2-}] = K [\text{CaSO}_4] = \text{KSP} \quad (8)$$

According to Nakayama and Bucks (1991) pH levels of <7.0, 7.0–8.0 and > 8.0 could lead to slight, medium and severe clogging, respectively. Therefore, pH values of well water (table 2 and Figure. 3) certify sever clogging problems during the experiment. Although, it has been reported that the pH may not have a direct impact on clogging; but it could accelerate the chemical reactions or biological growth involved in clogging process (Nakayama and Bucks 1991; Dehghanisanij et al. 2004, 2005).

3.2 Emitter performance

The manufacturing coefficient of variation (Vm) values determined in the laboratory ranged from 3 to 8 (table 1). The minimum value (3.05%) was corresponded to EM5 and the maximum value (7.69%) to EM2. According to ASAE standard EP405.1 (ASAE standards, 2003a), Vm value between 3-11% classify emitters in a moderate category.

Figure 4 (a, b and c) shows the relative flow rate in different emitters during the experiment in subunits irrigated with well water, acidic water and magnetic water, respectively. This Figure illustrates a fairly continuous flow rate reduction in all emitters over the time, especially at the first month of the experiment, which could be attributed to emitter clogging.

Figure 4 also show a less reduction rate in emitters irrigated with acidic water, as compared to the ones irrigated with well and magnetic water. The reason can be explained by the fact that the acid injection to well water lowering the pH which consequently prevents and reduces carbonate precipitation.

Table 4 presents the total discharged and total nominal water and reduction rate in all emitters under different management schemes (well, acidic and magnetic irrigation water).

According to table 4, the maximum reduction of flow rate was occurred in EM4 (14.98%) and after that in EM3 (12.32%) irrigated with non-treated well water and the minimum reduction was occurred in EM2 (2.76%) irrigated with acidic water. The flow rate in EM1, EM2 and EM5 with self flushing feature did not reduce significantly during the experiment, even in subunit irrigated with untreated well water. Self flushing system allows the emitters to pass large particles but the orifice is constricted at normal operation pressure to regulate the flow rate (Hills and El-Ebaby1990).

Dehghanisanij et al. (2004, 2005 and 2007) showed that emitters with pressure compensating feature have smaller variation in emitter discharge than non pressure compensating emitters when low quality water was induced by algae and protozoa and they showed that emitters with self flushing and pressure compensating features have priority for use in drip irrigation under saline water.

The EM2 with the largest nominal discharge, self flushing and pressure compensating features showed less flow rate reduction than the other emitters in different management schemes.

Figure 5 shows the variation of Uc, Eu and Vf in different subunits irrigated with well, acidic and magnetic water. These parameters help us to evaluate the effect of emitter clogging and manufacturing coefficient of variation on flow rate changes of different emitters. Figure 5 shows a declined trend for Uc and Eu in different subunits over the time. However, the acidic water subunit had the highest values of Uc and Eu for all emitters. The Uc provides a measure of deviation from the average (Wilcox and swales 1947). According to Bralts (1986), the performance of emitters is classified and evaluated in three categories; “good” for the Uc values greater than 89%, “medium” for the Uc values between 71% and 89%, and “poor” for the Uc values less than 71%. In this study, the value of Uc was greater than 71% for all emitters under different water management and during the experiment. The high values of Uc belong to EM2 and the low values belong to EM3 and EM4. At the end of experiment, Uc values for EM2 were measured 93.1, 96.19 and 92.86 for well, acidic and magnetic water, respectively. These values for EM4 were measured 89.7, 93.89 and 87.94. The nominal discharge was the same for EM2 and EM4. The better performance of EM2 can be attributed to the self flushing feature. The nominal discharge in EM4 was higher than that in EM5. However, EM5 with self flushing feature showed better performance as compared to EM4.

The Eu parameter provides a measure of the least water consumed plants condition. According to Bralts (1986), if the Eu values are greater than 84%, then the performance is classified as high; between 66% and 84%, the performance is medium; and less than 66%, the performance is poor. According to Eu values, the performance of emitters in subunits is

organized as acidic>well>magnetic water. But the Eu value decreased suddenly at the end of experiment (The 10th of October) due to complete clogging of one emitter among 25 emitters.

Both Eu and Uc have negative correlation with the percentage of complete clogged emitters (Dehghanisani et al. 2007). EM5 and EM4 showed better performance as compared to the others in different water management schemes. The Eu value was greater than 84% for all emitters in acidic water subunit, so their performance classified as high. Consequently, the hazard of chemical clogging under saline water usage can be reduced by acidification and high performance can be promised.

The Vf values also show differences among different types of emitters (Figure. 5). According to Bralts (1986), emitters are classified as high, medium and poor performance on the basis of Vf values; less than 11%, between 11 and 29% and greater than 29%, respectively. As the Vf values become smaller, the variation of discharge among emitters on the laterals are decreased (Bralts 1986). According to Figure 6, the performance of all emitters (with the exception of EM4) in different subunits was high. The performance of EM4 in well, acidic and magnetic water subunits was medium, high and medium, respectively. These results suggest that the mist spray emitter type was characterized as less clogging and could be used for more than one irrigation season with good result.

4. Conclusion

The results of this study showed that the application of well saline water in drip irrigation system have the potential to induce emitter clogging. The concentration of Fe and Mg in well water was lower than the hazardous levels that could clog emitters. It was found that the flow rate reduction in emitters is affected by emitter characteristics and water treatment methods. The priority of emitters according to flow rate reduction and clogging are as follow: EM4> EM3> EM5> EM1> EM2.

It was found that the acid injection treatment provide better performance than the magnetic field. In the other hand, less flow rate reduction was occurred in emitters using acidic water. There was no significant difference between subunits using untreated well water and treated magnetic water. All the emitters used in this experiment (with the exception of EM4) were ranked as high performance class. The performance of EM4 with untreated well water and treated magnetic water was moderate and with treated acidic water was high. The Eu and Uc decreased over the time and increased by emitter clogging. Regarding to Eu and Uc, acidification was found to be one of the most effective method in controlling emitter clogging and conserving high distribution uniformity.

Acknowledgment

Appreciation is made for Mr. Hemn AhmadAali, Kaveh Ahmadzade, Kaveh Mollazade, Rahmat Mohammadi, Jalal Khorshidi, Younes Khoshkhoo, Zakarya Abdolla Amirani, and Mrs. Parang Nikmaram for their helps.

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Table 1. Some characteristics of the five different types of drip irrigation emitters

Emitter	Type of Emitter	Nominal			V _m %	Other Characteristics ^[a]
		Discharge (Lh ⁻¹)	k	x		
EM1	On - line PC	4	5.009	-0.0559	3.61	self-flushing
EM2	On - line PC	4	2.843	0.136	7.69	self-flushing
EM3	In - line PC	3.6	3.2846	0.036	4.04	anti-drain
EM4	On - line NPC	4	1.1957	0.5258	4.76	turbulent flow emitter
EM5	In - line PC	2.4	0.95	0.4202	3.05	turbulent labyrinth flow, self cleaning

^[a] According to manufacture's manual.

