Fatty acids characters and grain yield of *Brassica napus* L. as affected by tillage systems and sowing date

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Abstract

In order to study the effect of ttillage systems and sowing dates on yield, yield components and oil content of rapeseed genotypes (*Brassica napus* L.), field experiment was carried out at the Sari Agricultural Experimental Station, 25 km of Eastern Sari, Iran in 2001–2002 and 2002–2003. The experimental design was split plot factorial with three replications. Three tillage system levels were allocated to main plots, and combination of rapeseed genotypes (Hyola 401, PF) and sowing dates (including 8 Sep, 23 and 7 Oct) were randomised to sub-plots. The tillage levels were consisting of: no- tillage, minimum tillage (residue return with two disks) and conventional tillage (residue return with plow and one disk). Results of ANOVA using SAS Procedures showed that tillage systems had significant differences at the 1% probability level on grain yield, conventional tillage produced more grain yield and there were no difference between minimum and no tillage. Combined analysis showed that tillage system had significant of brassica (P<0.05), and conventional tillage and no tillage with means of 42.68 % and 42.54 % oil content in comparison minimum tillage with 41.63% have advantage. Varieties were differences (p<0.01) for grain yield, oil content and oil yield. Result showed that sowing date had significant difference at the 5% probability level on oleic acid content, in which 23 and 7 Oct produced the most oleic acid content and 8 Sep (first sowing date) was located in the end rank. Overall the result of this research showed that different sowing date and agronomic techniques have different effects on saturated fatty acids, these parameters were no equal direct on fatty acids combination.

Key words: Tillage systems, Brassica napus L., Yield and Oil Content, Sowing date and Fatty acids

Introduction

Different problems such as uneconomical current tillage system and gradual erosion in these systems and on the other hand reduction of consume water and fertilizer rates in canola, cultivation of canola can apply in rotation for increasing organic matter of soil. These low rainfall environments are where average annual rainfall is less than 240 mm in Iran. In these areas with low rainfall, canola is often sown dry at a set date (early to mid May) rather than waiting for the germinating rains, to optimise yield and oil content. In oilseed rape, oil is deposited late during seed development. We observed 5-10% oil (when the seed size is about 0.5 mg), 200 degree days after pollination when profile consisted of 50-70% saturated fatty acids (C16:0, C18:0), and a greater content of C18:2 (15-27%) than C18:1 (6-12%). The fatty acid composition changed rapidly in subsequent days, and the proportions approached those observed in mature seed. At maturity, high temperature resulted in a significant increase of the C18:1 content and a decrease of the C18:3 content. This is consistent with the results of Lagravere et al. (2000) and Izquierdo et al. (2003) who showed that low temperature increased C18:1 and C18:2 desaturation, resulting in a higher C18:3 content of mature oilseed rape seeds. At low temperature and under conditions of water stress, the C18:1 content decreased and the C18:3 content increased. The highest seed oil and erucic acid contents were detected in the Norin 16 (B. napus) and 181026 (B. juncea) lines. However, line 68-5702 (B. carinata), in virtue of its high yielding capacity, produced a higher oil yield than all the other lines and an erucic acid yield second only to that produced by Norin 16. In oilseed rape, oil is deposited late during seed development. Seeds were sampled every 200 degree days after pollination. We observed 5-10% oil (when the seed size is about 0.5 mg), 200 degree days after pollination when profile consisted of 50-70% saturated fatty acids (C16:0, C18:0), and a greater content of C18:2 (15-27%) than C18:1 (6-12%). The fatty acid composition changed rapidly in subsequent days, and the proportions approached those observed in mature seed. At maturity, high temperature resulted in a significant increase of the C18:1 content and a decrease of the C18:3 content. In research effect of temperature (18/10°C and 26/18°C day/night) and water deficit (50%) was examined in winter rapeseed cv. Samourai from the end of the flowering period to maturity. Yield and the yield components, number of siliques/m² and seed number/m², significantly decreased at high temperature and reduced water. Single seed weight decreased at high temperature and also at low temperature under conditions of water stress. Nitrogen content increased and seed oil content decreased at elevated temperature and with water stress. However, for almost a decade attention was focused on double low (00) winter and spring varieties and only recently has interest been directed towards a number of brassica species characterized by a fatty acid composition different from those traditional cultivars previously mentioned. Conservations tillage practices are arable option for increasing nutrient use efficiency by crops, since these practice retain residues after the crop has harvested (Hoching, 2001; Ahmad et al., 2000). Conservation tillage practices are a viable option for increasing nutrients use efficiency by crops, since these practices retain residues after the crops have been harvested. Residues play a critical role in nutrient distribution and plant growth (Bayer, 2004) and affect the amount of soil nutrients available to the crops (Tobares et al. 2002; Tawainga et al. 2002). Residues allow N to be plant available for longer periods of time, by initially immobilizing, and then gradually mineralizing, the N (Pike D and Clarke 2004; Momoh et al., 2001). NT has more advantages on plant growth and yield in soil with low organic matter and poor structure than soil with high organic matter and good structure (Miralles et al. 2000). NT has not affect on seedling growth; however soya grain yield also has not been reduced (Dreccer, et al., 2000). These alterations for plant growth and yield production depended on soil texture and structure; climate factors such as precipitation rate and weed control. NT in comparison with CT have ordinary more loss water and low availability of elements, therefore efficiency of soil environment alteration in point of view mineralization nitrification and nitrogen fertilizer requirement at NT system is more than CT. The aim of this work is to obtain the first information on the productive and biological behaviour and on the variation in fatty acid composition in a number of varieties belonging to Brassica napus in an environment in Iran. The objective of these experiments is to evaluate of tillage Systems and sowing date on fatty acids characters and grain yield of Brassica napus. L.High variation was observed in canola varieties for fatty acid composition.

