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Effect of Time of Growing Season and Time of Day for Flower Harvest on Flower Yield and Essential Oil Quality and Quantity of Four *Rosa* Cultivars

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ABSTRACT

The performance of four *Rosa* cultivars representing different species (*R. damascena* 'Gulqandi', *R. centifolia* 'Sahiwal', *R. borboniana* 'Lahori' and *R. hybrida* 'Gruss an Teplitz') was evaluated with respect to their floral characteristics under Faisalabad, Pakistan agro-climatic conditions. There was a positive and highly significant interaction between temperature and number of flowers for 'Sahiwal'. As temperature increased, so too did the number of flowers, with a maximum number of flowers/plant in March, April and May. This was significantly more than other months of the year. 'Sahiwal' formed the most flowers, followed by 'Gruss an Teplitz' then 'Gulqandi'. The effect of time of day of flower harvest (05:00 or 17:00 hr) on the quality and quantity of rose oil produced from these four roses was examined to determine the best harvest time for maximum oil yield, ideal oil content and suitable oil composition. Harvest time significantly affected the quantity and quality (i.e. oil profile and composition) of the oil recovered. Early morning was the best time to harvest flowers for maximum quality and yield of essential oil from all four roses. 'Sahiwal' showed the greatest potential for commercial production of rose essential oil by Faisalabad farmers due to an overall high flower yield over an extended flowering period and relatively high yield of high quality essential oil.

Keywords: gas chromatography, harvest time, rose, Soxhlet apparatus
Abbreviations: evening A. oil, absolute oil form flowers harvested in the evening; **evening C. oil**, concrete oil form flowers harvested in the evening; **GC**, gas chromatography; **GA**, gibberellic acid; **morning A. oil**, absolute oil from flowers harvested in the morning; **morning C. oil**, concrete oil form flowers harvested in the morning

INTRODUCTION

The genus *Rosa* contains over 130 recognized species native to diverse climatic regions (Cairns 2000). Species are native to the northern hemisphere and are widely distributed from the arctic to the tropics in Europe, Asia, the Middle East and North America (Krüssmann 1981; Nilsson 1997). The performance of any particular rose is strongly influenced by the environmental conditions and how well adapted the genotype is to those conditions.

Temperature is one of the most critical factors affecting the growth and morphological characteristics of plants (Hu and Willson 2000). The interaction of temperature and a plant's ability to adapt to temperature influences its survival, ecological niche, competitiveness, reproduction, and usefulness to humans (Boyer 1982). Temperature is a critical cue for most rose species and some cultivars to bloom as they have an obligate vernalization requirement for flowering. Roses that repeat flowering over the growing season (most modern cultivars have been selected to possess this trait) have a non-obligate vernalization requirement; however, the initial cycles of flowering right after a vernalization treatment tend to be more productive (Gudin 1992).

General growth and productivity of roses is also strongly dependent on temperature (Pasian and Lieth 1994). Optimization of rose flower production requires the ability to predict the response of plants to various environmental conditions, particularly air temperature. Computer software has been developed for the scheduling of cut roses that can be optimized based on cultivars and is based on thermal units (Mattson and Lieth 2007). Cut roses and potted miniature roses are generally cultivated over a range of 20–30°C during the day and 16–20°C at night with particular temperatures chosen based on cultivar, desired growth response and convenience (Bredmose 1998; Jiao and Grodzinski 1998; Dole and Wilkins 1999; Damake and Bhattacharjee 2000; Gonzalez-Real and Baille 2000; Shin *et al*. 2001; Kim and Lieth 2003).