Note: PC = pressure compensating; NPC = non-pressure compensating; k = emitter discharge coefficient and x = emitter discharge exponent

Table 2. Mean chemical characteristics of the waters

Parameters	Unite	Well water	Magnetic water
EC	dS/m	4.8	4.76
pH*		8.19	8.09
Ca	(me/L)	9.7	9.65
Mg	(me/L)	9.08	8.85
Na	(me/L)	31.96	32.02
HCO ₃	(me/L)	7.3	7.01
SO ₄ ⁻²	(me/L)	21.46	23.4
Cl	(me/L)	21.29	20.05

* During acid injection, pH was less than 6.5

Table 3. Chemical analyses of well water and the Possibility of CaCO₃ and CaSO₄ precipitation

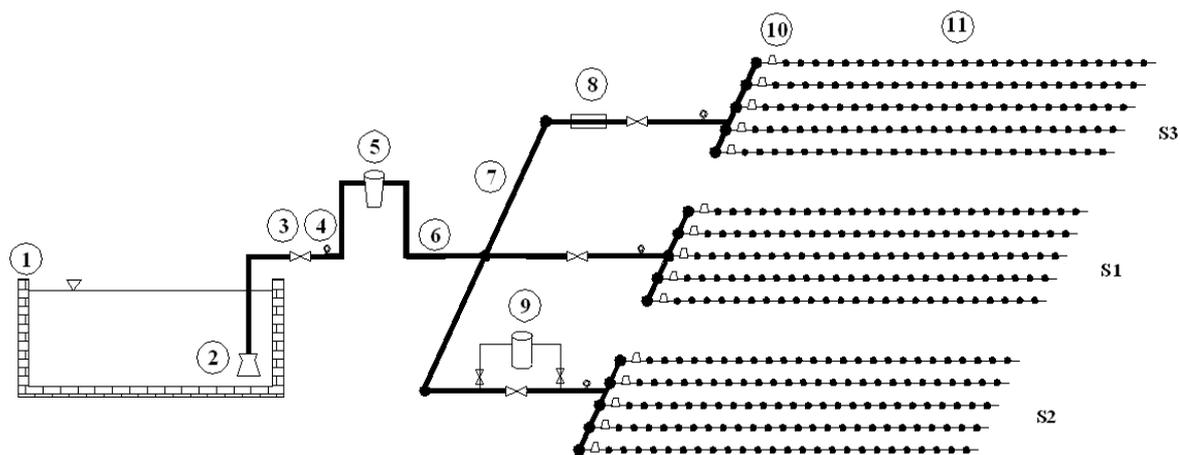
Date	pHm	pHc	LSI	Ca ⁺² (mol/l) × 10 ³	SO ₄ ⁻² (mol/l) × 10 ³	[Ca ⁺²][SO ₄ ⁻²] (mol ² /l ²) × 10 ⁵	Possibility of CaCO ₃ precipitation	Possibility of CaSO ₄ precipitation
8/24	8.12	7.01	1.11	18	47.4	85.32	+	+
9/7	8.05	7.02	1.03	18.8	50.16	94.3008	+	+
9/21	8.27	6.93	1.35	18	39.46	71.028	+	+
10/5	8.03	6.95	1.08	21	38.54	80.934	+	+
10/19	8.11	7.02	1.09	20.8	39.68	82.5344	+	+
11/2	8.02	6.87	1.16	17.4	43.62	75.8988	+	+
11/16	8.70	6.89	1.81	21.8	41.6	90.688	+	+

Table 4. Total discharged and nominal water and reduction rate in all emitters under different management schemes

Lateral No.	Total nominal water use (m3)	S1		S2		S3	
		Total water used (m3)	Reduction (%)	Total water used (m3)	Reduction (%)	Total water used (m3)	Reduction (%)
EM1	284.66	270.24	5.07	271.66	4.57	269.45	5.34
EM2	286.94	273.28	4.76	279.03	2.76	278.54	2.93
EM3	249.98	219.18	12.32	241.21	3.51	222	11.19
EM4	272.16	231.39	14.98	256.77	5.65	233.82	14.09
EM5	181.44	170.58	5.99	172.22	5.08	170.92	5.80



Figure 1. Activity area in Iran



- | | | | |
|--------------|-------------------|-------------------|---------------------------|
| 1. Reservoir | 4. Pressure gages | 7. Sub-main line | 10. Flow meters |
| 2. Pump | 5. Screen filter | 8. Magnetic Water | 11. Emitters and laterals |
| 3. Valves | 6. Main line | 9. Injector | |