Materials and Methods:

In order to study the effect of tillage systems and sowing dates on yield and fatty acids composition of rapeseed genotypes (Brassica napus L.), field experiment was carried out at the Sari Agricultural Experimental Station, 25 km of Eastern Sari, Iran in 2001–2002 and 2002–2003. The experimental design was split plot factorial with three replications. Three tillage system levels were allocated to main plots, and combination of rapeseed genotypes (Hyola 401, PF) and sowing dates (including 8, 23 Sep and 7 Oct) were randomised to sub-plots. The tillage levels were consisting of: no- tillage, minimum tillage (residue return with two disks) and conventional tillage (residue return with plow and one disk). The distance between plots and plants on rows were 3 M and 5 cm respectively. Soil practice was performed at early September. Plots received enough fertilizer and application of 2.5litter/ha trephlan before planting for weed control. Fertilizer was applied in basis of soil analysis (all plots and phosphors fertilizer and one third nitrogen fertilizer requirement in basis of canola production directions in country different climate at planting stage). The maximum fertilizer requirement was 50kg/ha net nitrogen, 59-kg/ha P2O2 and 100 kg/ha K2O.One third nitrogen fertilizer was applied at bolting time and final amount of fertilizer was used at the beginning of flowering. All protection stages were performed as traditional method. Plants were harvested when % 40- 50 of seed on main and branch pods became bright brown or black. Grain weight and fatty acids content were measured on all treatments. Grain yield was determined on middle row of plot after omitting the margin effect (half meter from beginning and end of each row). In each treatment, seeds were harvested (3 replicates) every 200 degree days until maturity. Oil content was determined by Nuclear Magnetic Resonance. Fatty acid composition was analysed using gas chromatography of methyl esters. Each fatty acid was expressed as a percent of the total fatty acids. At harvest, yield were determined for the three replicates (0.5 m²) per treatment The data were analysed using the general linear model (GLM) procedure of the statistical analysis system, SAS (SAS Inst., Cary, Nc) to perform analysis of variance and selected correlation when analysis of variance showed significant treatment effects, Duncan's multiple range test was applied to compare the means at p = 0.05. Bartlet test determined the homogeneity of variance in all traits. Mix analysis was applied to the rest of trait.