Roses are of great economic importance as a crop for the florist, landscape, culinary, medicinal and perfume industries. Mostly roses are grown for ornamental purposes, however, there are some rose cultivars having fragrance in their flower petals and are used to extract EOs. *Rosa damascena* Mill. (Damask rose, oil-bearing rose, pink rose), *R. gallica* L. and *R. centifolia* L. are the most important species for producing high-value aromatic EO. This EO is used as a fragrance component in pharmaceutical preparations (e.g., ointments and lotions) and is extensively used as a fragrance ingredient in perfumes, creams and soaps (Lawrence 1991). The value of rose EO has been recognized for centuries and today there are four main centers located in Bulgaria, Turkey, Iran and France where roses are grown for the production of rosewater and rose EO. Temperature during production and harvest can also be a crucial factor affecting the quality of the EO produced in rose flowers, which are typically grown in outdoor field conditions. EOs are highly volatile and can easily be lost. Rose EO is comprised of 56 components, of which 13 are aroma-bearing compounds, e.g. citronellol, methyl eugenol, geraniol, linalool, and others (Younis *et al.* 2007). When considering the quality and quantity of EO, it is necessary to consider temperature at the time of petal harvest; the time of day is also

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a critical aspect (Charn 1979; Chalchat *et al.* 1997). In other species and plant organs grown for EO, such as leaves of mint (*Mentha*), it is well documented that the time of harvest greatly influences the EO yield, content, and composition (Clark and Menary 1984).

In a previous study to characterize rose cultivars and compare extraction methods for rose EO production in Pakistan, EO was extracted from four rose cultivars (*R. damascena* 'Gulqandi', *R. centifolia* 'Sahiwal', *R. borboniana* Desp. 'Lahori' and *R. hybrida* L. 'Gruss an Teplitz' (='Surkha')) using solvent extraction through hexane, solvent extraction through ether and steam distillation (Younis *et al.* 2007). Through solvent extraction (hexane) 'Gulqandi' yielded 0.145% absolute oil (on a petal weight basis) and 'Sahiwal' yielded 0.11%, whereas 'Gruss an Teplitz' yielded the least (0.035%) absolute oil. Solvent extraction through hexane yielded more absolute oil (0.11%) than steam distillation (0.075%) and solvent extraction (0.07%) through ether on a petal weight basis (Younis *et al.* 2007). With a greater understanding of the oil produced by these cultivars and a high yielding extraction process identified, the next step is to characterize these *Rosa* cultivars for overall flower yield and performance in a standard production environment in Pakistan.

Thus, the objectives of the present study are to a) identify the ideal rose cultivar that can be used for cultivation and flower production under local climatic conditions in Pakistan (yield, EO quality and duration of bloom production) and b) to identify the best time of day for flower harvest to obtain the highest quality EO.

MATERIALS AND METHODS

The study was carried out at the Institute of Horticultural Sciences, University of Agriculture, Faisalabad during 2004-2005. Four rose cultivars, *R. damascena* 'Gulqandi' (**Fig. 1A**), *R. centifolia* 'Sahiwal' (**Fig. 1B**), *R. borboniana* 'Lahori' (**Fig. 1C**) and *R. hybrida* 'Gruss an Teplitz' (= 'Surkha') (**Fig. 1D**) were evaluated for their floral characteristics under Faisalabad agro-climatic conditions (longitude 73° and 74° E, latitude 30° and 31.5° N, 184.4 m above sea level) (Pakistan Metrological Department, www.pakmet.com).

The rose plants were planted in the field in a Randomized Complete Block Design (RCBD). There were 30 blocks containing one plant of each cultivar randomized within each block. The four plants per block were planted in a square with a plant-to-plant distance of 91.4 cm. For this study, data were collected on the following floral characteristics: individual flower fresh and dry weight (g), flower diameter (cm) and flower yield (g/plant/month). Data were collected on two-year-old plants.

Fig. 1 Four rose cultivars used in this study. *R. damascena* 'Gulqandi' (**A**), *R. centifolia* 'Sahiwal' (**B**), *R. borboniana* 'Lahori' (**C**) and *R. hybrida* 'Gruss an Teplitz' (= 'Surkha') (**D**).

Collection and preparation of flowers for essential oil extraction

Flowers of all four rose cultivars were collected at the half-open stage in the morning (05:00 hr) and evening (17:00 hr). Flowers were bulked over multiple plants and when desirable quantity (~7 days to get the desirable quantity of petals) for one replication (i.e. 20 kg) was obtained. Petals were separated, weighed and spread in a tray out of sunlight at room temperature in the laboratory for 24 h before oil extraction in order to remove extra moisture, but not in order to dry them. Twenty kilograms of petals from each rose cultivar were used for each harvest time for extraction and it was replicated thrice over time during the month of April.