Figure 2. Field experiment layout

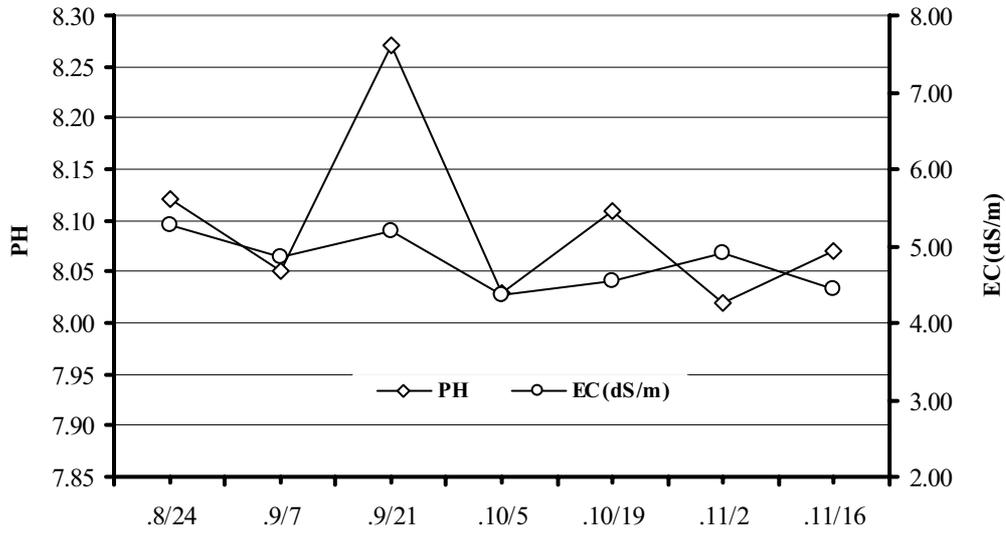


Figure 3. Variation of pH and EC during the experiment

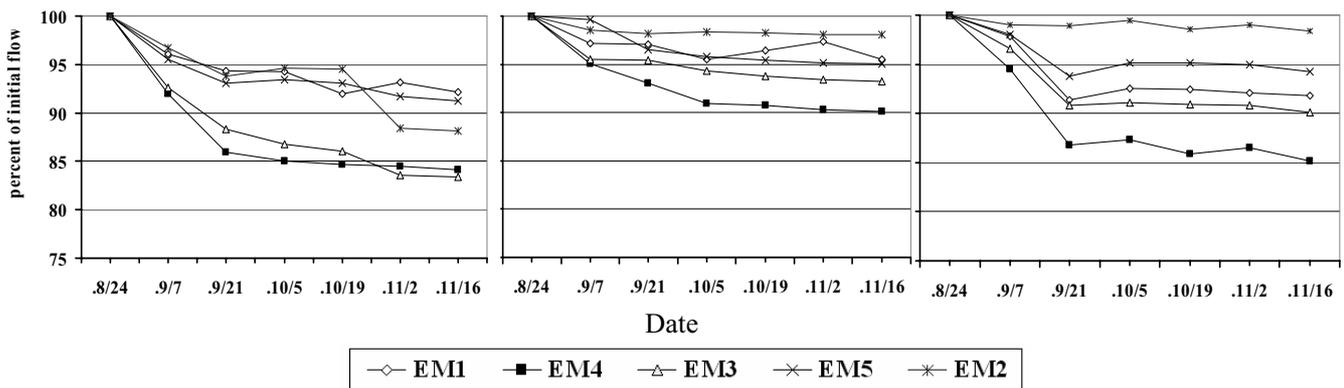


Figure 4. Variations of flow rate for different emitters

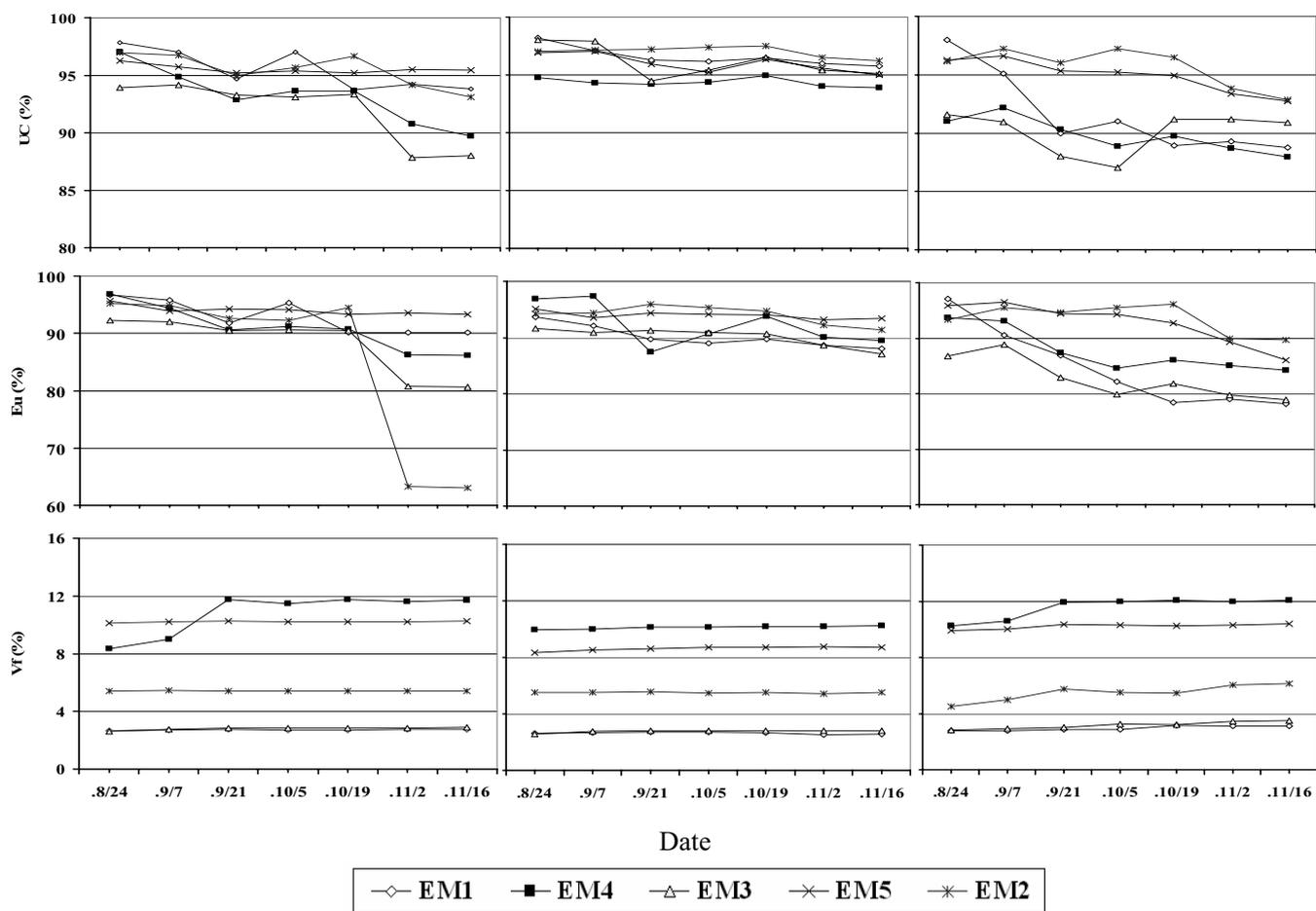


Figure 5. Variation of Uc, Eu and Vf in different subunits irrigated with well, acidic and magnetic water



Precision Forestry Using Airborne Hyperspectral Imaging Sensor

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The airborne mission campaign was funded by Aeroscan Precision (M) Sdn Bhd Project Office and Universiti Putra Malaysia (sponsoring information)

Abstract

Universiti Putra Malaysia in collaboration with a private company, Aeroscan Precision (M) Sdn Bhd based in UPM Serdang conducted a research and commercial applications of airborne hyperspectral sensing data in precision forestry. The UPM-APSB's AISA sensor was flown over a representative series of forested areas in Peninsular Malaysia from 12-20th July 2004. It is a pushbroom imaging spectrometer recording remote sensing images over a large spectrum of wavelengths from the visible (400 nm) to near infrared (1 000 nm). Images have a ground pixel size of 1m by 1 m at a flight altitude of 1 000 m a.s.l and a constant flight speed of 120 knots. The ground validation segment of the projects was focused around hill/montane dipterocarps,. In order to characterize the properties and status of the forests, a number of images and field spectrum were developed. Prior to and after the flight, field spectral reflectance measurements using a handheld FieldSpec spectroradiometer were taken over the timber species of interest. The use of spectral unmixing methods for the discrimination of individual timber species image components leading to a more accurate identification of timber species, timber inventory and volume estimates were evaluated. The results imply that UPM-APSB airborne hyperspectral imaging technology would enable the development of a rapid forest resources assessment, especially in the sustainable forest management in Malaysia and other tropical countries.

Keywords: Airborne, Hyperspectral imaging, Precision forestry, Sustainable management

1. Introduction

Airborne hyperspectral imagers are powerful diagnostic tools for remote sensing. They will play an increasing worldwide role in forestry and other disciplines (Held and Hendrawan, 1996). A targeted imager will view smaller specific "target" areas selected by the user. A targeted hyperspectral imager operating from a low altitude flying aircraft will typically provide spatial and spectral resolutions in the order of 0.5-4 m and 5-20 nm, narrow, contiguous over the visible (VIS)-and-near-infrared, respectively.

An airborne hyperspectral imager's ability to detect molecular absorption and particle scattering "signatures" of constituents is its defining advantage, compared with broadband multi spectral imaging sensors such as the Landsat TM, SPOT HRV, IKONOS and QuickBird. The finer spectral resolution of an airborne hyperspectral imager, along with an appropriately high signal-to-noise ratio (SNR) in the wavelength of interest, allows detection and inferences of biological processes that are characterized by specific emission or absorption features. In many situations, an airborne hyperspectral imager can unambiguously identify surface constituents and their abundance, captures the unique spectra, or spectral signature of an object, which can then be used to identify and quantify the material(s) of which it is composed (Kamaruzaman, 2006a and 2006b). For vegetated targets, it has the effect of measuring the status of the targets such as within field variation and timber inventory for precision agriculture and forestry, respectively (Kamaruzaman and Dahlan, 2006a, 2006b and 2006c).

This paper therefore concerns a push-broom airborne hyperspectral imaging sensor technology for precision forestry applications using reflected solar radiation (400-900 um wavelength range) developed and operationalized by Aeroscan Precision (M) Sdn Bhd in collaboration with UPM's Forest Geospatial Information and Survey Laboratory (FGISL), Universiti Putra Malaysia, Serdang.