Result and Discussion

Oil yield and percent were influenced by tillage system, sowing date and variety (p<0.01) (Table 5). So that conventional tillage system and no tillage system had more oil than minimum tillage system (Table 6). This differences probability rose from moisture alteration and nitrogen rate presence in soil. Tawainga et al. 2002 reported that tillage practices did not affect oil percent. Hyola variety produced less oil yield than PF variety. Grain yield and oil percent were more in 8 Sep sowing date s than 7 Oct sowing date (Table 8). Oil yield also was significantly affected by interaction among variety, sowing date and tillage system (p<0.01) PF variety with conventional tillage system and 8 Sep sowing date had the most oil yield (Table 9). Oil percent is affected by genetic factors (Izquierdo et al. 2003). If the plant is not under drought stress at late growth stage seed oil percent will be content in each variety. Bell C and Van Rees H 2003 showed that environmental factors in addition to genetic factors have affected on oil percent. Plant population density has addition effect on oil percent (Hernan, 2000). Other researchers showed that plant population density reduced oil percent (Herridge et al. 1992). At least it has known that oil percent heredity is higher than grain yield general result. Result showed that in spit of more grain yield in conventional tillage system, varieties can use in no or minimum tillage system with 8 cm sowing date for obtaining sustainable agriculture and utilization of soil sources science Hyola variety is early maturity, it can use in climate zone with lower growth period. In general, result in this experiment showed that minimum and no tillage system have many benefits in comparison with conventional tillage system. Environmental factors such as radiation moisture and temperature have less effect on it. Final seed size has very variation among genotypes under different environmental alteration (Cheema et al., 2001). Big seed productions that fill completely not only increase grain yield, but also produce suitable seed for cultivation. Tillage system, sowing date and variety have significantly effect on grain yield (Table 2). Minimum tillage system had lower grain yield than normal and no tillage system. There were not significantly differences between normal and no tillage system for grain yield (Table 6). Hyola variety had lower grain yield than PF variety (Table 7). Grain yield is resulted of plant canopy activity during a growth season and utilization manner of radiation and other environmental sources. In this case, leaf photosynthetic activity has been affected by radiation, its distribution and respiration (Hernan et al. 2000). Varieties were different in former mentioned characteristic varieties that germinate early, growth rapidly in winter and produce leaf area in early rapid growth, can utilize more radiation and increase radiation efficiency in leaves and as a result produce more yields. Since PF variety has more rapid

growth than Hyola variety therefore it has the most grain yield, since it utilize from environmental source as suitable manner in comparison with Hyola variety. This result is in agreement with other studies. 16 cm sowing date had lower grain yield than 8 Sep sowing date (Table 8). There is significant difference for interaction effects among variety, tillage system and sowing date (p<0.01) (Table 9). So that PF variety with 8 Sep sowing date and conventional tillage system had more grain yield than other treatment. Hyola variety in 16 Sep sowing date and minimum tillage system had the lowest grain yield (Table 9). There is relation among yield component, so that increasing one component decreases the other components. Sowing date may have a major effect on rapeseed oil characteristics during grain filling. Researcher reported a significant correlation between oleic acid content and sowing date between 21 and 70 days after flowering. Also 'in vivo' experiments with developing rapeseed seeds demonstrated that oleate desaturase activity was stimulated by low sowing date, repressed by high sowing date and rapidly restored by bringing the seeds back to low sowing date. This was due to the synthesis or activation of oleate desaturase at low sowing date and the reversible inhibition of this enzyme at high sowing date. In our experimental conditions, the prolongation of the grain filling phase in a cooler period observed in the irrigated treatment and the anticipation of the sowing date, resulted in lower mean sowing date during grain filling in both years. However, it is known that in high oleic hybrids $\Delta 12$ oleate desaturase is active only at the early days of the embryo development associated with synthesis activity of the lipids and that its transcript is not accumulated during the grain filling period (Lagravère et al., 2000). So while the early sowing date may have taken advantage of the lower sowing date of the early phase of seed development, the lower sowing date recorded at the last phase of grain filling in the irrigated treatment may not be responsible for the lower oleic/linoleic acid ratio observed. On the other hand, irrigation may influence the sowing date of the vegetative apparatus and the microclimate of a crop; so, we think that in our experimental conditions, lowering the plant tissue sowing date upon irrigation, might have caused a higher activity of oleate desaturase and a lower oleic/ linoleic acid ratio. The small differences observed between treatments are in accordance with Izquierdo, 2003 who reported that in high oleic acid mutants, the oleic and linoleic contents are less influenced by sowing date than in standard genotypes (Flagella et al., 2000). Anyway, other environmental factors also, such as the unfavorable cell water and probably nutritional status under non-irrigated conditions, may influence the genetically programmed activation or synthesis of oleate desaturase. Besides, the synthesis of fatty acids in seeds takes place up to 18:1 in proplastids and from 18:1 to 18:2 and 18:3 in the cytosol (Hakan Ozer. 2003). So, environmental factors may influence the proportions of fatty acids not only by altering the enzyme activity but also influencing the transport from an organelle to another (Steer and Seiler, 1990). In conclusion, in a Mediterranean environment, supplementary irrigation and early sowing resulted in a notable rise in seed yield and in a decrease in the oleic/linoleic ratio in high oleic rapeseed genotypes. A possible role of lower tissue sowing date during grain filling under irrigation and early sowing in activating oleate desaturase might be hypothesized. Further investigation is needed to get deeper insight into the effects on seed oil composition of the interaction between irrigation and sowing date and rapeseed crop sowing date.