Solvent extraction

For solvent extraction, a Soxhlet apparatus was used. *n*-Hexane was used as the solvent to extract the EO from the four rose cultivars. Solvent extracts of all the volatile compounds from the petals were collected in a flask (2L, Kinax, USA). This was the concrete oil, which was further processed to remove any remaining solvent from the rose EO. A distillation process was performed to recover the solvent from the concrete oil (organic solvent and rose oil) using a rotary evaporator. In this way all the organic solvent was recovered. Distillation by using a rotary evaporator is useful because the active ingredients of rose EO are not lost. Absolute oil from concrete oil was recovered by adding 2 ml of absolute alcohol to 20 ml of concrete oil. The alcohol removes all the natural waxes present in the EO. The EO was filtered and the absolute alcohol was removed by performing distillation with a rotary evaporator. Final traces of alcohol were removed by bubbling nitrogen gas through this EO.

Identification by gas chromatography

We also carried out gas chromatography (GC) for separation and qualitative and quantitative analysis of rose EO constituents. A Shimadzu 17-A gas chromatograph was used (Sp 2330 capillary columns, BPX70 – 70% phenyl column; column length 30 m; column thickness 25 µm). Other analytical parameters were: sample size (1 µl); carrier gas, N_2 (g) flow velocity = 5 ml/min; initial column temperature, 50°C; and final column temperature, 120°C. Initial hold was three min, final hold up time was 9 min and ramp rate was 5°C/min. Identification of constituents of EO of all four rose cultivars was done by comparison with gas chromatograms of a mixture of standards (Fluka, 99% GC grade, Switzerland) while quantitative analysis was carried out by calculating the area under the peak using software CSW-32 (http://www.dataapex.com/products/csw32.php).

Statistical analysis

Data were recorded and analyzed for significance using analysis of variance (ANOVA) and means were compared using Duncan's multiple range test at the 5% probability level (Steel *et al.* 1997). Minitab Statistical Software (Version 2.4) was used for analysis of data. Pearson's correlation was use to correlate monthly mean temperature and monthly means for the different floral characteristics.

RESULTS AND DISCUSSION

Flower production

Flower yield differed significantly ($P \leq 0.01$) for month of harvest, cultivar, and the month × cultivar interaction (**Table 1**). Production number of flowers for each cultivar per month is reported in **Table 2**. The maximum flower yield (121.7 flowers/month/plant) was produced in May by 'Sahiwal', followed by 'Gruss an Teplitz' (111.7 flowers/month/plant) in April. Both 'Gulqandi' and 'Lahori' yielded a maximum (77.7 and 57.9, respectively) number of flowers in April, significantly less then the other two cultivars during that month.

A relationship between temperature and flower production of four rose cultivars under Faisalabad climatic condi-

Table 1 Analysis of variance for flower yield/plant/month of four rose cultivars.

Source of variation	D.F.	S.S.	M.S.	F Value
Months	11	69542.92	6322.08	9534.82**
Cultivar	3	140568.97	46856.32	70667.62**
Month x Cultivar	33	24795.53	751.38	1133.21**
Error	96	63.65	0.66	
Total	143	234971.08		

* = Significant ($P \leq 0.05$), ** = Highly significant ($P < 0.01$)

tions was identified. There was a weak and negative association between temperature and individual flower fresh weight in all four rose cultivars. A weak and negative relationship between temperature and individual flower dry weight occurred in 'Gruss an Teplitz', 'Gulqandi' and 'Sahiwal', but in 'Lahori' it had a positive but non-significant association (**Table 3**). A positive and highly significant interaction between temperature and the number of flowers was observed for 'Sahiwal' (**Table 4**). As temperature increased there was also an increase in the number of flowers and a slight decrease in flower size for 'Sahiwal'. Temperature was negatively associated with all or most flower production traits in 'Gulqandi' (**Table 5**) and 'Gruss an Teplitz' (**Table 6**), respectively.