2. Materials and methods

2.1 Description and mounting of UPM-APSB's AISA Airborne Hyperspectral Imager

UPM-APSB's AISA is a state-of-the-art fixed wing aircraft mounted commercial hyperspectral imager operationalized by Aeroscan Precision (M) Sdn Bhd. project office in Lebuh Silikon, in collaboration with Forest Geospatial Information & Survey Lab (FGISL) of Universiti Putra Malaysia (Figure 1). It is designed to provide real time, frequent, repetitive, accurate and reliable pushbroom instrument that acquire images in 288 registered, contiguous narrow spectral bandpasses such that for each element it is possible to derive a complete reflectance spectrum. UPM-APSB hyperspectral imager is a complete system that consists of a compact hyperspectral imager head, miniature GPS/INS sensor for precise positioning, data acquisition unit and Caligeo post-processing software. This small portable instrument, with a total weight of only 15 kg was mounted on an aluminium metal plate that is compatible with a standard aerial camera mount, available in any fixed wing aircraft such as that of a Pan Malaysia Air Transport (PMAT) Short SkyVan SC7 and/or a Sabah Air GAF Nomad N22B. Swath width is 360 pixels and field of view (FOV) in cross track direction 20° which makes ground resolution from 1 km altitude approximately 1 m at a flight speed of 120 knots (60 m/s). In addition, 20 pixels per swath for downwelling irradiance were acquired via a fiber optic irradiance sensor (FODIS) on the N22B aircraft. Accurate position information, necessary for image rectification is measured with Systron C-MIGITS II integrated GPS/INS unit, which includes 3-axial inertial measurement unit based on solid-state gyros, GPS receiver and real time Kalman filter. The effect of the aircraft such as the lateral roll is monitored using data from an onboard gyroscope. The advantage of UPM-APSB airborne hyperspectral imager over other airborne hyperspectral instruments is the flexibility in selecting the sensor's spatial and spectral resolution characteristics. Reflected light from the target below the aircraft is transmitted through a sensor lens and directed to a prism-grating-prism optical system, which splits the light into its component wavelength spectra. The refractive properties of the two opposing prisms allow for a linear projection of light onto the CCD two-dimensional array.

<<FIGURE 1>>

2.2 Airborne data acquisition

The calibration flight by UPM's FGISL/APSB took place from 18-19 February 2004 in Selangor using a SC-7 aircraft. The commercial operating flights on the GAF Nomad N22B twin engine aircraft for precision forestry data applications were successfully conducted from 12-20th. July 2006. The airborne data presented here were only part of that collected by the overall project. Over 20 flight lines were collected for the project corresponding to better than 90% of the proposed data acquisition (by area). Some data were not acquired because of the weather and other operational considerations. Data delivery over some selected Area of Interest (AOI's) was accomplished within 24 hours of completion of data acquisition.

2.3 AISA data processing, field verifications and timber volume estimation

UPM-APSB's imaging spectrometer was configured to measure 20 spectral bands. The data was first pre-processed by a Caligeo software (a plug-in of the latest ENvironment for Visualizing Images [ENVI]) version 4.0 for a calibrated, rectified and georeferenced image after performing the atmospheric, radiometric and geometric corrections. After pre-processing, the data were processed digitally using ENVI version 4.0 which is a user-friendly to search for image spectral signatures, in addition to the geospatial capabilities, which result in an effective identification, visualization, spatial and spectral data reduction and management tool with integrated decision-making capabilities. The data were subjected to a minimum noise fraction transform, pixel purity index, n-dimension visualizer, identification, spectral angle mapper and mixture tuned matched filtering processing before getting the final output, AeroMapTM. AeroMapTM's turn-around time is typically 24 hours from collected data to visualized information since a short-turn around time is required to keep data processing costs down and to achieve the maximum end user satisfaction.

After the individual tree species were identified, classified and mapped, the timber volume was estimated using the volume formulae and regression model shown in Figure 2.

<FIGURE 2 >>

3. Results and discussion

Figure 3 demonstrated the usefulness of UPM-APSB's hyperspectral imager for precision forestry applications especially, individual tree species identification, classification and mapping that has benefited from the operationalization of UPM-APSB's AISA airborne hyperspectral data.

<<FIGURE 3>>

Airborne hyperspectral sensing has the potential to be useful for minimal impact harvesting since individual commercial tree species can be easily identified and mapped due to the different spectral "signatures" registered by different species (Figure 4). Upon combining this data with ground derived models that relate crown size to tree size (allometric relationships) for the various species in the project area, it was possible to derive the individual tree counts, average tree size by diameter size class, tree height and hence timber volume per hectare (Figures 5a and 5b). Using airborne

hyperspectral imagery seems to improve timber inventory assessments, allowing for increased accuracy and detail of individual species for sustainable development and forest statistics. Measurements of forest chemistry in relation to tree health and stress is most likely possible due to spectral sample regions of healthy versus stressed timber trees was easily separated at the NIR spectrum using canopy reflectance.

<<FIGURE 4>>

<<FIGURE 5a>>

<<FIGURE 5b>>

Malaysia has both international and national reasons to assess the sustainable development of the nation's forests. Monitoring our forests is essential to preserving Malaysia's second largest revenue-generated industry next to petroleum. Also, there is a need to assess Malaysia's progress on international commitments eg. Kyoto Protocol products (forest inventory, afforestation, deforestation and reforestation) where their parameters can be derived from AeroMap™ products measured by UPM-APSB's AISA airborne hyperspectral imaging. The temporal components of these measures provide trends for policy makers and for sustainable forest managers.

4. Conclusion

UPM-APSB now has experience and expertise in the disciplines and technologies required for airborne hyperspectral imaging earth observation especially in precision forestry applications. Our data and product, AeroMap™ have demonstrated many useful precision forestry applications which can provide a potential and efficient means of managing and sustaining agriculture and forestry development with precise geospatial details of the targeted sites and very minimal ground control points (GCP's).

Geo-corrected data can be provided to clients/users as apparent reflectance to thematic base map and features extraction AeroMap™ products. Data, products and services from UPM-APSB's hyperspectral imager have being available for commercial purchase from Aeroscan Precision (M) Sdn Bhd. Project Office/FGISL at Lebu Silikon, UPM. Utilizing this potential, Aeroscan Precision (M) Sdn Bhd/FGISL, UPM will organize similar missions upon request to interested clients in precision forestry and other applications. The current challenge is to intensify precision forestry applications development and to broaden the usage of airborne hyperspectral imaging data in Malaysia for forest inventory and reduced impact harvesting. This will entail the coordinated effort and support of the Malaysian government, industry and academia.

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Figure 1. The airborne hyperspectral imaging system kit and mounting of sensor on-board a fixed wing aircraft

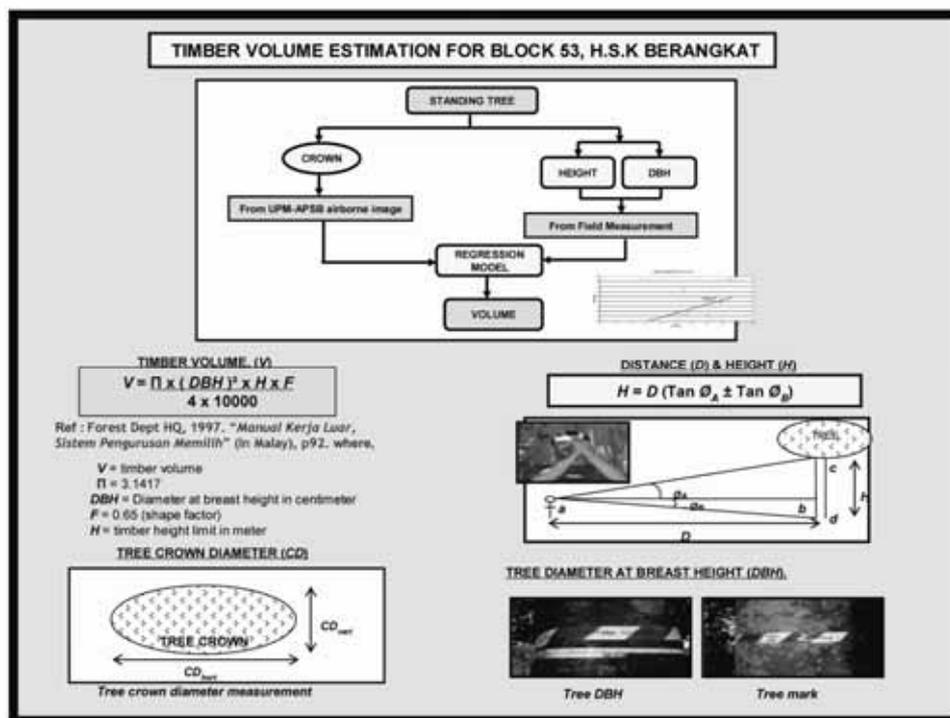


Figure 2. Estimating timber volume in Block 53 using the timber volume formulae and the derived regression model