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K (p pm)	Р(p pm)		Total N %	,)	EC (ds/m	12)	Р	H	Org	anic carbo	n (%)	Soil texture
400		31		0/05-0/06		0/6		7,	7		0/63		Silty clay loam
ATTRIBE	JAN	FEB	MAR	APR	MAY	JUNE	JULY	AUG	SEP	OCT	NOV	DEC	ANNUAL
EMPERATURE (C)	7	7.3	9.5	14.9	19.5	23.5	25.5	25.7	22.8	18.1	13	8.6	16.3
HUMIDITY %	84	83	84	81	78	79	81	82	83	84	85	86	82
PRECIPITATION	66.2	62.6	63.6	36.4	31.2	31.1	31.4	48.2	80.4	103.6	99.9	77	731.6
WIND	2.9	3	3.3	3.4	3.1	2.7	2.4	2.4	2.3	2.4	2.4	2.5	2.8
DEGREE DAYS	135.2	120.7	125.1	161	205.7	215.8	210.8	171.5	157.7	170.3	145.4	130.4	1949.6
SUNSHINE	135.2	120.7	125.1	161	205.7	215.8	210.8	171.5	157.7	170.3	145.4	130.4	1949.6

Table1: the results of soil analysis and climatology data means in Bay cola

Table2: Combined analysis of variance of agronomic practice in tillage systems and sowing date

Mean of square							
C18:0	C16:0	Oil yield	Oil content	Grain yield	1000-Grain weight	D.F.	S.O.V
0/0009	0/0093	3257/025	1/0376	2613/76	0/0533	2	Repetition(R)
0/0001	0/0001	347754/9	32/9	3046176/3	0/1025	1	Year
0/0005	0/03	38841/46	22/11	25652/2	0/0258	4	Y(R)
0/0034	0/0045	**208461/24	**11/66	**895488/1	**0/6738	2	Tilage systems
0/0042	0/0069	**469757/18	**4/21	**2619969/3	**0/534	2	Y* T
0/0031	0/0048	13856/17	2/18	52468/1	0/0484	8	R*T(Y)
0/0007	0/0045	**425301/54	**9/35	**1972602/9	*1/88	2	Sowind date (D)
0/0006	*0/030	**380770/69	**11/4	**1658971/6	**16/396	1	Variety
0/0012	0/0052	**72818/2	**10/778	**588727/2	**0/636	2	Y*D
0/0024	0/00014	**32398/79	**11/34	**433174/7	**1/125	1	Y*V
0/0017	0/00078	**133760/95	**2/56	**614830	**0/312	4	T*D
0/0038	0/01027	14389/5	1/73	86015/9	0/1397	2	T*V
0/0006	0/0052	**25887/8	0/1432	**199692/4	**1/66	2	D*V
0/0027	0/0012	*21747/6	**3/63	**213955/9	0/1347	4	Y*D*V