In 'Sahiwal' an increase in the number of flowers per plant was obtained with increasing temperature, which is a unique character in roses. Flower production of 'Sahiwal' was maximum in the hottest months of the year, i.e. May, June and July (**Table 2**). Several researchers (Mortensen and Moe 1995; Yamaguchi and Hirata 1998) reported a trend that as temperature increased to 30°C there was a decrease in flower production and quality in roses, a complete contrast to our findings for 'Sahiwal'. Even though May, June and July are the hottest months of the year, flower production was maximum in these months, highlighting that 'Sahiwal' is a well adapted cultivar in this production environment. 'Gruss an Teplitz' ranked second in terms of flower production (**Table 2**), whereas 'Gulqandi' produced flowers only in March and April, and as the temperature increased, flower production ceased (**Table 2**). A similar trend was observed in 'Lahori' (**Table 2**). Fern (2000) found, during his field trips to England, that 'Gulqandi' flowers from June to July; in our study it produced flowers for 7 to 8 weeks (March-April).

Most wild roses are non-recurrent bloomers and have a short flowering season of 2-4 weeks in spring. This flowering pattern is due to an obligate vernalization requirement for flowering and flower bud initiation occurring over a limited time period just after bud break. From flowering patterns in Faisalabad 'Lahori' and 'Gulqandi' appear to be non-recurrent cultivars (**Table 2**). Most modern cultivars flower continuously or recurrently, producing flowers throughout the growing season outside and throughout the year under glass. 'Sahiwal' and 'Gruss an Teplitz' fall into

Table 2 Mean number of flowers/plant/month of four rose cultivars throughout the year.

Month	Cultivar				Sahiwal
	Mean temperature (°C)	Gruss an Teplitz	Gulqandi	Lahori	
Jan	19.4	14.2 ± 0.42 s	0.0 ± 0.00 t	0.0 ± 0.00 t	60.6 ± 1.25 j
Feb	21.9	39.3 ± 0.47 op	0.0 ± 0.00 t	0.0 ± 0.00 t	77.7 ± 0.80 g
Mar	26.7	92.1 ± 0.52 f	57.1 ± 0.51 k	41.2 ± 0.45 n	99.3 ± 0.38 d
April	33.5	111.7 ± 0.37 b	77.7 ± 0.89 g	57.9 ± 0.58 k	99.0 ± 0.58 d
May	38.4	97.4 ± 0.40 e	0.0 ± 0.00 t	44.4 ± 0.31 m	121.7 ± 0.38 a
June	42.5	68.1 ± 0.48 h	0.0 ± 0.00 t	0.0 ± 0.00 t	106.1 ± 0.49 c
July	39.1	53.2 ± 0.44 l	0.0 ± 0.00 t	0.0 ± 0.00 t	100.2 ± 0.72 d
Aug	37.1	57.7 ± 0.90 k	0.0 ± 0.00 t	0.0 ± 0.00 t	107.0 ± 0.58 c
Sept	35.7	38.1 ± 0.59 p	0.0 ± 0.00 t	0.0 ± 0.00 t	92.0 ± 0.54 f
Oct	33.0	35.9 ± 0.52 q	0.0 ± 0.00 t	0.0 ± 0.00 t	64.7 ± 0.88 i
Nov	27.2	21.1 ± 0.48 r	0.0 ± 0.00 t	0.0 ± 0.00 t	61.8 ± 0.43 j
Dec	21.4	15.4 ± 0.32 s	0.0 ± 0.00 t	0.0 ± 0.00 t	40.5 ± 0.78 no
Total		644.2	134.8	143.5	1030.6

Mean in a column sharing the same letter do not differ significantly using DMRT ($P<0.05$)

Table 3 Correlation between temperature and flower production for *Rosa borboniana* 'Lahori'.