Individual Species Information of Block 53, Beranghat P.F.R			
No.	Type of Species	No of Tree Counts	Timber Volume, m ³
1	Mahang	3	2.34
2	Berangan	14	136.92
3	Perah	25	51.75
4	Tualang	43	73.53
5	Nyatoh	74	99.90
6	Mersawa Durian	152	101.08
7	Kulim	183	206.79
8	Merawan Siput Jantan	237	457.41
9	Keledang	293	5.86
10	Medang Daun Besar	337	33.70
11	Ara Bertih	355	170.40
12	Meranti Tembaga	481	1,382.39
13	Minyak Beruk	315	3,244.50
14	Medang	234	348.66
15	Rengas	504	549.36
16	Keledang Bangkok	867	589.56
17	Kelat	1,768	2,422.16
18	Kedondong	1,485	1,257.79
19	Jelutong	943	3,423.88
20	Penarahan	596	426.14
21	Ara	330	83.49
22	Nyatoh Taban	2	4.28

Figure 5a. Mapping of individual timber species with different unique spectral libraries for precision forest inventory

Timber Distribution by Size Class (dbh)					
Species	DBH < 55cm	DBH = 55cm	55cm < DBH < 60cm	DBH > 60cm	Total No. of Tree Counts
Mahang	3	0	0	0	3
Berangan	0	0	4	0	14
Perah	12	0	12	0	25
Tualang	43	0	0	0	43
Nyatoh	74	0	0	0	74
Mersawa Durian	152	0	0	0	152
Kulim	183	0	0	0	183
Merawan Siput Jantan	162	0	75	0	237
Keledang	293	0	0	0	293
Medang Daun Besar	337	0	0	0	337
Ara Bertih	355	0	0	0	355
Meranti Tembaga	244	0	203	34	481
Minyak Beruk	257	0	0	48	315
Medang	234	0	2	0	234
Rengas	487	0	17	0	504
Keledang Bangkok	867	0	0	0	867
Kelat	1752	0	0	16	1768
Kedondong	1328	0	155	0	1485
Jelutong	902	0	0	41	943
Penarahan	559	0	27	0	596
Ara	183	0	227	0	330
Nyatoh Taban	2	0	0	0	2
Total	6355	0	779	147	9241

Figure 5b. Mapping of individual timber species with different unique spectral libraries for precision forest inventory. Managing Mountain Forest Resources of Gunung Stong, Kelantan using UPM-APSB's Airborne Hyperspectral Imaging. Paper Presented at the 3rd. *Bangi World Conference on Environmental Management*, 05-06 September 2006, Equatorial Hotel, Bangi, Selangor. 10p.



Effects of Soil Texture and Water Retaining Agent on the Emergence of Processing Tomatoes

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Abstract

In the present paper, effects of two factors including soil texture(sand content rate) and water retaining agent on the emergence ratio of processing tomatoes were investigated through general regression of agricultural design testing and data-processing system (DPS),with attempts to obtain the best agronomic measures according to the model The linear relationship between design factors and target values (Emergence rate) of the test model and effects of one degree item, quadratic item and interaction item were also observed in the present paper. Result showed the optimal intervals of X_1 and X_2 ranged from 0.83251~1.167949(g/100g) and 15.1765~34.8235%, respectively.

Keywords: Processing tomato, Soil texture, Water retaining agent, Emergence, Regression design and analysis

1. Introduction

Currently, processing tomato has been the leading industry and biggest export-oriented enterprise. Planting and processing of tomatoes has been the significant economic pillar for Xinjiang people of all nationalities to increase yield and benefit and wealth accumulation in rural areas(He, 2008, PP. 42~44). In order to solve the problem of supplying in balance and obtain high quality and high yield, planting patterns of processing tomato seedling transplantation have been extensively spread in recent years, especially plug-seedling in Xinjiang which resulted in extensive popularization and good results(Wang, 2007, P. 17). However, in actual practice, observation of emergence rate and its resultant control technique, especially effects of soil texture in combination with water retaining agent on the emergence of processing tomatoes have been less documented, and we thus know less about technological measures of increasing emergence rate of processing tomatoes. Therefore, in the present paper, we investigated effects of two factors including soil texture (sand content rate) and water retaining agent on the emergence ratio of processing tomatoes in order to obtain the best agronomic measures and look forward to provide scientific theoretical basis for producing(Zhi, 2003, PP. 360~361).

2. Materials and methods

2.1 Materials

The present study was undertaken in the NO.5 greenhouse of Vegetable institute of Shihezi in March, 2008, seeded on 1st March. The variety used was "rieger 87-5", a early maturing and main variety in Shihezi with 1000-grain weight of 3.0g and germination of 98%. Shufeng water-saving agent, loam soil and silt soil(diameter: 0.0625~0.0039mm) were applied in the present study. Basic nutritional constituent of loam was 3.6% organic matter, 0.18% total nitrogen, 0.27% total phosphorus, 6.5×10^{-6} available potassium and 2.2×10^{-5} available phosphorus (Fan, 2008, PP. 199~201, Jie, 2000, PP. 22~24). Tomatoes were seeded by the 72 plugs of polystyrene (50cm×30cm×30cm). 72 seeds were selected for treatment of each group with a thermometer, and 1 seed per plug.

2.2 Methods

According to the preliminary tests and empirical data, upper limit(+R) of water retaining agent consumption(per 100g soil) X_1 was valued as 2g/100g while lower limit(-R) as 0; loam soil and silt soil(diameter: 0.0625~0.0039mm) were

prepared by different volume ratios (sand content rate) of percentage X_2 with the upper limit (+R) of 50% and lower limit (-R) of 0 in the present two factors model. The consumption used for study was prepared according to the data listed in Table 1. 13 tests were performed to study the effects of two factors on emergence rate (%), namely variable Y by the quadratic general rotary unitized design with the aid of DPS v3.01. Processing flow was $X_1 \rightarrow X_2$. Maximum and minimum temperatures of greenhouse were recorded during the period of trial.

3. Results and analysis

3.1 Observation of experimental conditions

Air temperature of the greenhouse was observed during the period of trial, used as references for administration and emergence. Average lowest and highest temperature was 18.8°C and 35.7°C, respectively, during the period from 1st March to 17th March while maximum and minimum temperature was 42.0°C and 12.0°C, respectively. During the period, the hotter days were five in the early stage lasting shortly and thus had less interference with emergence.