	Mean of square									
C18:0	C16:0	C16:0 Oil yield Oil content Grain yield 1000-Grain weight D.F. S.O.V								S.O.V
0/0008	0/000	9 **22	27902/88	**5/59	**1218904/8		0/1194	2		Y*T*V
0/0025	0/000	9 18	079/79	0/1831	82687/6		0/0182	2		Y*D*V
0/0015	0/0042	3 12	2214/4	0/8357	54211/1		0/1134	4		Y*T*D*V
0/0019	0/0034	4 **1	7646/24	0/653	**111537/9		0/1194	4		V*D*T
0/0023	0/0062	2 7	323/9	0/719	35772/5		0/112	60		Error
Ns, * and **	*: not signific	ant, significan	t at the 5 and 1 %	levels of probabili	ty, respectively.					
Table3: Combined analysis of variance for quality charactersin tillage systems and sowing date										
	Mean of square									
TU/TS	UAP	TUSFA	LNAP (C18: 3)) LAP (C18: 2)	18:1	16:1	TSFA	C20:0	df	S.O.V
0/0227	0/1022	0/0980	0/5998	0/150	0/0007	0/0146	0/0089	0/0001	2	Repetition(R)
0/0001	0/0198	0/0149	0/2205	0/024	0/00003	0/0004	0/0003	0/00001	1	Year
0/0786	0/0359	0/0043	0/0998	0/1908	0/00002	0/0043	0/0352	0/0007	4	Y)(R
0/0142	0/0148	0/0342	0/0449	0/0605	0/00003	0/0057	0/0042	0/0017	2	Tilage systems
0/01405	0/0212	0/0193	0/2046	*0/744	0/0001	0/0051	0/0067	0/0003	2	Y* T
0/0264	0/07583	0/0821	0/3609	0/613	0/0001	0/0030	0/0099	0/0009	8	R*T(Y)
0/0134	0/01452	0/0326	0/5121	0/0407	*0/0004	0/0045	0/0042	0/00067	2	Sowind date (D)
0/0431	0/05917	0/1233	0/0844	0/0060	0/00006	0/0009	0/0116	0/0003	1	Variety
0/0017	0/02040	0/0237	0/0799	0/1360	0/00004	0/0018	0/0002	0/00084	2	Y*D
0/0083	0/05512	0/02520	0/1760	0/0211	0/00009	0/0018	0/0057	0/00006	1	Y*V
0/00578	0/07725	0/08388	0/6804	*0/6445	0/00007	0/0036	0/0013	0/0015	4	T*D
0/0052	0/0049	0/00051	0/07206	0/347	0/00004	0/0026	0/0025	0/00025	2	T*V
0/0035	0/0873	0/0933	*1/234	0/1711	0/00005	0/0078	0/00038	0/0003	2	D*V
0/01806	0/0517	0/0223	0/1135	0/2311	0/0001	0/0003	0/0095	0/00049	4	Y*D*V
0/0061	0/0244	0/02417	0/09786	0/1356	0/000034	0/0004	0/0024	0/00087	2	Y*T*V
0/03073	0/15255	*0/20081	0/09014	0/1439	0/00034	0/0020	0/0074	0/0012	2	Y*D*V
0/01559	0/1844	*0/2213	0/4690	0/1204	0/0004	0/0024	0/0019	0/00033	4	Y*T*D*V
0/03073	0/15255	*0/20081	0/09014	0/1439	*0/00034	0/0020	0/0074	0/0012	4	V*D*T
0/02098	0/0739	0/0830	0/3645	0/2417	0/0001	0/0028	0/0073	0/0011	60	Error
Ns, * and ** C16:0(Palm C18:1 :(Ole C18: 2 (Lin	Ns, * and **: not significant, significant at the 5 and 1% levels of probability, respectively C16:0(Palmitic Acid) C18:0(Stearic Acid) C20:0 (Arashidic acid) TSFA :(Total saturated fatty acid) 16:1(palmitolic Acid) C18:1 :(Oleic acid) C18: 2 (Linoleic acid)									
C18: 3 (Lin	olenic acids)	TUSEA (To	tal I IN saturated f	atty acid) LIAP	·(UN known acid	k) TU/T	S (ratio Total	UN saturate	d to to	tal saturated fatty

C18: 3 (Linolenic acids) TUSFA (Total UN saturated fatty acid) UAP :(UN known acids) TU/TS :(ratio Total UN saturated to total saturated fatty acids

Table4: Means comparison of tillage systems of agronomic characters.

	•	3.	
Oil yield	Oil content	Grain yield	Tillage System
(kg/ha)	(%)	(kg/ha)	i iliuge system
b1112/7	42/54 a	2612/2 ь	(No.T)
b1105/5	41/63 b	2661/9 ь	(Min.T)
a1240/8	42/68 a	2906/8 a	(Con.T)
Manue fallance d baseling annual lat			

Mean followed by the same letters in each column are not significantly different (Duncan multiple rang test 5 %).

Table5: Means comparison of sowing date in ccombined analysis of agronomic characters

C18:1	Oil yield (kg/ha)	Oil content (%)	Grain yield (kg/ha)	sowing date			
0/642 b	1239/21 a	42/86 a	2887/41 a	8 Sep			
0/648 a	1188/94 b	41/89 в	2835/16 a	23 Sep			
0/644 ab	1030/92 c	42/09 в	2458/38 ь	7 Oct			
(confollowed by the same latters in each column are not configuratly different (Dynam multiple range test 5.0)							

Mean followed by the same letters in each column are not significantly different (Duncan multiple rang test 5 %).