	Temp.	Fresh flower wt.	Dry flower wt.	№ of flowers	Size of flowers
Fresh flower wt.	0.115				
Dry flower wt.	0.113	1.000**			
No. of flowers	0.139	0.985**	0.985**		
Size of flowers	0.119	1.000**	1.000**	0.985**	
Flower yield/plant/month	0.135	0.984**	0.985**	1.000**	0.984**

* = Significant (P<0.05), ** = Highly significant (p<0.01)

Table 4 Correlation between temperature and flower production for *Rosa centifolia* 'Sahiwal'.

	Temp.	Fresh flower wt.	Dry flower wt.	№ of flowers	Size of flowers
Fresh flower wt.	-0.458				
Dry flower wt.	-0.367	0.884**			
No. of flowers	0.767**	-0.809	-0.665		
Size of flowers	-0.640	0.507	0.656	-0.487	
Flower yield/plant/month	0.781**	-0.744	-0.603	0.994**	-0.461

* = Significant (P<0.05), ** = Highly significant (P<0.01)

Table 5 Correlation between temperature and flower production for *Rosa damascena* 'Gulqandi'.

	Temp.	Fresh flower wt.	Dry flower wt.	№ of flowers	Size of flowers
Fresh flower wt.	-0.075				
Dry flower wt.	-0.076	1.000**			
No. of flowers	-0.039	0.983**	0.982**		
Size of flowers	-0.074	1.000**	1.000**	0.984**	
Flower yield/plant/month	-0.040	0.984**	0.983**	1.00**	0.984**

* = Significant (P<0.05), ** = Highly significant (P<0.01)

Table 6 Correlation between temperature and flower production for *Rosa hybrida* 'Gruss an Teplitz'.

	Temp.	Fresh flower wt.	Dry flower wt.	№ of flowers	Size of flowers
Fresh flower wt.	-0.915				
Dry flower wt.	-0.852	0.848**			
No. of flowers	0.553	-0.287	-0.588		
Size of flowers	-0.945	0.861**	0.785**	-0.499	
Flower yield/plant/month	0.526	-0.258	-0.570	0.999**	-0.476

* = Significant (P<0.05), ** = Highly significant (P<0.017)

this category (**Table 2**). Flowering of recurrent roses is described as self-inductive because there is no evidence that floral initiation is environmentally regulated (Halevy 1972). The recurrent-flowering characteristic is determined by a single locus and a recessive mutant allele in two key sources of this trait of modern cultivars: *R. chinensis* Jacquin and *R. wichurana* Crépin (Hurst 1941; Semeniuk 1971).

Gibberellic acid (GA) appears to play a strong role in flower induction in roses and concentration appears to be the key difference between recurrent and non-recurrent roses. In spring, roses produce shoots from axillary buds on overwintered stems from the previous season. The terminal meristem of each axillary shoot initiates a number of leaf primordia that is characteristic of a particular genotype and then the shoot terminates in an inflorescence: further growth involves the development of axillary shoots on these stems (Horridge and Cockshull 1974; Cockshull and Horridge 1977). From the data of Cockshull and Horridge (1977) it may be concluded that floral induction in *R. damascena*, a non-recurrent species, occurs shortly after bud break in spring when concentrations of GAs are low and is inhibited by subsequently higher levels of GAs. Consistent with this, it was shown that leaves of non-flowering shoots of *R. damascena* contained higher concentrations of GA and GA-like substances than flowering shoots (Farooqi *et al.* 1994). Roberts *et al.* (1999) report that in the non-recurrent cultivar 'Félicité et Perpétue', floral initiation occurs when concentrations of GAs are low just after bud break and GA concentrations quickly rise to levels that inhibit floral initiation. In contrast, in 'Little White Pet', a recurrent sport of 'Félicité et Perpétue', concentrations of GAs remain below a critical threshold for flowering throughout the growing season. Furthermore, applications of GA_3 reduced the number of flowers per plant and the proportion of flowering plants when applied at bud break to non-recurrent roses.