3.2 Regression relation of water retaining agent, soil texture and seedling number

As seen from the emergence of each test, along with the level values of -R → +R, emergence rate decreased gradually. Y was obtained through seedling numbers, used as target values. Binary quadratic regression relation of two factors and target value was observed. Resultants Y (Table 2, 3) were input into programme, and through calculating, the following regression equation was obtained:

$$Y = 42.50000 - 2.76777X_1 + 0.52678X_2 + 2.01134X_1^2 + 12.76250X_2^2 - 1.05000X_1X_2 \quad (1)$$

Variance analysis and F value testing was undertaken to investigate the fitting degree and reliability of equation (1). As seen from Table 3, due to $F_1 = 1.880 < 3.97$ (Critical value of $F_{0.05}$), lack of fit term was not significant, and thus we could taken further statistics analysis and test the quadratic regression model; due to $F_2 = 4.036 > 3.97$, quadratic regression equation was significant at the level of 0.05 which indicated that experimental data was in line with the applied quadratic mathematic model, and quadratic regression equation fit actual situation closely and could be used as references for forecasting. At the level of 0.05, P values of X_1 , X_2 and X_2^2 term were all lower than 0.05 which indicated that effects of one degree term of the three factors level on the emergence rate of rieger 87-5 were significant; P values of X_2 and X_1X_2 term were higher than 0.05 which indicated that effects of these two terms on the emergence rate were not significant and could be eliminated for no references.

At the significant level of $\alpha = 0.10$ after eliminating the insignificant term, regression equation was briefed as follows:

$$Y = 42.50000 - 2.76777X_1 + 2.01134X_1^2 + 12.76250X_2^2 \quad (2)$$

Mathematical model equation (2) provide a information base which could be used as a reference to analysis effects of one degree term and square term and obtain the best agronomic measures for produce.

3.3 Effects analysis of each term according to the target value of emergence rate

According to equation (2), partial regression equation of one degree and square term X_i ($i=1,2$) against Y_{x_i} was obtained as follows:

$$\hat{Y}_{X_1} = 42.50000 - 2.76777X_1 + 2.01134X_1^2 \quad (3)$$

$$\hat{Y}_{X_2} = 42.50000 + 12.76250X_2^2 \quad (4)$$

Through derivation of equation (3) and (4), we have

$$\hat{Y}_{X_1}' = -2.76777 + 4.02268X_1 \quad (5)$$

$$\hat{Y}_{X_2}' = 25.5250X_2 \quad (6)$$

Different level values were obtained through solving equation, and compared to value of zero level, within the range of $-R \leq X_i \leq +R$, level value X_1 which showed effects on emergence was 0.55 at the range of 0 ~ +1 while X_2 was 0 at the range of -1 ~ +1. Therefore, we could speculate that water retaining agent had the greater effects on emergence rate of processing tomatoes and soil texture followed.

3.4 Optimization of agronomic measures

Maximum of Y attained 91.67% in the tests. Taken emergence rate of 56.54% as target value, frequency analysis were undertaken through the designed mathematical model (Table 4), and corresponding factor values of two factors in 95% confidence interval were obtained, which were qualified for our interval estimate values of optimized agronomic measures (Table 5).

$$X_1 = 0.83251 \sim 1.167949 \text{g}/100\text{g}$$

$$X_2 = 15.1765 \sim 34.8235\%$$

4. Conclusions

Effects of two factors, namely water retaining agent consumption X_1 (X_1 g/100g) and soil texture X_2 (sand content rate, %) on emergence rate Y (%) were investigated in the present paper. According to the quadratic general rotary unitized design and analysis, results showed that there was a linear relationship between Y and two factors X_i ($i=1, 2$) and past through F_1 and F_2 value test. Through effect analysis of one degree and square term of equation, results showed that level value X_1 which showed effects on emergence was 0.55 at the range of 0~+1 while X_2 was 0 at the range of -1~+1 compared to value of zero level. Taken average emergence rate of 56.54% as ideal target value in each test, the optimal intervals of X_1 and X_2 ranged from 0.83251~1.167949(g/100g) and 15.1765~34.8235%, respectively. In the trial, we also could speculate that if water retaining agent consumption increased along with the increasing of soil content rate within the designed interval range, high emergence rate was obtained.

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Table 1. Values and levels of the independent variables ($R=1.41$)

independent variables	levels				
	-R	-1	0	+1	+R
X_1 g/100g	0	0.29	1	1.71	2
(x_2 ,%)	0	7.3	25.0	42.7	50.0

Table 2. Experimental design and results

Experiment NO.	X_1	X_2	Seedling number
1	1	1	41
2	1	-1	43
3	-1	1	66
4	-1	-1	63
5	-R	0	47
6	R	0	32
7	0	-R	44
8	0	R	45
9	0	0	38
10	0	0	29
11	0	0	35
12	0	0	28
13	0	0	20

Table 3. Variance analysis of the test results

Source of variance	Sum of squares	Degree of freedom	Mean square	F ratio	P value
X ₁	75.5124	1	75.5124	11.6599	0.0112
X ₂	110.1997	1	110.1997	0.0160	0.9030
X ₁ ²	17.8574	1	17.8574	8.4204	0.0229
X ₂ ²	140.8696	1	140.8696	14.4316	0.0067
X ₁ X ₂	6.1951	1	2.1951	0.0438	0.8402
Regression	361.2026	5	167.5777	F ₂ =4.036	0.0686
Residual	52.4888	7	46.9384		
Lack of fit	17.3528	3	55.268	F ₁ =1.880	0.1664
Pure error	32.3840	4	28.552		
Total	423.4902	12			

Table 4. Each factor group of top value

Frequency distribution of variances in 18 tests with target value over 56.54%				
Level	X ₁	Frequency	X ₂	Frequency
-1.4142	3	0.2278	3	0.2778
-1.0000	3	0.2278	3	0.1667
0.0000	2	0.1111	1	0.1111
1.0000	2	0.1111	1	0.2667
1.4142	1	0.2222	2	0.2778

Table 5. Frequency of of variances in 18 tests with target value over 56.54%

	Weighted mean square	Standard error	95% confidence interval
x ₁	-0.2450	0.2720	-0.778... 0.287
x ₂	0.1000	0.2630	-0.555...0.555

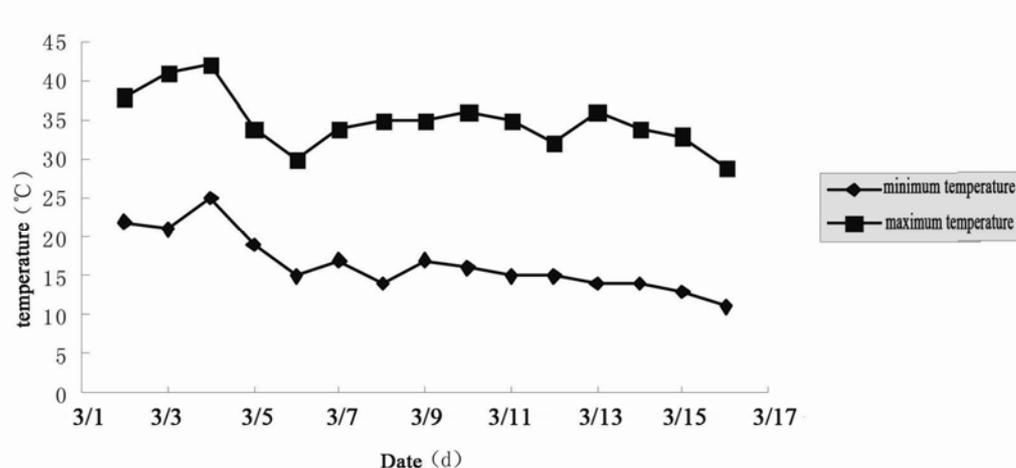


Figure 1. Changing curves of temperature during the period of trial



Effect of Densities of Planting on Yield and Essential Oil Components of Fennel (*Foeniculum vulgare Mill Var. Soroksary*)

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Abstract

In order to study the effect of different densities of planting on yield and essential oil components of Fennel (*Foeniculum vulgare Mill Var. Soroksary*), an experiment was carried out in college of agriculture Karaj at 2008. Experiment was conducted based on completely randomized block design with three replication and five plant densities. Five plants spaces were 10, 15, 20, 25 and 30cm on the row. The distance between rows in all treatments was 40cm. The essential oil extracted by water distilled method from seeds and essential oil was analyzed by gas chromatography (GC). The higher essential oil percentage (% 3.53) was obtained with the lowest densities of planting. The higher percentage of anethole (%83.07), estragol (%3.47), fenchone (%8.04), p-cymene (%4.45), α -terpinene (%0.54), sabinene (%0.51), and α -Pinene (%0.48) were obtained with space between plants 25, 10, 20, 20, 15, 20, and 25cm, respectively.