Table6: Means comparison of brassica varieties of agronomic characters							
C16:0	Oil yield	Oil content	Grain yield	Variaty			
	(kg/ha)	(%)	(kg/ha)	Variety			
4/65 b	1093/64 b	41/96 b	2603/04 ь	Hyola401			
4/69 a	1212/40 a	42/61 a	2850 /92 a	PF			
Manue fullance d las de							

Mean followed by the same letters in each column are not significantly different (Duncan multiple rang test 5 %).

Table 7. Weats interaction of thage systems and sowing date on agronomic characters							
Linoleic Acid Content (18:2)	Oil Content(%)	Sowing date	Tillage System				
21/22 a	42/75 ь						
20/96 ab	42/72 ь	8 Sep	No.Till				
20/95 ab	42/13 bc						
21/09 ab	42/22 bc						
21/03 ab	40/99 d	23 Sep	Minimuim.Till				
20/76 ab	41/68 cd						
20/72 ь	43/61 a						
21/08 ab	41/98 bc	7 Oct	Conventional.Till				
21/17 ab	42/45 b						

Table7: Means interaction of tillage systems and sowing date on agronomic characters

Mean followed by the same letters in each column are not significantly different (Duncan multiple rang test 5 %).

Fable8: Means interaction between	brassica	varieties and	l sowing	date on	agronomic	characters
abico. Micans mici action between	or assica	varioues and	1 SOWING	uate on	agronomic	characters

Linolenic A (18:3)	Variety	Sowing date
11/17 ab	Hyola 401	2 San
11/25 ab	PF 7045.91	8 Sep
11/53 a	Hyola 401	22 Sam
11/05 ь	PF 7045.91	25 Sep
11/33 ab	Hyola 401	70-4
11/56 a	PF 7045.91	7 Oct

Mean followed by the same letters in each column are not significantly different (Duncan multiple rang test 5 %).

Table9: Means comparison of interaction among brassica varieties and sowing date and tillage systems on agronomic characters and content of saturated fatty acid

TUSFA	18: 1	Oil yield(kg/ha)	Grain yield(kg/ha)	Variety	Sowing date	Tillage systems
91/91 a	0/643 ab	1374/56 bc	3164/7 ь	Hyola 401	8 Sam	
91/76 ab	0/638 b	1300/19 cd	2912/9 bcd	PF	8 Sep	
91/69 ab	0/656 a	1183/74 ef	2754/8 cde	Hyola 401	22 Sam	NT
91/54 ab	0/645 ab	1107/25 f	2463/3 fg	PF	25 Sep	IN.1
91/63 ab	0/641 b	1168/88 ef	2697/7 def	Hyola 401	7 Oct	
91/70 ab	0/648 ab	1363/04 bc	2986/8 bc	PF	7001	
91/53 ab	0/652 ab	1127/59 ef	2675 def	Hyola 401	Q C	
91/74 ab	0/638 b	1294/62 cd	3012/3 b	PF	8 Sep	
91/77 ab	0/643 ab	809/95 h	2693/6 def	Hyola 401	22 Sam	МТ
91/71 ab	0/648 ab	1152/90 ef	1958/9 i	PF	25 Sep	IVI. I
91/83 ab	0/643 ab	967/02g	3148/4 ь	Hyola 401	7 Oct	
91/51 ab	0/645 ab	1412/67 b	2197/4 h	PF	7001	
91/71 ab	0/638 b	1421/56ь	3162/1 ь	Hyola 401	9 Can	
91/63 ab	0/643 ab	1694/36 a	3689/2 a	PF	8 Sep	
91/78 ab	0/645 ab	971/54 g	2306 gh	Hyola 401	22 Sam	C.T
91/45 b	0/652 ab	1085/18f	2523/7 efg	PF	25 Sep	
91/54 ab	0/647 ab	1100/76 f	2554/7 efg	Hyola 401	7 Oct	
91/75 ab	0/637 b	1234/09de	2709 def	PF	/ 001	

Mean followed by the same letters in each column are not significantly different (Duncan multiple rang test 5 %).

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