'Sahiwal' performed extremely well under Faisalabad climatic conditions and had remarkable flowering throughout the year (Younis *et al.* 2006); a single plant could produce about 1,000 fragrant flowers/year (**Table 2**). This character of 'Sahiwal' distinguished it from all other rose cultivars, most of which did not produce flowers throughout the year and which, with an increase in temperature, resulted in a decrease in flower production. 'Sahiwal' was identified among a shipment of what was labeled as 'Des Peintres'. It was recognized as different from 'Des Peintres' and named 'Sahiwal' to distinguish it. 'Sahiwal' is unique among *R. centifolia* cultivars because *R. centifolia* is not known to be recurrent (Krüssman 1981). On the other hand, *R. borboniana* cultivars are known to be recurrent (Krüssman 1981) and 'Lahori' was not recurrent in Faisalabad. Morphological characteristics of these cultivars are consistent with their respective species and an inadvertent mix up of these cultivars is unlikely.

From the results it was observed that there was a weak and negative association between temperature, fresh flower weight and dry flower weight of all four rose cultivars, which is consistent with other reports. For instance, Shin *et al.* (2001) studied the response of *R. hybrida* 'Kardinal' to temperature and reported that flower dry weight increased from 0.7 to 3.0 g as temperature decreased from 30 to 15°C. They observed that when plants were moved to a lower temperature at the visible bud stage, flower dry weight increased. Besides temperature, variations in climate, soil conditions, and cultural practices all affect the growth and performance of plants. Therefore, it is unclear to what extent the increased yield (number and biomass of flowers) by 'Sahiwal' as the season progressed and temperatures increased can be attributed directly to temperature or adaptability to other local macro- and micro-environmental conditions in Faisalabad.

Effect of flower harvesting time on essential oil extraction

The second objective of this experiment was to assess the effect of harvest time of rose petals on oil quality and quantity (using 20 kg of petals per harvest time per cultivar). **Table 7** indicates considerable differences when the petals were harvested at different times of the day. 'Gulqandi' yielded 39 g of concrete oil (and 29 g of absolute oil) when petals were harvested in the morning, but when petals were harvested in the evening they yielded 25 g of concrete oil (and 16 g of absolute oil). Considerable differences in oil yield were observed among the four rose cultivars between the two harvest times. In 'Sahiwal', 30 g of concrete oil and 22 g of absolute oil were recovered when petals were harvested in the morning while 18 g of concrete oil and 11 g of absolute oil was obtained when petals were harvested in the

Table 7 Relative percentage of components of rose essential oil obtained from flowers harvested in the morning (05:00 hr) and evening (17:00 hr) identified through gas chromatography.