Keywords: Fennel, Density, Essential oil, Anethole

1. Introduction

In the world today, the traditional food, forage and fiber crops are not the only plants of key agricultural and trade significance, but they also include plants whose secondary metabolites are valued for their characteristic aromatic or therapeutic attributes, or as main natural inputs to the proliferating perfumery and chemical industries. Bitter fennel (*Foeniculum vulgare Mill. Var. soroksary*) is a perennial hemicryptophyte, common in the Mediterranean basin and known since antiquity as a medicinal and aromatic herb. The fruit of bitter fennel is commonly used as a natural remedy against digestive disorder. Bitter fennel is also used to flavor foods, liqueurs and in the perfumery industry (Lucinewton et al., 2005). Present world market is around US\$ 80 million. Iran exports these produce worth US\$ 10 million (Masood et al., 2004). The production of essential oils not only depends upon the metabolic state and present developmental differentiation program of the synthesizing tissue, but also is highly integrated with the physiology of the whole plant. Besides, the oil productivity is friendly to ecophysiological, environmental and other factors (Voirin et al, 1990). Plant spacing is an important factor in determining the microenvironment in the fennel field. The optimization of this factor

can lead to a higher yield in the crop by favorably affecting the absorption of nutrients and exposure of the plant to the light. According to results of Verzalova et al (1988) row spacing did not effect on the plant height but number of umbel and seed yield per plant was increased at the wider spacing. Masood et al (2004) investigated the effect of row spacing (40, 50, 60, and 70cm) on morphological characters and seed yield of fennel and reported that the greatest plant height, seed yield per umbel, and seed yield per hectare were obtained with the lowest row spacing but the lowest plant height, seed yield per umbel, and seed yield per hectare were obtained with the greatest row spacing.

Bianco and Damato (1994) reported that plant density not affected on plant height at flowering of primary umbel, number of stem and umbel per plant, yield per plant and per hectare. Akbariani et al (2006) studied the effect of plant density on seed yield of Coriander (*Coriandrum sativum*) and showed with increasing of plant density seed yield decreased significantly. Hasanali et al (2002) with study the effect of different plant densities on yield dry material of Thyme (*Thymus vulgaris*) showed that the higher yield of dry material was obtained with 15 cm densities of planting.

Arabasi and Bayran (2004) with planting sweet basil in three plant density (20, 40 and 60 plants m⁻²) reported that the highest amount of dry matter, percentage and the yield of effective substances produced in 20 plant m⁻². Aflatuni (2005) mentioned that planting pattern had no significant effect on essential oil amount and composition of mint. In this study was considered the effect of different densities of planting on yield and essential oil components of Fennel.

2. Materials and methods

This experiment was carried out in the Tehran university, college of agriculture of Karaj in 2008 (Table1). Field was plowed during the fall season and was disked before sowing time to provide a proper seedbed. Experiment was conducted based on completely randomized block design with three replications and five plant densities. The experiment includes 3 blocks and each block is contained 5 plots. Each plot size was 2.5x1.5m. Distance between blocks and plots were 1m. Five plant spaces were 10, 15, 20, 25, and 30cm. The distance among rows in all treatments was 40cm. Each plot was consisted of five rows. The bitter fennel seeds were sown at the 7th March 2008. Irrigation were done as: 1. 2-3 days interval irrigation until germination stage, 2. 4-5 days interval irrigation from germination to appearance first flowers stage, and 3. 7 days interval irrigation from appearance first flowers to harvest stage. Thinning was done when plants had 4-5 leaves. In order to better growing of plants, crust breaking operation were done at three stages (18th April, 4th May, and 19th May). Ten plants were selected at random from each plot for recording individual plant observation. All agronomic practices were keeping normal and uniform for all the treatments. Then seeds harvested after ripening at two stages (20th August and 30th August) and dried in shade for 72 hour. In order to extraction of essential oil, 15gr of seeds powdered and then essential oil isolated by water distillation for 4h and with three replications. The essential oils were separated from the aqueous layer, dried over anhydrous sodium sulfate and calculated average of essential oil yield for three replications and finally were stored in the refrigerator until analysis time. In order to identify the essential oil components, gas chromatography was carried out using a GC Thermo-UFM with Ph-5 column (10m, 0.1mm ID, 0.4 FT). Oven temperature was performed as follow: 60°C to 285°C at 80°C/min; injector temperature 280°C; detector temperature, 280°C; carrier gas, He (0.5ml/min) with chrome-card software and area normalization method. In order to identify the essential oil components, relative retention time of components was compared together.

3. Results and discussion

Results indicated that essential oil percentage was affected significantly by different densities of planting, as the maximum essential oil percentage (%3.53) was obtained with the minimum plant density (space between plants on the 30cm row) and the minimum essential oil percentage (%3.1) was obtained with the maximum plant density (space between plants on the 10cm row)(see Table2). The higher percentage of anethole (%83.07), estragol (%3.47), fenchone (%8.04), p-cymene (%4.45), α -terpinene (%0.54), sabinene (%0.51), and α -pinene (%0.48) were obtained with space between plants on the 25, 10, 20, 20, 15, 20, and 25cm row, respectively. While the minimum percentage of anethole (%81.98), estragol (%3.39), fenchone (%6.97), p-cymene (%3.74), α -terpinene (%0.45), sabinene (%0.39), and α -pinene (%0.32) were obtained with space between plants on the 20, 15, 25, 10, 25, 10, and 10cm row, respectively (see Table3, and Fig 1 to 7). In general, with decreasing of space between plants on the row essential oil percentage was increased. Ahmad and Haque (1986) with study the effect of different plant densities on yield essential oil and oil of Nigella (*Nigella sativa*) showed that the higher yield of essential oil and oil were obtained with the lowest of plant density. Shalaby and Razin (1994) with study on the Thyme (*Thymus vulgaris*) reported that the maximum yield of dry matter and essential oil were obtained with the lowest of plant density. El-Gandi et al (2001) suggested that the highest of essential oil percentage per plant in Sweet basil (*Ocimum basilicum* L.) was obtained with the lowest of plant density but total yield of essential oil was increased with the increasing of plant density. Najafi and Moghadam (2002) reported that with increasing the plant density, seed and biological yield were increased. Wajid et al (2004) showed that plant population had significant effect on yield component. For production of essential oil, multiple factors are effective including: plant ontogeny, site of oil production, photosynthesis, photoperiodic modulation, light quality, seasonal and climatic variations, nutritional relationships, plant growth regulators, plant density, moisture, salinity, temperature. In

general, each factor that is influenced on the photosynthesis can be affected on the production of essential oil. Plant density by affecting the absorption of nutrients and exposure of the plant to the light can be affected on the photosynthesis and production of essential oil.

4. Conclusion

Fennel plant is one of the most interesting research plants. It is between medicinal and aromatic plant. Plant density is an important factor in determining of fennel yield. Results of this study showed that influence of different densities of planting on yield essential oil were significant at the 0.05 probability levels. But different densities of planting have different effect on the essential oil component.