Volatile component	Cultivar							
	Gruss an Teplitz		Gulqandi		Lahori		Sahiwal	
	Morning	Evening	Morning	Evening	Morning	Evening	Morning	Evening
Citronellol	14.57 ± 0.93	13.12 ± 0.54	60.63 ± 0.09	48.58 ± 0.97	26.93 ± 1.64	21.56 ± 1.31	55.61 ± 1.69	48.54 ± 2.44
Methyl eugenol	3.49 ± 0.14	2.67 ± 0.11	5.01 ± 0.25	2.05 ± 0.10	1.7 ± 0.93	1.08 ± 0.03	2.93 ± 0.12	1.98 ± 0.06
Geraniol	2.99 ± 0.18	2.78 ± 1.12	2.05 ± 0.04	1.43 ± 0.09	1.89 ± 0.19	1.01 ± 0.03	2.79 ± 1.01	2.41 ± 0.05
Geranyl acetate	15.71 ± 0.65	14.89 ± 0.74	2.32 ± 0.07	1.89 ± 0.11	4.29 ± 0.26	2.35 ± 0.09	2.63 ± 0.08	1.79 ± 0.03
Phenyl ethyl alcohol	48.11 ± 0.55	42.01 ± 2.70	18.45 ± 0.56	14.01 ± 0.64	30.13 ± 1.25	27.01 ± 1.73	31.79 ± 0.97	30.01 ± 0.60
Linalool	1.59 ± 0.04	1.34 ± 0.04	1.24 ± 0.06	1.11 ± 0.01	1.24 ± 0.03	1.11 ± 0.01	1.78 ± 0.02	1.43 ± 0.01
Nerol	1.37 ± 0.04	1.23 ± 0.06	0.43 ± 0.01	0.41 ± 0.00	0.48 ± 0.09	0.34 ± 0.01	0.39 ± 0.07	0.28 ± 0.01
Benzaldehyde	2.14 ± 0.08	2.01 ± 0.02	0.13 ± .01	0.05 ± 0.00	1.03 ± 0.03	1.02 ± 0.03	1.06 ± 0.01	1.04 ± 0.03
Benzyl alcohol	3.28 ± 0.32	2.87 ± 0.09	1.75 ± 0.08	1.21 ± 0.02	5.29 ± 0.85	5.01 ± 0.17	1.08 ± 0.23	0.91 ± 0.01
Rhodinyl acetate	3.01 ± 0.12	2.93 ± 0.15	2.94 ± 0.08	1.65 ± 0.12	5.90 ± 1.81	5.78 ± 0.39	2.05 ± 0.13	1.67 ± 0.09
Citronellyl acetate	1.78 ± 0.05	1.23 ± 0.07	1.89 ± 0.07	1.21 ± 0.02	2.21 ± 0.10	1.99 ± 0.03	0.98 ± .01	0.79 ± 0.10
Benzyl acetate	0.56 ± 0.03	0.34 ± 0.01	1.04 ± 0.03	0.56 ± 0.03	1.10 ± 0.26	1.01 ± 0.03	0.74 ± 0.05	0.70 ± 0.16
Phenyl ethyl formate	3.01 ± 0.06	2.89 ± 0.06	0.88 ± 0.01	0.86 ± 0.01	2.13 ± 0.24	1.98 ± 0.02	0.97 ± 0.01	0.81 ± 0.18

Values are the mean of three replications.