Acknowledgement

The authors are grateful to University of Tehran, Mr Kaveh Mollazade, Rahmat Mohammadi, and Khaled Ahmad Aali for their valuable assistance and technical support.

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Table 1. Geographical coordinates, average annual rainfall and mean annual temperature of Karaj

longitude	latitude	height of sea level(m)	average annual rainfall(mm)	mean annual temperature(°c)
50° 59'E	35° 47'N	1312/5	230	14/3

Table 2. Relationship between space between plants and percentage of essential oil

Space between plants(cm)	10	15	20	25	30
Essential oil (%)	3.1	3.22	3.33	3.33	3.53

Table 3. Relationship between plant density and percentage of essential oil component

RT	RI	compound	10cm	15cm	20cm	25cm	30cm
1.42	954.03	α -Pinene	0.32	0.38	0.41	0.48	0.45
1.5	984.88	Sabinene	0.39	0.44	0.51	0.42	0.47
1.59	1039.64	α -Terpinene	0.51	0.54	0.5	0.45	0.51
1.62	1054	P-Cymene	3.74	3.95	4.45	4.06	3.8
1.76	1117.65	Fenchone	7.74	7.63	8.04	6.97	7.94
1.96	1223	Estragol	3.47	3.39	3.43	3.44	3.41
2.14	1321.05	E-Anethol	82.52	82.51	81.98	83.07	82.68

RT: Retention time

RI: Retention index

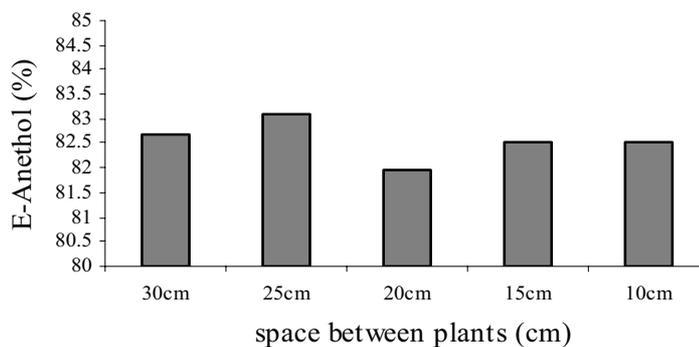


Figure 1. Relationship between percentage of E-Anethol and space between plants

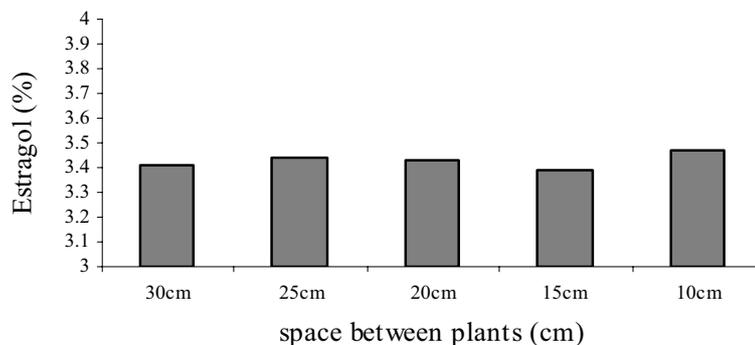


Figure 2. Relationship between percentage of Estragol and space between plants

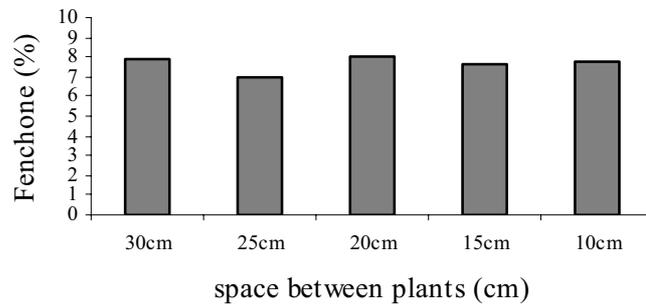


Figure 3. Relationship between percentage of Fenchone and space between plants

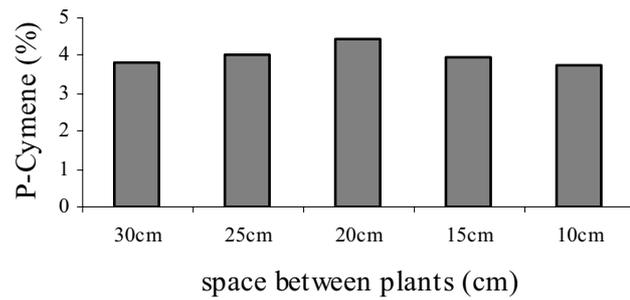


Figure 4. Relationship between percentage of P-Cymene and space between plants

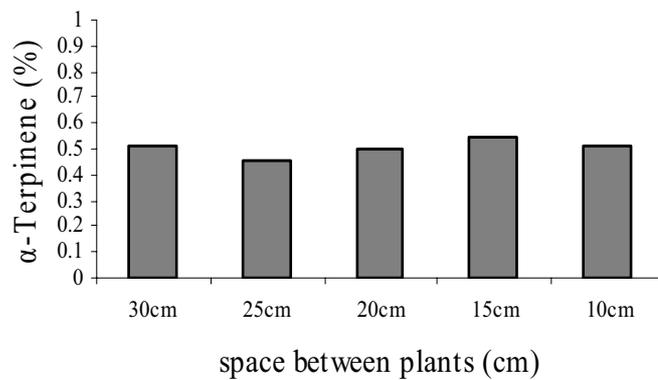


Figure 5. Relationship between percentage of α -Terpinene and space between plants

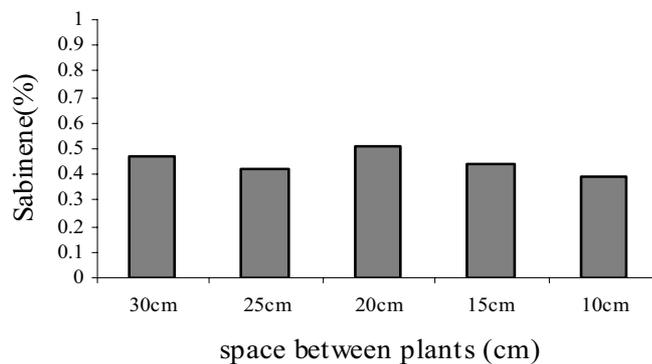


Figure 6. Relationship between percentage of Sabinene and space between plants

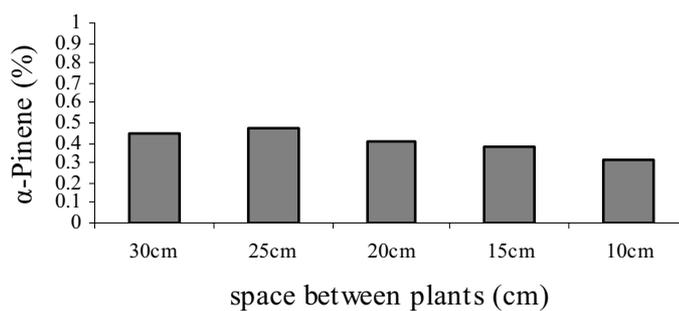


Figure 7. Relationship between percentage of α -Pinene and space between plants

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Journal of Agricultural Science

Semiannual

Publisher Canadian Center of Science and Education
Address 4915 Bathurst St. Unit # 209-309, Toronto, ON. M2R 1X9
Telephone 1-416-208-4027
Fax 1-416-208-4028
E-mail jas@ccsenet.org
Website www.ccsenet.org
Printer William Printing Inc.
Price CAD.\$ 20.00