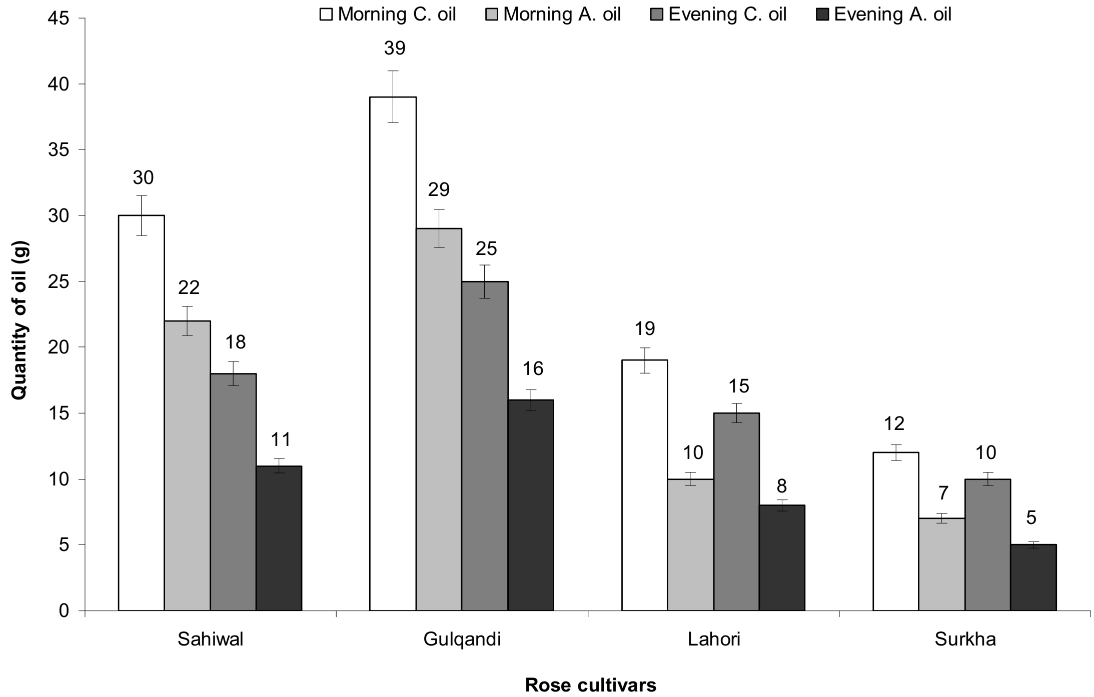

Fig. 2 Effect of flower harvesting time on *Rosa* essential oil extraction. Each value is the mean of 3 replications. Morning C. Oil = Concrete oil form flowers harvested in the morning; Morning A. oil = Absolute oil from flowers harvested in the morning; Evening C. Oil = Concrete oil form flowers harvested in the evening; Evening A. oil = Absolute oil form flowers harvested in the evening.

evening.

GC analysis showed that the concentration of citronellol, methyl eugenol and geraniol components were higher in the oil obtained from flowers harvested in the morning compared to those harvested in the evening (**Table 7**). Rose EO obtained in the morning is of superior quality because of its higher percentage of citronellol, methyl eugenol and geraniol, components responsible for the characteristic fragrance in rose.

The amount and composition of EO depends on the developmental stage (ontogeny) of the plant and flower, and therefore time of harvest is one of the most important factors influencing EO quality and quantity (Croteau and Gershenon 1994; Ulseth 1994; Kothari and Singh 1995; Ram and Kumar 1999). The objective of studying this parameter was to find out the effect of harvesting time of rose petals on the EO yield. The time of petal harvesting considerably affected the EO yield. Petals yielded more EO when they were harvested in the early morning from half-opened flowers of those 2-year old plants (**Fig. 2**). The quantitative composition of the EOs of many aromatic plants is greatly influenced by the genotype and agronomic conditions, such as harvesting time, plant age and crop density (Marotti *et al.* 1994), although, in our experience, in most roses, flower production remains similar for up to 10-12 years.

For some crops, like *Eucalyptus*, harvesting time is not critical but for roses, it is important to harvest petals during the cooler period of the day to get high yielding, top quality oil (Knox 1998). Environmental factors such as temperature, relative humidity, irradiance, photoperiod and cultivation practices can influence the composition of EOs from aromatic plants (Bruneton 1995).

The potential for post-harvest increases in extracts and volatiles also changes with harvesting time (Jamoussi *et al.* 2004). For instance, the maximum concentration of total volatiles at harvest occurred when *Boronia* flowers were harvested in the morning, and subsequently declined (MacTavish 1995; MacTavish and Menary 1997). Kitchounow (1937) reported that *R. centifolia* and *R. gallica* flowers picked after 10 am yielded 59% less EO than those that picked during early morning hours (5 am). The results of this study suggest that variations in EO components in roses is influenced by harvesting time of petals and confirms that the best time to harvest rose petals, for both the EO content and its components, is the morning (Cherchi *et al.* 2001). Much of the relevant published work has focused on the potential role of harvesting time of flowers in the generation of volatiles during post-harvest processing (Francis and Allcock 1969; Ackermann *et al.* 1989; Winterhalter *et al.* 1990, 1991; Watanabe *et al.* 1993, 1995). Le Grice (1976) reported that climatic conditions greatly affect the production and dissemination of essential oil, so that all assessments can only be comparative. Thus, the present findings that early morning (5:00 hr) is the best time for harvest petals to obtain a higher quality yield of EO are substantially supported by studies carried out on various aromatic plants.

CONCLUSION

Faisalabad climatic conditions favored the cultivation of 'Sahiwal' as it produced a greater number of overall flowers, especially so during the hotter months of the year. This character of 'Sahiwal' made it distinct from the other rose cultivars examined. Most of the roses did not flower throughout the year, and as temperature increased they exhibited a decrease in flower production. For extraction of EO from roses, harvest of rose flowers in Faisalabad should be done in the early morning in order to achieve higher yield and higher quality EO. From a commercial perspective, we recommend that farmers consider growing 'Sahiwal' to increase their income. The extended flowering season and overall greater yearly yields of 'Sahiwal' compared to traditionally grown *R. damascena* is beneficial as overall higher EO yields per unit of land can be achieved and workers have more consistent employment.

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